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Preface

This textbook is the first edition of *Non-Hodgkin's Lymphoma in Childhood and Adolescence* and represents the first book completely focused on this topic. Recent extraordinary advances in diagnostic and therapeutic modalities, triggered by exceptional progress in molecular biology, have revolutionized the world of pediatric and adolescent Non-Hodgkin lymphoma (NHL). One of the highest priorities of this book is to incorporate all the cellular and molecular advances together with the most cutting-edge therapies available today into a comprehensive volume, providing a state-of-the-art overview on NHL in children and adolescents. It is divided into seven sections, each of which focuses on a critical component of pediatric NHL, including history and epidemiology, pathology and molecular biology, pathogenesis of B- and T-cell lymphomas, as well as the most recent insights into the pathogenesis of anaplastic large-cell lymphoma, disease evaluation and response, common and rare subtypes of NHL, and current and novel treatment strategies including a chapter on hematopoietic stem cell transplant. The text also comprehensively reviews the late effects of treatment, quality of patient life, and NHL treatment in countries with limited resources.

All chapters were written by distinguished world experts in each field, and we would like to thank all of them for their efforts and hard work, extending our thanks also to several junior physicians and fellows who assisted these experts on some chapters. In addition, we are extremely grateful for the editorial assistance of Andy Kwan in New York and Prakash Marudhu in India who have demonstrated outstanding patience and dedication in managing the flow of many chapters, figures, and permissions.

We are hopeful that this book will serve as a comprehensive and updated resource for all pediatric oncologists, pathologists, biologists, and trainees who are looking after children and adolescents with NHL.

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Part I

Introduction

1 History of Diagnoses and Treatment Strategies in Pediatric Non-Hodgkin's Lymphomas

Georg Mann

Introduction

In comparison to the extended and detailed spectrum of non-Hodgkin's lymphoma (NHL) entities in adulthood, there is only a limited variety of defined diagnoses in pediatric oncology, with 90% of the diseases being assigned to only 5 diagnoses, starting with Burkitt's lymphoma (BL), followed by lymphoblastic lymphoma (LL; 3/4 of T-cell origin), anaplastic large cell lymphoma (ALCL), diffuse large B-cell lymphoma (DLBCL), and primary mediastinal B-cell lymphoma (PMBCL) [16, 55]. Rare pediatric entities with only 1–2% are composed of pediatric follicular lymphoma (FL), mucosa-associated lymphoid tissue (MALT) lymphoma, marginal zone lymphoma (MZL), subcutaneous panniculitis-like T-cell lymphoma (SPTCL), and others.

The History of Cancer from Old Egypt to the Nineteenth Century

To understand the difficulties that medical research had to overcome on the way to clinically useful classification systems, especially with respect to pediatric NHLs, a brief look back at the evolution of medicine concerning cancer diagnosis might prove helpful. Possibly the first mention of cancer was found in an old Egyptian papyrus in which *Imhotep* described what presumably was a breast tumor in 2625 BC. The term Karkinos was first used in ancient Greece by *Hippocrates* around 400 BC. *Galen* in the second century AD believed that the genesis of diseases mainly to be caused by the dysregulation of body fluids or "humors" which he associated with certain personality traits or moods. Thus, the melancholic state, which he considered to be the worst, was thought to cause tumors by an excess of black bile. As the black bile was thought to reappear after surgical removal, forming cancer again, surgery was not recommended. This inadvertently saved patients with tumors from procedures which were at that time often incomplete and followed by what we now know are infections, worsening their conditions and shortening their lives. It was only in the Renaissance that autopsies set the foundations for pathologic anatomy. When *Vesalius,* in the middle of the sixteenth century, tried, but failed, to find the black bile by autopsy, he started a process of scientific discoveries that led to the development of anatomic pathology and its correlation to the causes of diseases [60].

The Early History of Lymphomas: Big Steps Forward Ending in Confusion

In 1832, *Thomas Hodgkin* was the first to describe what we now call lymphoma by correlating the clinical course of a disease of the glands and the spleen to its macro-pathologic appearance in six patients by autopsy. Interestingly, although microscopy was invented nearly a century earlier, it did not play a role at that time; a few decades later, however, microscopic examinations revealed the uniform appearance of the original tissue conserved by Hodgkin, thus confirming the diagnosis. *Rudolf Virchow* at the Charité in Berlin, by means of what he called cellular pathology, was the first to use the terms "leukemia", "lymphoma", and "lymphosarcoma" describing clinicopathologic entities in 1862 [2, 50, 89].

The term "lymphosarcomatosis" to emphasize the systemic character of the disease and to distinguish it from Hodgkin's disease was coined by *Johann Kundrat* in Vienna [43], who used staining techniques developed by Paul

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Ehrlich at the end of the nineteenth century in Berlin. *Ehrlich* was awarded the Nobel Prize in 1908 for his contributions to the understanding of the immune system. The enormous medical progress starting in the middle of the nineteenth century, characterizing the immune system and its pathologies, led to various attempts to classify "malignant lymphomas", a term introduced by the surgeon *Theodor Billroth* in Vienna [12].

During the following decades, dependent on the development and availability of diagnostic techniques and the knowledge of clinical courses, several different classification systems, some of which ending up with a broad variety of diagnoses, evolved sometimes even simultaneously. The systems defined cytomorphology with respect to size and shape of the cells, their cytoplasm and nuclei, the distribution within the lymph node, the clinical behavior, and the estimated origin of the cells. Not having enough evidence to reveal the genesis of the lymphoma cells, apparently unripe large cells were thought to stem from histiocytic/reticulocytic origin – another misunderstanding that worsened the Babylonian confusion and made clinicopathologic series incomparable when different systems, or even the same, were used by different groups, explainable by the lack of understanding of the cellular physiology of the immune system and the limitations of cytomorphological interpretation and characterization [50, 79].

On the Way to a Valuable Classification of Non-Hodgkin's Lymphomas (Table 1.1)

In 1966, *H. Rappaport* in the USA created a classification system by taking into account lymph node architecture (nodular vs. diffuse), postulated cellular origin (lymphocytic vs. histiocytic), and cytomorphology (well differentiated vs. poorly differentiated) [50, 74]. The detection of immunoglobulins on the surface of lymphoma cells by A.C. *Aisenberg* was another important step toward a biologic characterization of NHL [3]. A better understanding of the immune system led *R. J. Lukes* and *R.D. Collins* to develop a classification which was taking into account the B- or T-cell origin of lymphomas and which became widely accepted in the U.S. [48]. At around the same time, *Karl Lennert* in Germany created a classification system which, in addition to histological, immunohistochemical, cytologic, and ultrastructural techniques, also took into account immunochemical and immunomorphological methods in order to define the T- or B-cell origin of the tumor cells [46]. The result was the so-called Kiel classification 1974, which was then broadly used by lymphoma experts in Europe.

Table 1.1 Evolution of NHL classification systems – most relevant pediatric diagnoses: High grade or "intermediate grade"(DLBCL)

Rappaport, 1956 [74] **(1966,)** Nodular or diffuse:		**Working formulation 1982**
Well-differentiated lymphocytic lymphoma		Large cell immunoblastic
Poorly differentiated lymphocytic lymphoma		Lymphoblastic
Histiocytic/mixed lymphoma		Small non-cleaved cell (Burkitt and non-Burkitt type)
Burkitt		Diffuse large cell
	Lukes and Collins [48]	
B		**T**
Large non-cleaved FCC		Convoluted lymphocytic
Small non-cleaved FCC		Undefined
Burkitt type		Unclassifyable
Immunoblastic		Immunoblastic
	Kiel 1975 [46]	
B		**T**
Centroblastic		Pleomorphic, medium and large cell
Immunoblastic		Immunoblastic
Large cell anaplastic (Ki-1+)		Large cell anaplastic (Ki-1+)
Burkitt's lymphoma		
Lymphoblastic		Lymphoblastic
	REAL 1994 [36]	
B		**T**
Precursor B-cell neoplasm:		Precursor T-cell neoplasm: T-cell and natural killer cell neoplasms
Precursor B-lymphoblastic leukemia/lymphoma		Precursor T-lymphoblastic lymphoma/leukemia
Mature (peripheral) B-cell neoplasms		Mature (peripheral) T-cell neoplasms
Diffuse large cell B-cell lymphoma		Anaplastic large cell lymphoma, T/null cell, primary cutaneous type
Mediastinal large B-cell lymphoma		Peripheral T-cell lymphoma, not otherwise characterized
Burkitt's lymphoma/ Burkitt's cell leukemia		Anaplastic large cell lymphoma, T/null cell, primary systemic type
	WHO 2008, rev. 2016 [81, 82]	
B		**T**
Precursor B-cell neoplasm		Precursor T-cell neoplasm

Table 1.1 (continued)

Precursor B-lymphoblastic leukemia/lymphoma		Precursor T-lymphoblastic lymphoma/leukemia
Precursor B-cell acute lymphoblastic leukemia		Precursor T-cell acute lymphoblastic leukemia
Mature (peripheral) B-cell neoplasms		Mature (peripheral) T-cell neoplasms
Diffuse large B-cell lymphoma		Anaplastic large-cell lymphoma, T-/null cell, primary cutaneous type
Mediastinal large B-cell lymphoma		Peripheral T-cell lymphoma, not otherwise characterized
Burkitt lymphoma/ Burkitt cell leukemia		Anaplastic large-cell lymphoma, T-/null cell, primary systemic type
Pediatric follicular lymphoma (2016)		EBV+ T-cell lymphoma of childhood (2016)

The Availability of Immunophenotyping

Georges Köhler and *Cesar Milstein* fused distinct antigen-specific B cells to myeloma cells, thus creating immortal clonal B-cell lines which were able to produce a specific monoclonal antibody to any desired antigen and in any desired amount [41]. This technique enabled researchers to unequivocally define lymphoma cells as either being of B- or T-cell origin. Additionally, it was attempted to correlate the immunophenotype of NHLs to the physiologic maturation stage of lymphatic development with respect to the protein expressions occurring at different stages [4]. To unify the language and specificity of monoclonal antibodies developed by different groups, the international IUIS-WHO Nomenclature Subcommittee was founded and launched the cluster denomination (CD) system starting with the definition of 15 entities [37].

Detection of Clonal Immunoglobulin and T-Cell Receptor Gene Rearrangements

A prerequisite for a valuable classification system appeared to be the understanding of the normal cellular immune system [49] and the description of the malignancies as counterparts of different physiologic cell types and development stages of their precursors. An important contribution to the biology and pathology of the immune system was the detection of the hierarchical organization of clonal immunoglobulin and T-cell receptor gene rearrangements [42, 56, 90]. The understanding of the physiology helped to support the definition of biologic entities and formed a basic tool by using the clonal rearrangement detection for minimal residual and disseminated disease measurement. Interestingly, these fundamental developments were not reflected in the so-called working formulation [64], whose intention was to create a translation among the various systems, to facilitate clinical comparisons of case reports and therapeutic trials and to unify and simplify the language in order to have a common NHL classification, mainly based on cellular appearance and clinical behavior.

In 1994, the Revised European-American Lymphoma (REAL) classification took into account immunophenotypic features in identifying distinct clinicopathologic entities as being either of B- or T-cell origin, by following and unifying the European and American systems on the currently available diagnostic tools. Of pediatric relevance is the definition of diffuse large B-cell lymphomas (DLBCL) and primary mediastinal B-cell lymphomas (PMBCL), which in adolescents account for a relevant proportion [36]. In 2000, by means of distinguishable gene expression analysis, two subtypes – germinal center B-cell-like (GCB) and activated B-cell-like (ABC) DLBCL – were defined [5]. The GCB type is more common in pediatric DLBCLs [83]. Further modifications by the WHO 2008 and 2016 classification ([81], revised 2016 [82]) helped to better characterize low grade and cutaneous entities, very rare in the pediatric age group, overall describing close to 50 entities with 18 subentities whereof only one fifth occurs in the pediatric age cohort in a quantitatively relevant number. What was most relevant in the revised version of 2016 for the diagnosis of pediatric NHL, was the introduction of, first, *systemic EBV+ T-cell lymphoma of childhood* (the name was changed from lymphoproliferative disorder to lymphoma, due to its fulminant clinical course and a desire to clearly distinguish it from chronic active EBV infection) and, second, *pediatric-type follicular lymphoma* (a localized clonal proliferation with excellent prognosis where a conservative therapeutic approach may be sufficient [9].

The Historical Characterization of Burkitt's and Anaplastic Large Cell Lymphomas

These two entities deserve closer description as they shed light on the development of medical research and clinical progress in oncology, unlike lymphoblastic, mainly T-cell, NHLs, where the evolution of characterization parallels the diagnostic development of acute lymphoblastic leukemia of childhood.

Burkitt's Lymphoma

The history of the diagnosis of Burkitt's lymphoma reads like an orchestration influencing the progress of medicine. Disciplines like epidemiology, histology, tropical medicine,

virology, cytogenetics, and molecular genetics were all components helping to establish a particular diagnosis while adding new knowledge, thus fostering general medical research. The Irish surgeon *Denis Parson Burkitt* worked in Africa on behalf of the British colonial Ministry when he encountered jaw tumors in small children in the late 1950ties [19]. The uncommon manifestations with localizations not typical of any then well-known malignancies drove him to thoroughly investigate the courses of the patients and the possible epidemiology. He found that these tumors had been described already earlier, by *Dr. Albert Cook* who travelled to Africa at the end of the nineteenth century. At that time these tumors were diagnosed as organ-specific sarcomas, depending on their localization as either Wilm's tumors, neuroblastomas, or rhabdomyosarcomas. But the spread to Burkitt seemed to be atypical, since several tumors of similar size appeared in the same child without forming a primary and metastases. Recognizing the nature of the disease as systemic, he called it a syndrome [20]. Later, with the help of the pathologist *O'Connor*, the uniform microscopic appearance together with the clinical appearance confirmed the lymphomatous origin**.** Besides the finding that the disease was geographically confined to the peri-equatorial belt, the next epidemiologically important issue was the fact that these tumors only occurred up to certain altitudes above sea level. This suggested temperature as a possible environmental influence. Additionally, the thereby created map of lymphomas distribution resembled the spread of malaria. When tumor material was sent to England, an agent was detected within the cells of all samples [26], the Epstein-Barr virus (EBV), now bearing the name of its discoverers [13]. Identical appearing lymphomas in children outside Africa, which were then called non-endemic Burkitt's lymphomas, harbored the virus only in about half of the cases. Together with the later detected universal presence of the virus, doubts were raised about its causative role in cancer. However, some time later, the concept of virally induced cancer proved to be of importance after all. Iatrogenic immunosuppression necessary after organ transplantation poses patients at risk for chronic EBV proliferation in B-lymphatic cells which stepwise causes lymphoproliferation named posttransplant lymphoproliferative disease (PTLD) [28, 73], from oligo clonal populations up to monoclonal lymphomas. The solution to Burkitt's observation in Africa identifies chronic malaria as the cause of chronic immunosuppression. The spread by anopheles mosquitoes around the equatorial belt to altitudes of about 1000 meters above sea level, up to which Anopheles reproduces, explains the epidemic findings. The failure to control EBV leading to the development of lymphomas resembles the way PTLDs evolve. Similarly, EBV-positive B-NHLs may develop in immunodeficient children, like in the X-linked lymphoproliferative syndrome, the most striking example [35]. Another important step was the cytogenetic characterization of Burkitt's lymphoma. The chromosomal translocation t(8;14)) (q24;q32) was one of the first reproducible genetic alterations in human cancer [93], not only enabling the molecular characterization of the lymphoma but also stimulating the research toward the genetic causes of cancer. This resulted, among other things, in the characterization and functional description of the fused genes C-MYC on chromosome 8 and IGH on chromosome 14 [23, 52, 84].

Anaplastic Large Cell Lymphoma

Many aspects in the development of this now well-defined entity have considerably influenced clinical knowledge and cancer research. Historically, anaplastic large cell lymphomas have been described very well according to their morphologic appearance. Yet, before lineage-bound cytogenetic or molecular genetic diagnoses became available, they could hardly or not at all be distinguished from histiocytic diseases [40]. The morphologic appearance even resembled the aspect of carcinomas. The following characterization with (monoclonal) antibodies, one of them binding to the so-called Ki-1 antigen, first detected on Hodgkin's lymphomas and later named CD30, helped to finally define the entity (also positive for the epithelial membrane antigen (EMA) [24, 80]. Diseases with a rapid but also a relatively long course of growing and shrinking lymph node enlargements, with local as well as systemic inflammatory signs mimicking infectious or autoimmune diseases could be diagnosed as a clinicopathologic entity. The chromosomal translocation t (2;5) (p23;q35) [45] and its molecular counterpart, the fusion gene *NPM-ALK* found in ALCL [58] gave rise to cancer research, when ALK alterations were found in several other cancers such as lung carcinoma and neuroblastoma. Another very intriguing finding was the immunogenicity of the fusion neo-oncoprotein in case of ALCLs [71]. This fact conveniently explained the clinical wax-and-wane courses as well as inflammatory and autoimmune phenomena which all mimicked non-neoplastic diseases and substantially delayed exact diagnosis. The presence of antibodies against the fusion gene, together with the lack of the circulating fusion gene, could be correlated with a better prognosis [63]. These various milestones on the way to more exact diagnoses reflect the progress in oncologic research, where pediatric oncology has always been playing a leading role (Table 1.2).

History of Staging Procedures

The St. Jude's staging system by *Murphy*, an adapted Ann-Arbor system used for Hodgkin's lymphoma, upgraded mediastinal (universally stage III) and abdominal (localized stage II, extended stage III) manifestations, and reserved stage IV for bone marrow or central nervous system involvement. It was broadly accepted worldwide by pediatric

Table 1.2 Time table of some diagnostic milestones relevant for pediatric NHLs

Year	Author	Finding
1864	Virchow	Aleukemic leukemia
1871	Billroth	Multiple lymphomas
1893	Kundrat	Lymphosarkoma
1956/1966	Rappaport	Cytomorphologic differentiation
1958	Burkitt	Endemic African lymphomas
1964	Epstein	Virus detection
1972	Aisenberg	B- and T-cell NHL by surface markers
1972	Manolov	Burkitt NHL: chromosome 14
1974	Lukes, Collins	Immunologic characterization
1975	Lennert	Kiel classification: B- and T-cell NHLs
1975	Köhler	Monoclonal antibody production
1976	Zech	t(8;14) in Burkitt's lymphomas
1981	Korsmeyer	Hierarchic IG gene rearrangement
1982	Dalla-Favera	C-MYC oncogene in Burkitt's NHLs
1982	Taub	C-MYC-IGH fusion in Burkitt's NHL
1982	NCI	Working formulation
1983	Aisenberg	Immunophenotyping of NHLs
1985	Stein	Ki-1 antigen (CD30)
1985	Waldmann	T-cell receptor rearrangement
1988	Delsol	ALCL diagnosis
1989	Le Beau	t(2;5) in ALCL
1994	Harris	REAL classification
1994	Morris	NPM-ALK fusion in ALCL
2000	Alizadeh	DLBCL subtypes by gene expression
2000	Pulford	Auto-antibodies against ALK
2008	Swerdlow	WHO classification
2009	Park	NOTCH and FBXY7 in LBL T-NHL
2017	Szepanovsky	Pediatric DLBCL molecular diagnosis

hemato-oncologists [62]. Before Burkitt's lymphomas were recognized as systemic diseases in most cases, with no imaging techniques available, and local treatment being the only possible therapy, invasive procedures to establish the spread were undertaken. Extensive staging laparotomies, however, were challenging, with defined lymph node regions to be inspected and biopsies to be taken. By using new imaging techniques and realizing that systemic diseases require systemic treatments, these invasive procedures could be avoided in pediatric B- NHLs [8, 75]. In the case of Hodgkin's disease, where local treatment with irradiation played a curative role for a long time for the majority of patients, staging laparotomies were continued to be used until imaging techniques like ultrasound, CT and MRI scans and, last but not least, FDG PET scintigraphy eventually put an end to surgical staging procedures, at least in most cases.

A Brief History of Treatment Concepts

Local Treatment

The development of successful treatment concepts was initially influenced by first successes in Hodgkin's lymphomas with radiotherapy, dating back to the beginning of the twentieth century ([72], rev. in Aisenberg [2]). The successful radiotherapy for Hodgkin's lymphomas stimulated its use for NHL as well. It was only when pediatric NHLs were recognized as systemic diseases, at least in most cases, and multi-agent chemotherapy was studied successfully by several groups, that the role of radiotherapy was challenged. For some time, combined therapies were used [10]. With the introduction of a more precise diagnosis and stage-specific chemotherapeutic treatments, the role of radiotherapy for pediatric NHL came to an end [17, 39, 47, 78], and in due course, surgery met a similar fate. With the exception of small localized tumors and the possible necessity of gut resection in the case of ileo-cecal intussusception by a Burkitt's lymphoma, the role of surgery turned out to be confined to diagnostic interventions [8, 75], making dangerous and mutilating attempts to remove mediastinal tumors in T-NHLs or exenterating attempts to resect disseminated abdominal Burkitt's a thing of the past.

Development of Chemotherapy

The start of chemotherapy for NHLs dates back to the late 1940s when monotherapies proved to cause responses. The use of N-mustard, the pro-drug of oxazaphorines like cyclophosphamide or ifosfamide, was first documented in 1946 [34]. Aminopterin, a prodrug of methotrexate, was able to cause bone marrow remissions in children with acute lymphoblastic leukemia (ALL) [27], and in Africa, *Burkitt,* using whatever he could get hold of in the pioneering situation of limited resources in Africa, recorded responses to single agents like vincristine, cyclophosphamide, and methotrexate in the early 1960s (reviewed in Bösner 1994 [13]).

The Way to Specific Therapies

Two different regimens developed simultaneously using multi-agent chemotherapy. LSA2-L2 [92], stimulated by the successes of multi-agent treatment for childhood lymphoblastic leukemia based on remission induction, consolidation and maintenance treatment, was one of the first to be documented as successful treatment regimen for children with NHL. The other concept represents a condensed repetitive block therapy, stimulated by the successes in Hodgkin's lymphoma and modified for the treatment of NHLs in adults, where it became a standard of care, based on cyclophosphamide, daunomycin, vincristine and prednisolone (CHOP) [29, 51, 53]. As long as the pathologic entities were poorly distinguishable, no clear advantage of the different approaches was obvious**.** Then, in the 1970s, cooperative study groups started to investigate multi-agent chemotherapeutic interventions. In a randomized study for the treatment of localized pediatric NHL of the American Children's

Cancer Study Group (CCG), the specific advantage of the more ALL-treatment (LSA2-L2-like) regimen for lymphoblastic lymphomas and a better outcome for Burkitt's lymphomas with repetitive bloc therapy, consisting of cyclophosphamide, methotrexate, vincristine, and prednisolone (COMP) [38], became apparent. Nearly at the same time, the German Berlin-Frankfurt-Münster (BFM) group studied different regimens for different entities, also successfully using ALL-directed therapy for lymphoblastic lymphoma and condensed block therapy for mature B- (mainly Burkitt's) NHLs [61].

The more precise diagnostics enabled the development of specific therapeutic strategies aimed at the biologic entities. For the most common pediatric NHLs, mature B-cell, LL, ALCL, and PMBCL, different treatment protocols were developed.

Burkitt's Lymphomas

While still in Africa in the 1960s, Denis Burkitt described casuistic successes with monotherapies, since without adequate infrastructure and other supportive structures, intensive combination therapy was simply not available to him. Already at that time, the involvement of the central nervous system appeared to Burkitt to be a prognostically unfavorable factor, and so intrathecal therapy was administered to treat and prevent meningeal spread [94]. Combination therapies evolved, showing the benefit of intensified short block treatment; for extended diseases, high-dose methotrexate and high-dose cytarabine were added. The stepwise adaptation of treatment protocols in France by *Catherine Patte* [67] increased survival rates to about 90%, and this success led to a French American British cooperative protocol [22, 31, 68]. At the same time, a similarly intensified condensed block therapy led to the same results in the BFM group of *Alfred Reiter* [77]. Attempts to reduce the intensity to avoid acute (infections and mucosal damage) or late adverse effects failed for extended diseases but were possible in less disseminated lymphomas [21, 68, 91]. Today, high survival rates are obtained by intensive chemotherapy, but relapses remain challenging [18]. The implementation of immunotherapy by a CD20-specific antibody proved to be of advantage [33, 54] and is now under further clinical investigation.

Primary Mediastinal Large B-Cell Lymphomas

The treatment of PMBCL, when compared to the successes achieved with chemotherapy in Burkitt's and lymphoblastic lymphomas, turned out to be unsatisfactory with Burkitt-cell directed therapy protocols [16]. It was due to the positive experience in adult protocols for this entity that the regimen, involving an anti CD20 antibody and chemotherapeutic dose escalation, was adopted for pediatric PMBCLs, thus improving the diagnosis for this age group [25, 32].

Lymphoblastic Lymphomas

With the start of multi-agent treatment concepts, ALL-directed therapies started to be used for all kinds of pediatric NHLs. The LSA2-L2 protocol created by *Wollner* (see above) was one of the first, documenting response and cure rates in a considerable number, but also noting that for patients with extended disease, higher cure rates could only be achieved with a more intensive treatment. Event-free survival (EFS) rates improved substantially, and prophylactic cranial radiotherapy could be omitted. With one of the most successful multi-agent chemotherapy protocols, based on an ALL-like BFM scheme, a 90% cure rate could be achieved [78]. In an attempt to coordinate the treatment on a large scale, a European cooperation for pediatric NHLs, the European intergroup cooperation for pediatric NHLs (EICNHL), started a cooperative study using the structure of this successful pediatric BFM protocol for lymphoblastic lymphomas. While the high cure rates of the preceding BFM protocol could not be achieved, because of the high toxicity of the treatment, the overall EFS reached nearly 80% [44]. Nevertheless, at the end, the extended European cooperation per se was successful and the overall results agreed perfectly with current data. A risk-adapted treatment according to molecular changes (NOTCH, FBXY7 mutations) is currently under investigation ([65]; Burkhardt, ongoing).

Anaplastic Large Cell Lymphomas

One of the first structured cooperative treatment protocols for treatment of ALCLs, the BFM protocol 1990 to treat the then so-called Ki1 (CD30)-positive NHLs, was developed by the BFM group [76]. Within the international EICNHL group, it was decided to use the BFM backbone for a modified treatment strategy [14]. While EFS rates appeared to be improving, overall survival rates (with respect to survival after a relapse) were favorable in comparison to the other entities. This, together with reported responses to minimal interventions with vinblastine, aimed at palliation and producing durable remissions [15] and since an immunologic response to the t(2;5)-derived NPM-ALK fusion protein was detectable [71], treatment with immune modulation measures became attractive. The toxin-conjugated anti-CD30 antibody was effective in relapsed and resistant diseases [70]. As the fusion protein proved to have tyrosine kinase activity, an anti-ALK tyrosine kinase inhibiting treatment was appropriate and proved valuable in chemo-resistant ALCLs [30, 59].

Post-transplant Lymphoproliferative Disease

PTLDs are defined in the WHO classification. In some immunosuppressed patients, lymphadenopathy, mostly EBV-associated, may occur after transplantation and evolve into overt lymphomas. This resembles in part the evolution of EBV-associated Burkitt's lymphomas in children in Africa immunosuppressed by chronic malaria infection (see above). A strategy to reduce, whenever possible, immunosuppression and an anti-B-cell CD20 antibody therapy, to be applied before installing conventional lymphoma-directed treatment, were therefore developed [28] .

Conclusions and Future Perspectives

The look back at the evolution of diagnosis and treatment of pediatric NHLs brings to mind the enormous diagnostic and therapeutic efforts that were necessary to cleanse cancer of its mystical interpretations, and redefine it in terms of biology and medical treatment systems. This eventually led to excellent cure rates in formerly rapidly fatal diseases (Table 1.3). It is an excellent example of how clinical and laboratory research are mutually stimulating fields, striving for scientific and medical success. Since lymphatic diseases generally appear to be more susceptible to immunologic attacks than other cancers, concepts that favor immunotherapy are very attractive to researchers and clinicians alike. Monoclonal antibodies against B-cell epitopes, pure or toxin conjugated, chimeric antibodies harboring a T-cell receptor moiety or chimeric antigen receptor T-cell therapies are all going to be implemented in chemotherapy protocols sooner or later. Inhibitory substances aiming at specific molecular changes in the diseases are already contributing to current treatment regimens in clinical research. Further analyses of the somatic gene sequences or RNA sequencing might 1 day provide such highly detailed information that prognostically different tumor types can in the future be subjected to different treatment intensities and qualities.

Table 1.3 Most relevant pediatric studies and results for lymphoblastic and mature B-cell NHL

Treatment results for mature B-NHL in Childhood						
Author	Pub.year	Group	Protocol	N patients	EFS%	EFS% CNS+
Patte [67]	2001	SFOP	LMB 89	561	91	79
Patte [68]	2007	FAB/LMB	FAB/LMB96	1111	88	
Gerrard [31]	2008					
Cairo [22]	2007					
Reiter [77]	1999	BFM	BFM90	413	88	65
Wösmann [91]	2005	BFM	BFM95	505	89	69
Tsurusawa [86]	2014	JPLSG	B-NHL 03	321	87	

Modified after Minard-Colin et al. [55]
CNS central nervous system

	Treatment results for lymphoblastic NHL in childhood							
Author	Pub. year	Group	Period	Protocol	Structure	N patients	Radiotherapy	EFS%
Anderson [6]	1993	CCG	1977–1982	CCG-551	LSA2-L2	124 (st. III,IV)	Local	64
Mora [57]	2003	MSKCC	1971–1990	LSA2-L2	LSA2-L2	95	Local	75
Patte [66]	1992	IGR	1981–1989	LMT81	LSA2-L2+ HD MTX	84	Cranial for CNS+	75
Tubergen [87]	1995	CCG	1983–1990	CCG502	LSA2-L2 vs.ADCOMP	143	Local, Craniosp. For CNS+	74
Asselin [7]	2011	POG	1996–2000	POG9404	DF+/− HD MTX	137 incl. T-ALL	Cranial	85
Abromovic [1]	2008	COG	1994–1997	COG5941	DF mod., HD MTX, HD ARAC, CY	85	Cranial for CNS+	78
Reiter [76]	1995	BFM	1986–1990	BFM 86	BFM NHL Non-B	63	Cranial, stages III,IV	84
Reiter [78]	2000	BFM	1990–1995	BFM 90	BFM NHL Non-B	136	Cranial, stages III,IV	90
Burkhardt [16]	2006	BFM	1995–2001	BFM 95	BFM NHL Non-B	198	Cranial for CNS+	80
Uytterbroeck [88]	2008	EORTC	1989–1998	CLG58881	BFM NHL Non-B	119	No	77.5
Bergeron [11]	2015	SFOP	1997–2003	LMT96	Modif.BFM	79	Cranial for CNS+	85
Pillon [69]	2009	AIEOP	1992–1997	AIEOP92	LSA2-L2	55	Cranial for CNS+	69
Termuhlen [85]	2013	COG	2000–2005	COGA5971	CCG ALL vs. BFM	266	Cranial for CNS+	82
Landmann [44]	2017	EICNHL	2003–2008	BFM 90	Rand. Dexa vs.Pred	319	Cranial for CNS+	81

Modified after Minard-Colin et al. [55]
DF Dana Farber consortium ALL protocol, *CNS* central nervous system, *craniosp* craniospinal

References

1. Abromowitch M, Sposto R, Perkins S, Zwick D, Siegel S, Finlay J, Cairo MS, Children's Oncology Group. Shortened intensified multi-agent chemotherapy and non-cross resistant maintenance therapy for advanced lymphoblastic lymphoma in children and adolescents: report from the Children's Oncology Group. Br J Haematol. 2008;143(2):261–7.
2. Aisenberg AC. Historical review of lymphomas. Br J Haematol. 2000;109:466–76.
3. Aisenberg AC, Bloch KJ. Immunoglobulins on the surface of neoplastic lymphocytes. N Engl J Med. 1972;287:272–6.
4. Aisenberg AC, Wilkes BM, Harris NL. Monoclonal antibody studies in non-Hodgkin's lymphoma. Blood. 1983;61(3):469–75.
5. Alizadeh AA, Eisen MB, Davis RE, Ma C, Lossos IS, Rosenwald A, Boldrick JC, Sabet H, Tran T, Yu X, Powell JI, Yang L, Marti GE, Moore T, Hudson J Jr, Lu L, Lewis DB, Tibshirani R, Sherlock G, Chan WC, Greiner TC, Weisenburger DD, Armitage JO, Warnke R, Levy R, Wilson W, Grever MR, Byrd JC, Botstein D, Brown PO, Staudt LM. Distinct types of diffuse large B-cell lymphoma identified by gene expression profiling. Nature. 2000;403(6769):503–11.
6. Anderson JR, Jenkin RD, Wilson JF, Kjeldsberg CR, Sposto R, Chilcote RR, Coccia PF, Exelby PR, Siegel S, Meadows AT, et al. Long-term follow-up of patients treated with COMP or LSA2L2 therapy for childhood non-Hodgkin's lymphoma: a report of CCG-551 from the Childrens Cancer Group. J Clin Oncol. 1993;11(6):1024–32.
7. Asselin BL, Devidas M, Wang C, Pullen J, Borowitz MJ, Hutchison R, Lipshultz SE, Camitta BM. Effectiveness of high-dose methotrexate in T-cell lymphoblastic leukemia and advanced-stage lymphoblastic lymphoma: a randomized study by the Children's Oncology Group (POG 9404). Blood. 2011;118(4):874–83.
8. Attarbaschi A, Mann G, Dworzak M, Trebo M, Mühlegger N, Reiter A, Horcher E, Gadner H, Austrian Cooperative Study Group. The role of surgery in the treatment of pediatric B-cell non-Hodgkin's lymphoma. J Pediatr Surg. 2002;37(10):1470–5.
9. Attarbaschi A, Beishuizen A, Mann G, Rosolen A, Mori T, Uyttebroeck A, Niggli F, Csoka M, Krenova Z, Mellgren K, Kabickova E, Chiang AK, Reiter A, Williams D, Burkhardt B, European Intergroup for Childhood Non-Hodgkin Lymphoma (EICNHL) and the international Berlin-Frankfurt-Münster (i-BFM) Study Group. Children and adolescents with follicular lymphoma have an excellent prognosis with either limited chemotherapy or with a "watch and wait" strategy after complete resection. Ann Hematol. 2013;92(11):1537–41.
10. Aur RJA, Hustu HO, Simone JV, Pinkel D. Therapy of localized and regional lymphosarcoma of childhood. Cancer. 1970;27:1328–31.
11. Bergeron C, Coze C, Segura C, Pacquement H, Gandemer V, Ducassou S, Patte C, Société Française d'Oncologie Pédiatrique (SFOP), France. Treatment of childhood T-cell lymphoblastic lymphoma-long-term results of the SFOP LMT96 trial. Pediatr Blood Cancer. 2015;62(12):2150–6.
12. Billroth H. Multiple lymphome. Wien Med Wochenschr. 1871;21:1065–8.
13. Bösner S. Das Burkitt-Lymphom: Geschichte und Epidemiologie. Marburg (Lahn)/Basilisken –Presse; 1993 ISBN 3-925347-26-7. Marburg, Germany.
14. Brugières L, Le Deley MC, Rosolen A, Williams D, Horibe K, Wrobel G, Mann G, Zsiros J, Uyttebroeck A, Marky I, Lamant L, Reiter A. Impact of the methotrexate administration dose on the need for intrathecal treatment in children and adolescents with anaplastic large-cell lymphoma: results of a randomized trial of the EICNHL Group. J Clin Oncol. 2009a;27(6):897–903.
15. Brugières L, Pacquement H, Le Deley MC, Leverger G, Lutz P, Paillard C, Baruchel A, Frappaz D, Nelken B, Lamant L, Patte C. Single-drug vinblastine as salvage treatment for refractory or relapsed anaplastic large-cell lymphoma: a report from the French Society of Pediatric Oncology. J Clin Oncol. 2009b;27(30):5056–61.
16. Burkhardt B, Zimmermann M, Oschlies I, Niggli F, Mann G, Parwaresch R, Riehm H, Schrappe M, Reiter A, BFM Group. The impact of age and gender on biology, clinical features and treatment outcome of non-Hodgkin lymphoma in childhood and adolescence. Br J Haematol. 2005;131(1):39–49.
17. Burkhardt B, Woessmann W, Zimmermann M, Kontny U, Vormoor J, Doerffel W, Mann G, Henze G, Niggli F, Ludwig WD, Janssen D, Riehm H, Schrappe M, Reiter A. Impact of cranial radiotherapy on central nervous system prophylaxis in children and adolescents with central nervous system-negative stage III or IV lymphoblastic lymphoma. J Clin Oncol. 2006;24(3):491–9.
18. Burkhardt B, Reiter A, Landmann E, et al. Poor outcome for children and adolescent swith progressive disease or relapse of lymphoblastic lymphoma: a report from the Berlin-Frankfurt-Muenster Group. J ClinOncol. 2009;27(20):3363–9.
19. Burkitt DP. A sarcoma involving the jaws of African children. Br J Surg. 1958;46:218–23.
20. Burkitt DP, O'Conor GT. Malignant lymphoma in African children. I. A clinical syndrome. Cancer. 1961;14:258–69.
21. Cairo MS, Krailo MD, Morse M, Hutchinson RJ, Harris RE, Kjeldsberg CR, Kadin ME, Radel E, Steinherz LJ, Morris E, Finlay JL, Meadows AT. Long-term follow-up of short intensive multi-agent chemotherapy without high-dose methotrexate ('Orange') in children with advanced non-lymphoblastic non-Hodgkin's lymphoma: a Children's Cancer Group report. Leukemia. 2002;16(4):594–600.
22. Cairo MS, Gerrard M, Sposto R, Auperin A, Pinkerton CR, Michon J, Weston C, Perkins SL, Raphael M, McCarthy K, Patte C, FAB LMB96 International Study Committee. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. Blood. 2007;109(7):2736–43, Abstract.
23. Dalla-Favera R, Bregni M, Erickson J, Patterson D, Gallo RC, Croce CM. Human c-myc oncogene is located on the region of chromosome 8 that is translocated in Burkitt lymphoma cells. Proc Natl Acad Sci U S A. 1982;79:7824–7.
24. Delsol G, Al Saati T, Gatter KC, et al. Coexpression of epithelial membrane antigen (EMA), Ki-1, and interleukin-2 receptor by anaplastic large cell lymphomas. Diagnostic value in so-called malignant histiocytosis. Am J Pathol. 1988;130(1):59–70.
25. Dunleavy K, Pittaluga S, Maeda LS, Advani R, Chen CC, Hessler J, Steinberg SM, Grant C, Wright G, Varma G, Staudt LM, Jaffe ES, Wilson WH. Dose-adjusted EPOCH-rituximab therapy in primary mediastinal B-cell lymphoma. N Engl J Med. 2013;368(15):1408–16.
26. Epstein MA, Achong BG, Barr YM. Virus particles incultured lymphoblasts from Burkitt's lymphoma. Lancet. 1964;1:702–3.
27. Farber S, Diamond LK, Mercer RD, Sylvester RF Jr, Wolff JA. Temporary remissions in acute leukemia in children produced by folic acid antagonist, 4-aminopteroyl-glutamic acid (ssaminopterin). N Engl J Med. 1948;238:787–93.
28. Faye A, Vilmer EP. Post-transplant lymphoproliferative disorder in children: incidence, prognosis, and treatment options. Paediatr Drugs. 2005;7(1):55–65.
29. Fisher RI, Gaynor ER, Dahlberg S, Oken MM, Grogan TM, Mize EM, et al. Comparison of a standard regimen (CHOP) with three intensive chemotherapy regimens for advanced non-Hodgkin's lymphoma. N Engl J Med. 1993;328:1002–6.
30. Gambacorti Passerini C, Farina F, Stasia A, Redaelli S, Ceccon M, Mologni L, Messa C, Guerra L, Giudici G, Sala E, Mussolin L, Deeren D, King MH, Steurer M, Ordemann R, Cohen AM, Grube M, Bernard L, Chiriano G, Antolini L, Piazza R. Crizotinib in advanced, chemoresistant anaplastic lymphoma kinase-positive lymphoma patients. J Natl Cancer Inst. 2014;106(2):djt378.

31. Gerrard M, Cairo MS, Weston C, Auperin A, Pinkerton R, Lambilliote A, Sposto R, McCarthy K, Lacombe MJ, Perkins SL, Patte C, FAB LMB96 International Study Committee. Excellent survival following two courses of COPAD chemotherapy in children and adolescents with resected localized B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study. Br J Haematol. 2008;141(6):840–7.
32. Giulino-Roth L, O'Donohue T, Chen Z, Bartlett NL, LaCasce A, Martin-Doyle W, Barth MJ, Davies K, Blum KA, Christian B, Casulo C, Smith SM, Godfrey J, Termuhlen A, Oberley MJ, Alexander S, Weitzman S, Appel B, Mizukawa B, Svoboda J, Afify Z, Pauly M, Dave H, Gardner R, Stephens DM, Zeitler WA, Forlenza C, Levine J, Williams ME, Sima JL, Bollard CM, Leonard JP. Outcomes of adults and children with primary mediastinal B-cell lymphoma treated with dose-adjusted EPOCH-R. Br J Haematol. 2017;179(5):739–47. https://doi.org/10.1111/bjh.14951.
33. Goldman S, Smith L, Galardy P, Perkins SL, Frazer JK, Sanger W, Anderson JR, Gross TG, Weinstein H, Harrison L, Shiramizu B, Barth M, Cairo MS. Rituximab with chemotherapy in children and adolescents with central nervous system and/or bone marrow-positive Burkitt lymphoma/leukaemia: a Children's Oncology Group report. Br J Haematol. 2014;167(3):394–401.
34. Goodman LS, Wintrobe MM, Dameshek W, Goodman MJ, Gilman AZ, McLennan MI. Nitrogen mustard therapy. Use of methyl-bis (beta-chloroethyl) amino hydrochloride for Hodgkin's disease, lymphosarcoma, leukemia and certain allied and miscellaneous disorders. J Am Med Assoc. 1946;132:26–132.
35. Harrington DS, Weissenberger DD, Purtilo DT. Malignant lymphomas in X-linked lymphoproliferative disease. Cancer. 1987;59:1419–29.
36. Harris NL, Jaffe ES, Banks PM, Chan JKC, Cleary ML, Delsol G, De Wolf-Peeters C, Falini B, Gatter KC, Grogan TM, Isaacson PG, Knowles DM, Mason DY, Muller-Hermelink H-K, Pileri SA, Piris MA, Ralfkiaer E, Warnke RA. A revised European-American classification of malignant lymphoid neoplasms: a proposal from the international Lymphoma Study Group. Blood. 1994;84:1361–92.
37. IUIS-WHO Nomenclature Subcommittee. Nomenclature for clusters of differentiation (CD) of antigens defined on human leukocyte populations. Bull World Health Organ. 1984;62(5):809–15.
38. Jenkin RD, Anderson JR, Chilcote RR, Coccia PF, Exelby PR, Kersey JH, Kushner JH, Meadows AT, 7Siegel SE, Sposto R J et al. The treatment of localized non-Hodgkin's lymphoma in children: a report from the Children's Cancer Study Group., Clin Oncol 1984;2(2):88–97.
39. Jereb B, Wollner N, Kosloff C, Exelby P. The role of local radiation in the treatment of non-Hodgkin lymphoma in children. Med Pediatr Oncol. 1981;9(2):157–66.
40. Kaneko Y, Frizzera G, Edamura S, Maseki N, Sakurai M, Komada Y, Sakurai M, Tanaka H, Sasaki M, Suchi T, et al. A novel translocation, t(2;5)(p23;q35), in childhood phagocytic large T-cell lymphoma mimicking malignant histiocytosis. Blood. 1989;73(3):806–13.
41. Köhler G, Milstein C. Continuous cultures of fused cells secreting antibody of predefined specificity. Nature. 1975;256:495–7.
42. Korsmeyer SJ, Hieter PA, Ravetch JV, Poplack DG, Waldmann TA, Leder P. Developmental hierarchy of immunoglobulin gene rearrangements in human leukemia pre-B cells. Proc Natl Acad Sci U S A. 1981;78:7096–100.
43. Kundrat H. über Lympho-sarkomatosis. Wien Klin Wochenschr. 1893;6:234–9.
44. Landmann E, Burkhardt B, Zimmermann M, Meyer U, Woessmann W, Klapper W, Wrobel G, Rosolen A, Pillon M, Escherich G, Attarbaschi A, Beishuizen A, Mellgren K, Wynn R, Ratei R, Plesa A, Schrappe M, Reiter A, Bergeron C, Patte C, Bertrand Y. Results and conclusions of the European intergroup EURO-LB02 trial in children and adolescents with lymphoblastic lymphoma. Haematologica. 2017;102(12):2086–96.
45. Le Beau MM, Bitter MA, Larson RA, Doane LA, Ellis ED, Franklin WA, Rubin CM, Kadin ME, Vardiman JW. The t(2;5)(p23;q35): a recurring chromosomal abnormality in Ki-1-positive anaplastic large cell lymphoma. Leukemia. 1989;3(12):866–70.
46. Lennert K, Mohri N, Stein H, Kaiserling E. Histopathology of malignant lymphomas. Br J Haematol. 1975;31:193–203.
47. Link MP, Donaldson SS, Berard CW, Shuster JJ, Murphy SB. Results of treatment of childhood localized non-Hodgkin's lymphoma with combination chemotherapy with or without radiotherapy. N Engl J Med. 1990;322(17):1169–74.
48. Lukes RJ, Collins RD. Immunologic characterization of human malignant lymphomas. Cancer. 1974;34(4 Suppl):1488–503.
49. Magrath IT. Lymphocyte differentiation: an essential basis for the comprehension of lymphoid neoplasia. J Natl Cancer Inst. 1981;67(3):501–14.
50. Magrath I. Historical perspective: the evolution of modern concepts of lymphoid neoplasia. In: Magrath I, editor. The lymphoid neoplasms. 3rd ed. London: Edward Arnold; 2010. p. 23–45.
51. Magrath IT, Janus C, Edwards BK, Spiegel R, Jaffe ES, Berard CW, Miliauskas J, Morris K, Barnwell R. An effective therapy for both undifferentiated (including Burkitt's) lymphomas and lymphoblastic lymphomas in children and young adults. Blood. 1984;63(5):1102–11.
52. Manolov G, Manolova Y. Marker band in one chromosome 14 from Burkitt lymphoma. Nature. 1972;237:33–4.
53. McKelvey EM, Gottlieb JA, Wilson HE, Haut A, Talley RW, Stephens R, Lane M, Gamble JF, Jones SE, Grozca DN, Gutterman J, Coltman C Jr, Moon TE. Hydroxyldaunomycin(adriamycin) combination chemotherapy in malignantlymphoma. Cancer. 1976;38:1484–93.
54. Meinhardt A, Burkhardt B, Zimmermann M, Borkhardt A, Kontny U, Klingebiel T, Berthold F, Janka-Schaub G, Klein C, Kabickova E, Klapper W, Attarbaschi A, Schrappe M, Reiter A, Berlin-Frankfurt-Münster Group. Phase II window study on rituximab in newly diagnosed pediatric mature B-cell non-Hodgkin's lymphoma and Burkitt leukemia. J Clin Oncol. 2010;28(19):3115–21.
55. Minard-Colin V, Brugières L, Reiter A, Cairo MS, Gross TG, Woessmann W, Burkhardt B, Sandlund JT, Williams D, Pillon M, Horibe K, Auperin A, Le Deley MC, Zimmermann M, Perkins SL, Raphael M, Lamant L, Klapper W, Mussolin L, Poirel HA, Macintyre E, Damm-Welk C, Rosolen A, Patte C. Non-hodgkin lymphoma in children and adolescents: progress through effective collaboration, current knowledge, and challenges ahead. J Clin Oncol. 2015;33(27):2963–74.
56. Minden MD, Toyonaga B, Ha K, Yanagi Y, Chin B, Gelfand E, Mak T. Somatic rearrangement of T-cell antigen receptor genes in human T-cell malignancies. Proc Natl Acad Sci U S A. 1985;82:1224–7.
57. Mora J, Filippa DA, Qin J, Wollner N. Lymphoblastic lymphoma of childhood and the LSA2-L2 protocol: the 30-year experience at memorial-Sloan-Kettering Cancer center. Cancer. 2003;98(6):1283–91.
58. Morris SW, Kirstein MN, Valentine MB, Dittmer KG, Shapiro DN, Saltman DL, Look AT. Fusion of a kinase gene, ALK, to a nucleolar protein gene, NPM, in non-Hodgkin's lymphoma. Science. 1994;263(5151):1281–4.
59. Mossé YP, Voss SD, Lim MS, Rolland D, Minard CG, Fox E, Adamson P, Wilner K, Blaney SM, Weigel BJ. Targeting ALK with crizotinib in pediatric anaplastic large cell lymphoma and inflammatory myofibroblastic tumor: a Children's Oncology Group Study. J Clin Oncol. 2017;35(28):3215–21.
60. Mukherjee S. The emperor of all maladies 39–50. In: Mukherjee S, editor. The emperor of all maladies. London: fourth estate; 2011.
61. Müller-Weihrich S, Beck J, Henze G, Jobke A, Kornhuber B, Lampert F, Ludwig R, Prindull G, Schellong G, Spaar HJ, et al. BFM study 1981/83 of the treatment of highly malignant non-Hodgkin's lymphoma in children: results of therapy stratified

according to histologic immunological type and clinical stage. Klin Padiatr. 1984;196(3):135–42. German.
62. Murphy SB. Classification, staging and end results of treatment of childhood non-Hodgkin's lymphomas: dissimilarities from lymphomas in adults. Semin Oncol. 1980;7(3):332–9.
63. Mussolin L, Damm-Welk C, Pillon M, Zimmermann M, Franceschetto G, Pulford K, Reiter A, Rosolen A, Woessmann W. Use of minimal disseminated disease and immunity to NPM-ALK antigen to stratify ALK-positive ALCL patients with different prognosis. Leukemia. 2013;27(2):416–22.
64. National Cancer Institute sponsored study of classifications of non-Hodgkin's lymphomas: summary and description of a working formulation for clinical usage. The non-Hodgkin's lymphoma pathologic classification project. Cancer. 1982;49(10):2112–35.
65. Park MJ, Taki T, Oda M, Watanabe T, Yumura-Yagi K, Kobayashi R, Suzuki N, Hara J, Horibe K, Hayashi Y. FBXW7 and NOTCH1 mutations in childhood T cell acute lymphoblastic leukaemia and T cell non-Hodgkin lymphoma. Br J Haematol. 2009;145(2):198–206.
66. Patte C, Kalifa C, Flamant F, Hartmann O, Brugières L, Valteau-Couanet D, Bayle C, Caillaud J-M, Lemerle J. Results of the LMT81 protocol, a modified LSA2L2 protocol with high dose methotrexate, on 84 children with non-B-cell (lymphoblastic) lymphoma. Med Pediatr Oncol. 1992;20(2):105–13.
67. Patte C, Auperin A, Michon J, et al. The Société Française d'Oncologie Pédiatrique LMB89 protocol: highly effective multiagent chemotherapy tailored to the tumor burden and initial response in 561 unselected children with B-cell lymphomas and L3 leukemia. Blood. 2001;97:3370–9.
68. Patte C, Auperin A, Gerrard M, Michon J, Pinkerton R, Sposto R, Weston C, Raphael M, Perkins SL, McCarthy K, Cairo MS, FAB/LMB96 International Study Committee. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. Blood. 2007;109(7):2773–80.
69. Pillon M, Piglione M, Garaventa A, Conter V, Giuliano M, Arcamone G, Mura R, Cellini M, D'Amore ES, Varotto S, Mussolin L, Rosolen A, AIEOP-NHL Committee. Long-term results of AIEOP LNH-92 protocol for the treatment of pediatric lymphoblastic lymphoma: a report of the Italian association of pediatric hematology and oncology. Pediatr Blood Cancer. 2009;53(6):953–9.
70. Pro B, Advani R, Brice P, Bartlett NL, Rosenblatt JD, Illidge T, Matous J, Ramchandren R, Fanale M, Connors JM, Yang Y, Sievers EL, Kennedy DA, Shustov A. Brentuximab vedotin (SGN-35) in patients with relapsed or refractory systemic anaplastic large-cell lymphoma: results of a phase II study. J Clin Oncol. 2012;30(18):2190–6.
71. Pulford K, Falini B, Banham AH, Codrington D, Roberton H, Hatton C, Mason DY. Immune response to the ALK oncogenic tyrosine kinase in patients with anaplastic large-cell lymphoma. Blood. 2000;96(4):1605–7.
72. Pusey WA. Cases of sarcoma and Hodgkin's disease treated by exposures to x-rays: a preliminary report. J Am Med Assoc. 1902;38:166–9.
73. Randhawa PS, Jaffe R, Demetris AJ, Nalesnik M, Starzl TE, Chen YY, Weiss LM. Expression of Epstein-Barr virus-encoded small RNA (by the EBER-1 gene) in liver specimens from transplant recipients with post-transplantation lymphoproliferative disease. N Engl J Med. 1992;327(24):1710–4.
74. Rappaport H. Atlas of tumor pathology. Washington DC, USA: United States Armed Forces Institute of Pathology; 1966. Tumors of the haematopoeitic system: Section 3 3, fascicle 8; p. 442.
75. Reiter A, Zimmermann W, Zimmermann M, von Schweinitz D, Riehm H, Mildenberger H. The role of initial laparotomy and second-look surgery in the treatment of abdominal B-cell non-Hodgkin's lymphoma of childhood. A report of the BFM Group. Eur J Pediatr Surg. 1994;4(2):74–81.
76. Reiter A, Schrappe M, Parwaresch R, Henze G, Muller-Weihrich S, Sauter S, Sykora K-W, Ludwig W-D, Gadner H, Riehm H. Non-Hodgkin's lymphomas of childhood and adolescence: results of a treatment stratified for biologic subtypes and stage, a report of the Berlin-Frankfort-Munster Group. J Clin Oncol. 1995;13:359–72.
77. Reiter A, Schrappe M, Tiemann M, Ludwig WD, Yakisan E, Zimmermann M, Mann G, Chott A, Ebell W, Klingebiel T, Graf N, Kremens B, Müller-Weihrich S, Plüss HJ, Zintl F, Henze G, Riehm H. Improved treatment results in childhood B-cell neoplasms with tailored intensification of therapy: a report of the Berlin-Frankfurt-Münster Group Trial NHL-BFM 90. Blood. 1999;94(10):3294–306.
78. Reiter A, Schrappe M, Ludwig WD, Tiemann M, Parwaresch R, Zimmermann M, Schirg E, Henze G, Schellong G, Gadner H, Riehm H. Intensive ALL-type therapy without local radiotherapy provides a 90% event-free survival for children with T-cell lymphoblastic lymphoma: a BFM Group report. Blood. 2000;95(2):416–21.
79. Robb-Smith AHT. Reticulosis and reticulosarcoma: a histological classification. J Pathol Bacteriol. 1938;47:457–80.
80. Stein H, Mason DY, Gerdes J, O'Connor N, Wainscoat J, Pallesen G, Gatter K, Falini B, Delsol G, Lemke H, Schwarting R, Lennert K. The expression of the Hodgkin's disease associated antigen Ki- 1 in reactive-and neoplastic lymphoid tissue: evidence that reed-Stemberg cells and histiocytic malignancies are derived from activated lymphoid cells. Blood. 1985;66:848–58.
81. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, et al., editors. WHO classification of tumours of haematopoietic and lymphoid tissues. 4th ed. Lyon: IARC Press; 2008.
82. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, Advani R, Ghielmini M, Salles GA, Zelenetz AD, Jaffe ES. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127(20):2375–90.
83. Szczepanowski M, Lange J, Kohler CW, Masque-Soler N, Zimmermann M, Aukema SM, Altenbuchinger M, Rehberg T, Mahn F, Siebert R, Spang R, Burkhardt B, Klapper W. Cell-of-origin classification by gene expression and MYC-rearrangements in diffuse large B-cell lymphoma of children and adolescents. Br J Haematol. 2017;179(1):116–9.
84. Taub R, Kirsch I, Morton C, Lenoir G, Swan D, Tronick S, Aaronson S, Leder P. Translocation of the c-myc gene into the immunoglobulin heavy chain locus in human Burkitt's lymphoma and murine plasmacytoma cells. Proc Natl Acad Sci U S A. 1982;79:7837–41.
85. Termuhlen AM, Smith LM, Perkins SL, Lones M, Finlay JL, Weinstein H, Gross TG, Abromowitch M. Disseminated lymphoblastic lymphoma in children and adolescents: results of the COG A5971 trial: a report from the Children's Oncology Group. Br J Haematol. 2013;162(6):792–801.
86. Tsurusawa M, Mori T, Kikuchi A, Mitsui T, Sunami S, Kobayashi R, Takimoto T, Saito A, Watanabe T, Fujimoto J, Nakazawa A, Ohshima K, Horibe K, Lymphoma Committee of Japanese Pediatric Leukemia/Lymphoma Study Group. Improved treatment results of children with B-cell non-Hodgkin lymphoma: a report from the Japanese Pediatric Leukemia/Lymphoma Study Group B-NHL03 study. Pediatr Blood Cancer. 2014;61(7):1215–21.
87. Tubergen DG, Krailo MD, Meadows AT, Rosenstock J, Kadin M, Morse M, King D, Steinherz PG, Kersey JH. Comparison of treatment regimens for pediatric lymphoblastic non-Hodgkin's lymphoma: a Childrens Cancer Group study. J Clin Oncol. 1995;13(6):1368–76.
88. Uyttebroeck A, Suciu S, Laureys G, Robert A, Pacquement H, Ferster A, Marguerite G, Mazingue F, Renard M, Lutz P, Rialland X, Mechinaud F, Cavé H, Baila L, Bertrand Y, Children's Leukaemia Group (CLG) of the European Organisation for Research and Treatment of Cancer (EORTC). Treatment of childhood T-cell lymphoblastic lymphoma according to the strategy for acute lymphoblastic leukaemia, without radiotherapy: long term results of the EORTC CLG 58881 trial. Eur J Cancer. 2008;44(6):840–6.

89. Virchow R. Die Krankenhaften Geschwulste. In: Dreissig Vorlesungen gehalten wahrend des Wintersemestes (1862–1863) an der Universitat zu Berlin, vol 2, pp 728–38. A Hirschwald, Berlin; 1864–1865.
90. Waldmann TA, Davis MM, Bongiovanni KF, Korsmeyer SJ. Rearrangements of genes for the antigen receptor on T cells as markers of lineage and clonality in human lymphoid neoplasms. N Engl J Med. 1985;313:776–83.
91. Woessmann W, Seidemann K, Mann G, Zimmermann M, Burkhardt B, Oschlies I, Ludwig WD, Klingebiel T, Graf N, Gruhn B, Juergens H, Niggli F, Parwaresch R, Gadner H, Riehm H, Schrappe M. Reiter A; BFM Group.The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. Blood. 2005;105(3):948–58.
92. Wollner N, Burchenal JH, Lieberman PH, Exelby P, D'Angio G, Murphy ML. Non-Hodgkin's lymophoma in children. A comparison of two modalities of therapy. Cancer. 1976;37:123–34.
93. Zech L, Haglund U, Nilsson K, Klein G. Characteristic chromosomal abnormalities in biopsies and lymphoid-cell lines from patients with Burkitt and non-Burkitt lymphomas. Int J Cancer. 1976;17(1):47–56.
94. Ziegler JL, Bluming AZ. Intrathecal chemotherapy in Burkitt's lymphoma. Br Med J. 1971;3(5773):508–12.

2 Epidemiology of Non-Hodgkin Lymphomas in Childhood and Adolescence

Nirav Thacker and Oussama Abla

Introduction

The incidence of cancer in children is relatively low, representing 0.5–4.6% of the total number of cancer cases in the whole population [1]. However, childhood cancer has a high mortality-incidence ratio with approximately 80.000 childhood deaths attributable to cancer in a single year, with nearly 90% of deaths occurring in less developed countries [1]. Even in the United States of America (USA) where the mortality-incidence ratio of cancer is one of the lowest (<0.15) and overall survival (OS) is greater than 80%, cancer is the second most common cause of death in children less than 15 years of age [2]. During the last three decades, the incidence of childhood cancer has increased by approximately 13% [3]. Lymphoma is the third most common childhood cancer in the 0–14 year-age group and the most common malignancy in the 15–19 year-age group [3].

Lymphoma settles for a "BRONZE" among childhood cancer with Non-Hodgkin lymphoma (NHL) accounting for 7% of all malignancies under the age of 19 years. It is estimated that nearly 1040 new cases of NHL (620 children and 420 adolescents) will be diagnosed in the USA annually, with an incidence of 12.6 per million under the age of 19 years, while having a striking geographical variation worldwide [4, 5]. The incidence of NHL in children and adolescents has increased over the last four decades and is more frequent in boys as compared to girls [4].

Non-Hodgkin lymphoma encompasses a variety of lymphoid malignancies divided in different subtypes according to their pathology, molecular biology, clinical presentation, and treatment, while being united by their staging. Unlike adult NHLs which are mostly of low- to intermediate-grade, most childhood NHLs are high-grade malignancies [6, 7]. Therapy of NHL has been one of the success stories in childhood malignancies with a near doubling of survival rates over the last four decades, which has contributed significantly to the pool of childhood cancer survivors. This has created a unique population at increased risk of various health concerns attributable to prior therapies [8]. Therefore, in recent years, concerted efforts across the global pediatric oncology community have been made to identify factors for understanding the biology, creating a better risk stratification and helping to reduce therapy in the appropriate group to reduce long-term side effects.

This chapter will focus on the epidemiology of NHL in childhood and adolescence and will explain the differences in incidence of NHL according to age, gender, and geographical regions.

Descriptive Epidemiology

Incidence

Lymphomas (Hodgkin's disease and NHLs) are the third most common malignancy in childhood after acute leukemias and brain tumors, accounting for nearly 15% of all cancers diagnosed in children and adolescents less than 20 years in the USA as highlighted in Fig. 2.1 [9]. Non-Hodgkin's lymphoma accounts for approximately 60% of all lymphomas diagnosed in children (0–14 years) in the more developed countries, and the ratio is reversed in adolescents (15–19 years) [5]. The incidence rates of NHL in children and adolescents range from 10 to 15 per million in most developed countries, with NHL accounting for almost 7% of all cancers diagnosed under the age of 20 years [10].

Being a heterogeneous disease (Fig. 2.2), NHL has a wide variation in incidence by geographical regions, age, pathology, and gender. These variations form an important part of the epidemiological discussion on NHL. There is scarcity of national cancer registries in low- and middle-income

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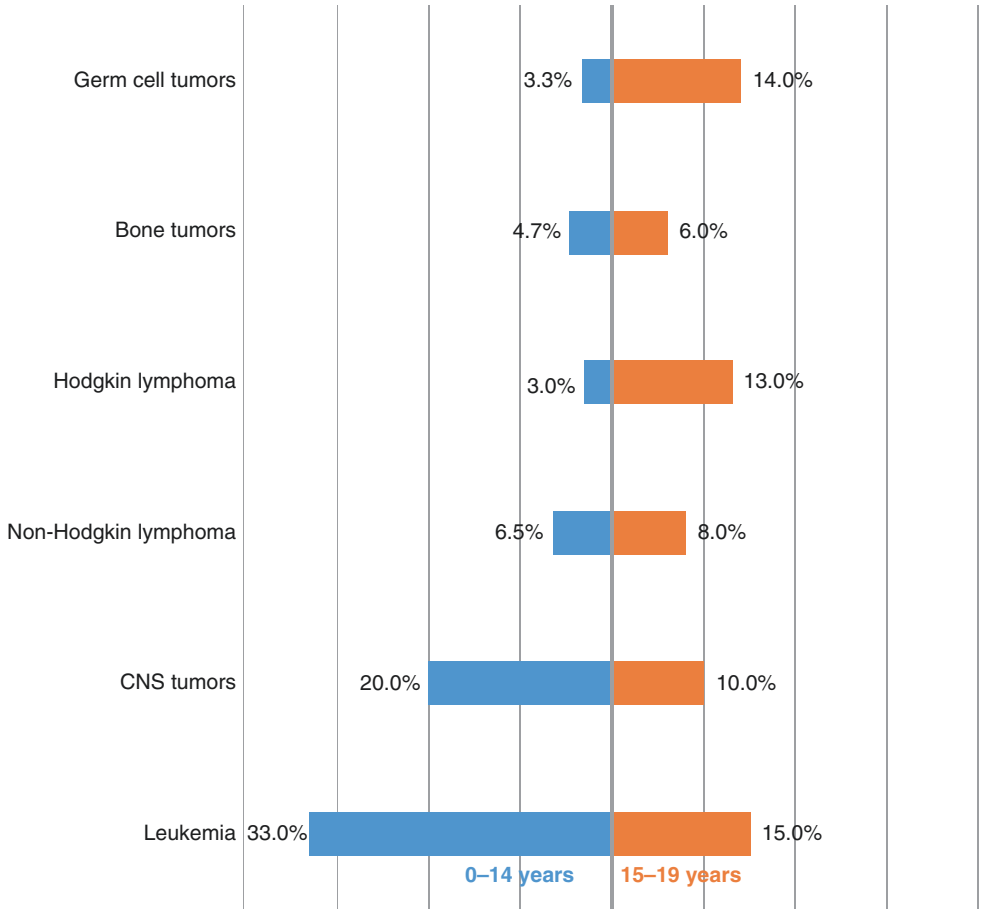

Fig. 2.1 Percentage Incidence of Cases of Childhood and Adolescent Cancers in the United States of America

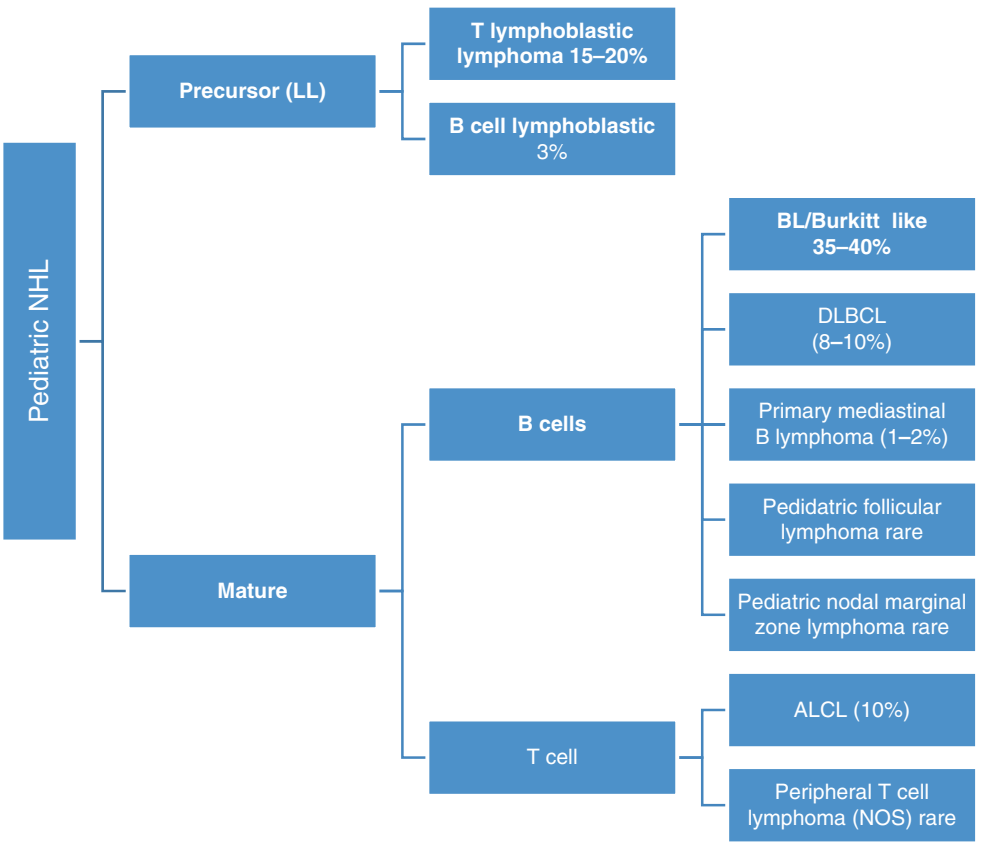

Fig. 2.2 Most common histologic subtypes of childhood NHL

countries (LMICs), limiting our ability to have a true estimate of childhood cancers not only in LMICs but also globally, since high-income countries (HIC) account for only 20% of the global incidence of pediatric cancers [11]. Therefore, in this chapter we will focus mainly on data from developed countries with inclusion of data from LMICs wherever available.

Incidence of NHL According to Histopathologic Entities

Burkitt's and Burkitt's-like lymphomas (BL) have a peak incidence between 5 and 14 years of age and have one of the highest gender gaps being almost 5.0-fold higher for male children (3.2 per million) than for female children (0.7 per million) [9]. The incidence of diffuse large B-cell lymphoma (DLBCL) and anaplastic large cell lymphoma (ALCL) gradually increases across childhood with a sharp increase in adolescence. Primary mediastinal large B-cell lymphoma (PMBCL) is rare before adolescence, while lymphoblastic lymphoma (LBL) has a relatively constant incidence across all age groups. Overall, NHLs are quite rare in infants (Fig. 2.3) [9, 12].

Endemic BL is mainly confined to equatorial Africa and Papua New Guinea, accounting for nearly half the cases of childhood cancers and 90% of cases of childhood lymphomas in the high-risk endemic areas, with an estimated incidence of 30–60 per million children per year and a peak incidence at 4–7 years of age with a male-to-female ratio of 2:1 [13, 14]. Nearly 95% of endemic BL are EBV-positive. In contrast, sporadic BL comprises nearly 30% of pediatric lymphomas in the USA accounting for 1% of all malignancies, with an incidence of 2.3 per million per year and a peak at 3–12 years of age. Sporadic BL has a much wider male-to-female ratio of 3.9:1.1. Only 20–30% of sporadic BL are EBV-positive [15].

The incidence of immunodeficiency-associated NHL variant is 22 per 100.000 person years in the USA. Among them, NHL is primarily seen in subjects infected by the human immunodeficiency virus (HIV) who are particularly prone to develop the acquired immunodeficiency syndrome (AIDS) and less commonly in patients with other types of immunodeficiency such as the posttransplant population. A steep incidence was noted since the late 1980s, due to the growing epidemics of HIV infection. In HIV-positive patients, BL typically affects those with a relatively high CD4+ T-cell count (>200/μl) and without other opportunistic infections. In contrast to other HIV-associated lymphomas, the rate of BL in the HIV-positive population has not decreased with the advent of powerful antiretroviral therapy. Around 40% of HIV-associated BL are EBV-positive [14, 16].

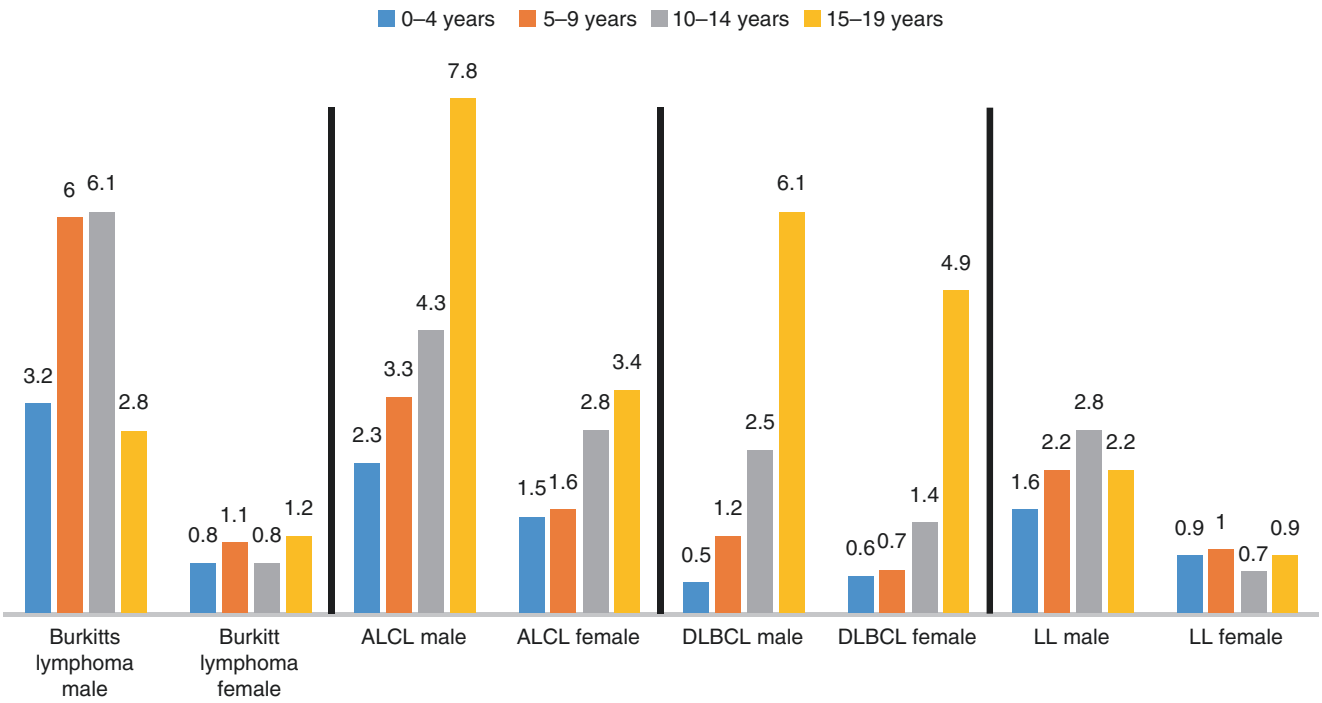

Fig. 2.3 NHL Age specific Incidence by Histolgic group, sex and age, SEER, 1977–95

Incidence of NHL According to Geographical Regions

Among childhood malignancies, NHL has the maximum variation in incidence across the globe as illustrated in Fig. 2.4. The incidence of NHL peaks in equatorial Africa, where it can constitute up to almost half of all malignancies [17]. Rates of NHL vary by almost 30-fold from Asia (India) to sub-Saharan Africa (Malawi) [10]. The higher incidence of NHL in sub-Saharan Africa is mainly contributed by endemic BL which has a strong association with EBV infection and immune modulation by malaria [14]. Endemic BL is the commonest malignancy in Africa where it alone accounts for almost 25% of all cancers in balanced registries. The so-called zone of "lymphoma belt" particularly lies in between 15 degrees north and south of the equator, receiving heavy rainfalls and temperatures above 60° F.

Overall, pediatric cancers are concentrated in LMICs, which account for more than 80% of the global burden of childhood cancer, and are estimated to contribute to 90% of childhood NHL cases diagnosed worldwide [11].

Incidence of NHL According to Age

Overall, NHL accounts for 7% of all malignancies under the age of 20 years; however, there is considerable variation in the incidence depending on the age group. NHL is quite rare in infancy, following which its incidence rapidly increases up to the age of 4 years; the rapidity of its rise subsequently decreases with additional spurt into adolescence. The tumor contributing to the bulk of cases changes from BL in childhood to DLBCL and other lymphomas in adolescence [4, 9]. As a proportion of total cancers, the incidence of NHL in the USA increases from 3% in the 1–4 year-age group to 8–9% in the 5–14 year-age group and remains stable throughout adolescence. Similar contribution from NHL is also noted in European registries where it accounts for 4.5% of total cancers in the 1–4 year-age group and 8–10% thereafter until adolescence as highlighted in Fig. 2.5 [4, 18].

The highest incidence of NHL in childhood is in the 15–19 year-age group with 18.3 and 15.9 per million in the USA and Europe. Nevertheless, NHL accounts for a lower proportion of cancer (8%) in this age group due to a relatively

Fig. 2.4 Global variation in incidence of NHL in children and adolescent

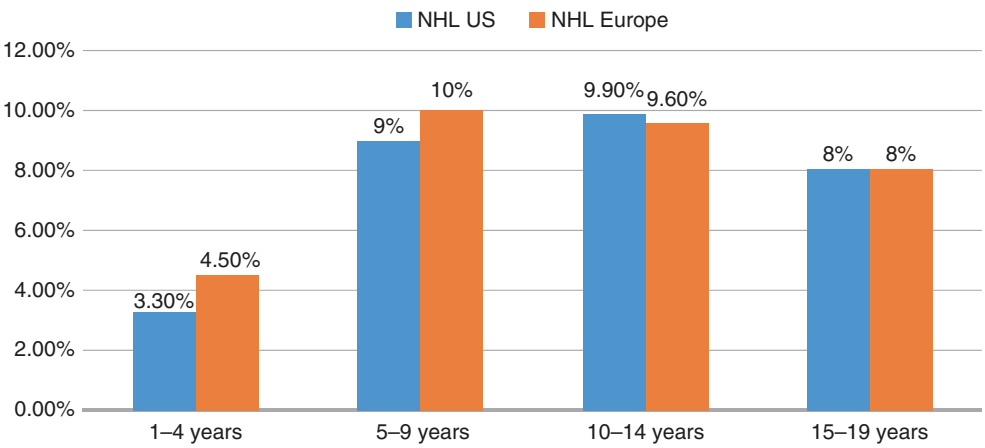

Fig. 2.5 NHL as percentage of total tumor by age

higher proportion of Hodgkin's lymphoma. Of note, the time period for the US data is 1975–2014, while for European data it is 1978–1997 [4, 18]. While the age-specific incidence increases for both males and females, the gap in the incidence rate between the two genders widens considerably after 4 years of age [9]. Furthermore, there is a geographical variation in the age distribution of NHL with the highest rates in Africa at 5–9 years of age in Malawi (318.3 per million person-years) and Uganda (106.6) while at 0–4 years in Egypt as compared to a peak at 15–19 years in the developed countries. On the other hand, there was minimal variation in incidence by age in Croatia, Ukraine, Brazil, Philippines, and India [10].

Incidence of NHL According to Gender

Across all international registries, males outweigh females in the incidence rates for NHL [10]. In the Surveillance, Epidemiology, and End Results program (SEER) registry, across all age groups, male children account for 70% for all cases of NHL. Male predominance is more pronounced in children less than 15 years when compared to adolescents (15–19 years) as illustrated in Fig. 2.3 [9]. The overall incidence of lymphoma increases with age for both genders, with males achieving a rapid increase from 4 years of age and females achieving similar increase after 10–14 years of age (Fig. 2.6) [9].

As for the histological subtypes, males continue their dominance for most subtypes except for PMBCL and B-cell precursor lymphoblastic lymphoma where the incidence is equal for both genders [12]. The male-to-female ratio is most pronounced for BL being 3.9:1.1 [15].

The incidence of DLBCL and other lymphomas (including ALCL) increases for both genders with age with a rapid rise in adolescents, while the incidence of lymphoblastic lymphoma remains fairly constant across all age groups for both male and female children. However, for BL, the incidence for females remains fairly constant across all age groups, while the incidence for male children peaks in the 5–14 year-age group followed thereafter by a decrease in adolescence (Fig. 2.3) [9].

Incidence of NHL According to Race/Ethnicity

Similar to the trend in most childhood cancers, the incidence of NHL is higher in whites than in African Americans [4]. However, among the most common three childhood cancers, the difference in incidence among lymphomas is the least. While looking more closely at age subgroups, the incidence is significantly higher in whites in the 5–9 year and 15–19 year-groups while in the other age groups the incidence is almost the same. Burkitt's lymphoma is more common in non-Hispanic whites than in Hispanic whites (3.2 vs. 2.0 cases per million) [15].

Time Trends of the Incidence of NHL

The incidence of NHL has increased from 9.5 per million to 13.4 per million from 1975 to 2014 (Fig. 2.7) at an annual percentage change of approximately 1% in the USA, while Europe showed similar trends with an average annual percentage change of 0.83% in children and 1.73% in adolescents from 1978 to 1997. This was mainly due to an increase in the incidence of non-BL (category IIb) as the incidence of BL (IIc) and others (IIe) has more or less remained constant [18].

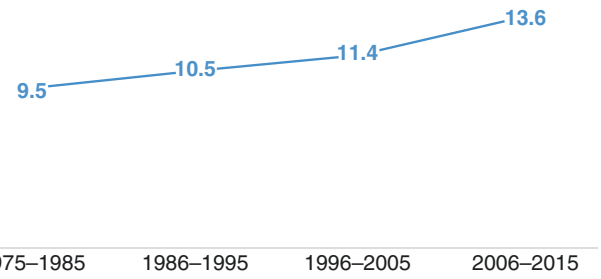

Fig. 2.7 Time trend for incidence of childhood and adolescent NHL

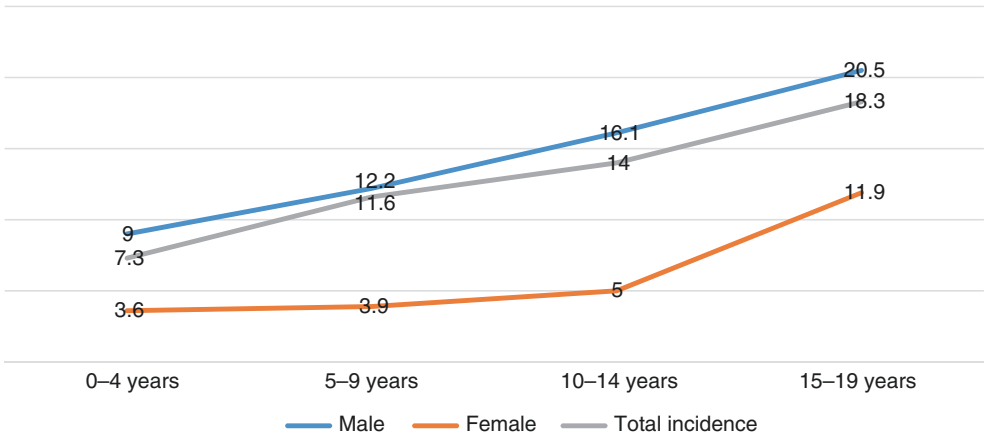

Fig. 2.6 Gender and age specific incidence of childhood NHL

Risk Factors

Proven Risk Factors

- *Immunodeficiency*
  Congenital and acquired immunodeficiency syndromes are significant risk factors for the development of lymphomas (Table 2.1). Most of these cases are EBV-positive. Acquired immunodeficiency is mainly due to HIV infection or posttransplant immunosuppression. Posttransplant lymphoproliferative disease (PTLD) accounts for about 3% of all pediatric NHL diagnoses; and the majority (65%) of them are DLBCLs with less than 10% having BL [19].
  HIV increases the risk of developing NHL in children by 150 times with the majority being of B-cell lineage (BL or BL-like). In fact NHL might be the first AIDS defining manifestation in some children [20–23].

- *Previous Neoplasm (Secondary Malignancy)*
  NHL can present as a second malignancy after cancer therapy; however its incidence is low with only 11 (0.3%) of 2968 of children in the German Childhood Cancer Registry diagnosed with secondary NHL [24].

Substantial Evidence: Cofactors

- *Epstein-Barr Virus (EBV)*
  EBV is present in almost all endemic BL, found in 20–30% of sporadic BL, 40% of HIV-associated BL, and in a high number of primary immunodeficiency-associated BL or PTLD. EBV is known to immortalize B cells and postulated to provide a block in the apoptotic clearance of B cells with *MYC*-translocations by either BHFR1, EBNA1, or suppression of pro-apoptotic BIM protein by LMP1, thereby helping in clonal evolution [25, 26]. A study from Malawi demonstrated strong association between high EBV antibody titers and endemic BL [27]. For more details on the role of EBV in the pathogenesis of B-cell lymphomas, please refer to Chap. 4.

Table 2.1 Immunodeficiency and NHL

Immunodeficiency	NHL
Primary	
Ataxia telangiectasia	BL, DLBCL, T cell LL
Wiskott-Aldrich syndrome	DLBCL
Severe Combined Immunodeficiency (SCID)	EBV-associated lesions
X-linked lymphoproliferative disorder	B-cell lymphoproliferative disorder
Autoimmune lymphoproliferative disorder	DLBCL, BL
Common variable immune deficiency (CVID)	DLBCL, EBV-associated lesions
X-linked hyperimmunoglobulin syndromes	DLBCL
Acquired	
Acquired immune deficiency syndrome (AIDS)	Kaposi sarcoma, BL
Post-organ/bone marrow transplant	DLBCL, BL

- *Malaria*
  There is an overlap in the geographical maps of endemic malaria and endemic BL. Epidemiological studies have shown that patients with high titers of EBV and malaria antibodies had the highest incidence of endemic BL [27, 28]. Malaria cooperates with EBV by modulating T-cell response in the pathogenesis of BL. Falciparum malaria causes EBV reactivation via cysteine-rich interdomain 1a of falciparum erythrocyte membrane protein. In addition, *Plasmodium falciparum* has a ligand for TLR-9 which is known to induce cytidine-deaminase in B cells and its overexpression can induce *MYC*-translocations. The translocated B cells would routinely be cleared by apoptosis which is in turn hampered by EBV [29, 30].

Unknown or Inconsistent Evidence

Radiation, arboviruses, schistosome parasites and as also some plants (*Euphorbiae tirucalli* and *Jatropha curcas*) have been suggested as possible causative factors/cofactors for NHL/endemic BL, although evidence is sparse and contrasting in some cases [14].

Mortality and Survival

Therapy for childhood NHL has been one of the big success stories in pediatric oncology with a 74% decrease in mortality rates from 1975 to 2010 and an annual percentage change exceeding 4% in the USA. Non-Hodgkin's lymphoma accounts for 3% and 7% of all cancer-related mortality in children and adolescents, respectively [31].

The survival of children and adolescents with NHL has doubled over the last 3–4 decades from around 45% to nearly 90% in the USA [4]. Similarly, in Europe, the outcome of children and adolescents with NHL is 83% and 78% (excluding BL), respectively [32].

However, as with most cancers there is a marked difference in the outcome of NHL between HICs and LMICs. It is difficult to have a true estimate of incidence and mortality rates due to the absence of robust population-based national cancer registries in LMICs, despite the fact that these countries account for more than 80% of children with cancer worldwide. In Malawi and Uganda (countries having the maximum incidence of NHL) the survival rates range from

20 to 50%. It is estimated that LMICs lag behind in survival rates by 20–30 years compared to the HICs [33–36].

As shown by the NHL-Berlin-Frankfurt-Münster (BFM) Study Group, age, gender, and biology also had an impact on the survival of children and adolescents with NHL. As compared to the average overall survival of NHL patients, PMBCL histology, adolescent girls with T-cell lymphoblastic lymphoma and DLBCL as well as younger children (<4 years) with ALCL and precursor B-cell precursor lymphoblastic lymphoma had inferior survival rates. In contrast, male patients with T-cell lymphoblastic lymphoma and DLBCL had superior survival rates [37].

Conclusions

Childhood NHLs are a heterogeneous group of diseases having a unique "epidemiological history" mainly attributable to BL and Denis Burkitt's legacy. Burkitt's lymphoma was one of the first tumors to be epidemiologically mapped and the first human tumor to be linked with a virus. In addition, BL was one of the first malignancies to be associated with a chromosomal translocation and the first lymphoma to be associated with HIV infections [14]. Since its first description in Africa in the 1960s, we have come a long way in understanding the biology and the therapy of childhood BL with survival rates exceeding 90% in the more developed countries. However, the irony remains that the place where the tumor was first described is still struggling to improve the survival from 1970, when the outcome for pediatric BL in Africa was as good as anywhere in the world.

Approximately 10% of pediatric NHL occurs in more developed countries and even if we further improve survival and cure all patients in these countries we would salvage only another 200 patients accounting for 1–2% of global numbers. However, if we could improve survival by even 15% in LMICs which account for nearly 90% of burden of childhood NHL we could save another 2500 children which is more than the total number of patients diagnosed in HICs.

References

1. Stuart BW, Wild CP. World Cancer report, vol. 2014. Lyon: IARC Press; 2014. p. 69–73.
2. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2018. CA Cancer J Clin. 2018;68(1):7–30.
3. Steliarova-Foucher E, Colombet M, Ries LAG, Moreno F, Dolya A, Bray F, et al. International incidence of childhood cancer, 2001–10: a population-based registry study. Lancet Oncol. 2017;18(6):719–31.
4. Howlader N, Noone AM, Krapcho M, Miller D, Bishop K, Kosary CL, Yu M, Ruhl J, Tatalovich Z, Mariotto A, Lewis DR, Chen HS, Feuer EJ, Cronin KA, editors. SEER cancer statistics review, 1975–2014. Bethesda: National Cancer Institute; 2017, https://seer.cancer.gov/csr/1975_2014/, based on November 2016 SEER data submission, posted to the SEER web site [Internet].
5. Ward E, DeSantis C, Robbins A, Kohler B, Jemal A. Childhood and adolescent cancer statistics, 2014. CA Cancer J Clin. 2014;64(2):83–103.
6. Murphy S. Classification, staging and end results of treatment of childhood non-Hodgkin's lymphoma: dissimilarities from lymphomas in adults. Semin Oncol. 1980;7:332–8.
7. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, Advani R, Ghielmini M, Salles GA, Zelenetz AD, Jaffe ES. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127:2375–90.
8. Armstrong GT, Liu Q, Yasui Y, et al. Late mortality among 5-year survivors of childhood cancer: a summary from the Childhood Cancer Survivor Study. J Clin Oncol. 2009;27:2328–38.
9. Percy CL, Smith M, Linet M, et al. Lymphomas and reticuloendothelial neoplasms. In: Ries LA, Smith MA, Gurney JG, et al., editors. Cancer incidence and survival among children and adolescents: United States SEER Program 1975–1995, NIH Pub. No. 99-4649. Bethesda: National Cancer Institute, SEER Program; 1999. p. 35–50.
10. Linet MS, Brown LM, Mbulaiteye SM, Check D, Ostroumova E, Landgren A, et al. International long-term trends and recent patterns in the incidence of leukemias and lymphomas among children and adolescents ages 0-19 years. Int J Cancer. 2016;138(8):1862–74.
11. Rodriguez-Galindo C, Friedrich P, Alcasabas P, Antillon F, Banavali S, Castillo L, et al. Toward the cure of all children with cancer through collaborative efforts: pediatric oncology as a global challenge. J Clin Oncol. 2015;33(27):3065–73.
12. Burkhardt B, Oschlies I, Klapper W, Zimmermann M, Woessmann W, Meinhardt A, et al. Non-Hodgkin's lymphoma in adolescents: experiences in 378 adolescent NHL patients treated according to pediatric NHL-BFM protocols. Leukemia. 2011;25(1):153–60.
13. Orem J, Mbidde EK, Lambert B, de Sanjose S, Weiderpass E. Burkitt's lymphoma in Africa, a review of the epidemiology and etiology. Afr Health Sci. 2007;7:166–75.
14. Molyneux EM, Rochford R, Griffin B, Newton R, Jackson G, Menon G, et al. Burkitt's lymphoma. Lancet. 2012;379(9822):1234–44.
15. Mbulaiteye SM, Biggar RJ, Bhatia K, Linet MS, Devesa SS. Sporadic childhood Burkitt lymphoma incidence in the United States during 1992-2005. Pediatr Blood Cancer. 2009;53(3):366–70.
16. Guech-Ongey M, Simard E, Anderson WF, et al. AIDS-related Burkitt lymphoma in the United States: what do age and CD4 lymphocyte patterns tell us about etiology and/or biology? Blood. 2010;116:5600–4.
17. Stiller CA, Parkin DM. International variations in the incidence of childhood lymphomas. Paediatr Perinat Epidemiol. 1990;4:303–24.
18. Izarzugaza MI, Steliarova-Foucher E, Martos MC, Zivkovic S. Non-Hodgkin's lymphoma incidence and survival in European children and adolescents (1978-1997): report from the Automated Childhood Cancer Information System project. Eur J Cancer. 2006;42(13):2050–63.
19. Yanik EL, Shiels MS, Smith JM, Clarke CA, Lynch CF, Kahn AR, et al. Contribution of solid organ transplant recipients to the pediatric non-hodgkin lymphoma burden in the United States. Cancer. 2017;123(23):4663–71.
20. Serraino D, Pezzotti P, Dorrucci M, et al. Cancer incidence in a cohort of human immunodeficiency virus seroconverters: HIV Italian Seroconversion Study Group. Cancer. 1997;79:1004–8.
21. Newton r ZJ, Beral V, et al. A case-control study of human immunodeficiency virus infection and cancer in adults and children residing in Kampala, Uganda. Int J Cancer. 2001;92:622–7.
22. Granovsky MO, Mueller B, Nicholson HS, et al. Cancer in human immunodeficiency virus-infected children: a case series from the Children's Cancer Group and the National Cancer Institute. J Clin Oncol. 1998;16:1729–35.
23. Caselli D, Klersy C, de Martino M, et al. Human immunodeficiency virus-related cancer in children: incidence and treatment outcome: report of the Italian register. J Clin Oncol. 2000;18:3854–61.

24. Landmann E, Oschlies I, Zimmermann M, et al. Secondary non-Hodgkin lymphoma (NHL) in children and adolescents after childhood cancer other than NHL. Br J Haematol. 2008;143(3):387–94.
25. Rowe M, Gemma K, Bell AI, Rickinson AB. Burkitt's lymphoma: the Rosetta Stone deciphering Epstein-Barr virus biology. Semin Cancer Biol. 2009;19:377–88.
26. Paschos K, Smith P, Anderton E, Middeldorp JM, White RE, Allday MJ. Epstein-barr virus latency in B cells leads to epigenetic repression and CpG methylation of the tumour suppressor gene Bim. PLoS Pathog. 2009;5:e1000492.
27. Aguilar R, Casabonne D, O'Callaghan-Gordo C, Vidal M, Campo JJ, Mutalima N, et al. Assessment of the combined effect of Epstein-Barr virus and Plasmodium falciparum infections on endemic Burkitt lymphoma using a multiplex serological approach. Front Immunol. 2017;8:1284.
28. Chene A, Donati D, Orem J, et al. Endemic Burkitt's lymphoma as a polymicrobial disease: new insights on the interaction between Plasmodium falciparum and Epstein-Barr virus. Semin Cancer Biol. 2009;19:411–20.
29. Parroche P, Lauw F, Goutagny N, et al. Malaria hemozoin is immunologically inert but radically enhances innate responses by presenting malaria DNA to toll-like receptor 9. Proc Natl Acad Sci U S A. 2007;104:1919–24.
30. Donati D, Zhang L, Chene A, et al. Identification of a polyclonal B-cell activator in Plasmodium falciparum. Infect Immun. 2004;72:5412–8.
31. Smith MA, Altekruse SF, Adamson PC, Reaman GH, Seibel NL. Declining childhood and adolescent cancer mortality. Cancer. 2014;120(16):2497–506.
32. Trama A, Botta L, Foschi R, Ferrari A, Stiller C, Desandes E, et al. Survival of European adolescents and young adults diagnosed with cancer in 2000–07: population-based data from EUROCARE-5. Lancet Oncol. 2016;17(7):896–906.
33. Redaniel MT, Laudico A, Mirasol-Lumague MR, Alcasabas AP, Pulte D, Brenner H. Geographic and ethnic differences in childhood leukaemia and lymphoma survival: comparisons of Philippine residents, Asian Americans and Caucasians in the United States. Br J Cancer. 2010;103(1):149–54.
34. Stanley C, Heimlich JB, El-Mallawany NK, Mtete I, Butia M, Kaimila B, Itimu S, Chikasema M, Chikasema F, Kampani C, Mzumara S, Montgomery ND, Fedoriw Y, Kazembe PND, Krysiak R, Liomba NG, Gopal S. Survival among CHOP-treated children with endemic Burkitt lymphoma (eBL) in Malawi. Br J Haematol. 2015;171:81, abstract 167.
35. Geriga F, Mutyaba I, Kambugu J, McGoldrick S, Casper C, Orem J. Presentation and treatment outcomes of children with Burkitt lymphoma at Uganda Cancer Institute. Br J Haematol. 2015;171:43, abstract 75.
36. Mutai M, Mtete I, Mehta P, Gopal S, Liomba G, Fedoriw Y, Krysiak R, Kazembe P, El-Mallawany NK. Clinical spectrum of lymphoma in children and adolescents in Central Malawi. Br J Haematol. 2015;171:78, abstract 159.
37. Burkhardt B, Zimmermann M, Oschlies I, Niggli F, Mann G, Parwaresch R, et al. The impact of age and gender on biology, clinical features and treatment outcome of non-Hodgkin lymphoma in childhood and adolescence. Br J Haematol. 2005;131(1):39–49.

Diagnosis and Classification

3

Thierry Jo Molina

Introduction

Non-Hodgkin lymphomas (NHL) are typical tumors for the pediatric pathologist to be familiar with as they represent around 7–10% of pediatric malignancies, the fourth most common one in children, being more frequent between 15 and 19 years of age [1–3]. These lymphomas could arise in any tissue, nodal or extranodal, and the pathologist should be prepared to diagnose its histological type according to the updated revised 2016 WHO Classification of Tumors of Hematopoietic and Lymphoid Tissues [4]. When dealing with a suspicious diagnosis of NHL, the pathologist should follow four major rules:

1. Eliminate the most frequent entities that might have a peculiar clinical or histological presentation before suggesting a rare subtype
2. Have the knowledge of entities that have not been described or are very rare in this age group, and consider therefore a reactive process mimicking NHL before assessing NHL
3. Consider a potential immune deficiency with an expansion of B or T cells before assessing a diagnosis of NHL, and avoid making a diagnosis without the full clinical information. It is therefore crucial before any diagnosis to rule out concurrent Cancer Predisposition Syndromes [5], Primary or Secondary Immunodeficiencies or Genetic diseases.
4. Ensure the use of ancillary techniques to confirm the diagnosis.

In this chapter, we will present the new updated 2016 revision of the WHO, discuss the importance of sampling (cytology, needle biopsy, open wedge biopsy) for diagnosis, and present the different techniques considered today as being instrumental for an optimal diagnosis of childhood and adolescence NHL.

The Revised 2016 Classification of Lymphoid Neoplasms

This revised 4th edition of the WHO classification includes some changes linked to a better understanding of some entities with a common agreement consensus between members of the European Association for Haematopathology, the Society for Hematopathology (US), as well as an international clinical advisory committee. The importance of an international nomenclature is crucial when comparing NHL arising in children within different countries, and the pathologist classifying the tumor should stick to these entities. There is no space for a pediatric classification of NHL. Although some entities are predominantly described in children, they can arise rarely among patients >18 years of age. However, a few entities are different (clinically and molecularly) from the classical adult type, giving rise in the WHO nomenclature to a "pediatric-type" to underline this specificity. On the other hand, some entities described in adults (mantle cell lymphoma, angioimmunoblastic T-cell lymphoma) have not been clearly described in children. The WHO classification underlines very strongly the importance of multiparametric approach to classify a tumor and this is even more important considering childhood NHL. The clinical presentation, the age, the morphologic pattern, the phenotype by immunohistochemistry and if possible flow cytometry, the genetic profile (translocation, gain or loss, mutations, expression profiling, etc.), and presence of blood tumor cells are all important factors to consider in the final diagnosis. This multiparametric concept should lead to collaborative efforts between pathology, hematopathology, and molecular and cytogenetics laboratories for an optimal

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diagnosis of childhood and adolescence NHL. At least, all these data should be gathered in a unified and integrated consolidated report, with the pathology report. The pathologist facing a suspicion of childhood and adolescence NHL should be familiar with the whole NHL classification. This classification differentiates precursor cell lymphoid neoplasm (Table 3.1) (either B, T, or NK lineage) from mature B-cell (Table 3.2) and T-cell neoplasms (Table 3.3) and immune deficiency-related lymphoproliferative disorders (Table 3.4). The revised WHO classification differentiates also provisional entities that need more data and studies to consider them as well as distinct entities. In addition, we believe, considering children NHL that it is crucial to differentiate entities well described in children from the ones classically not reported in children or rarely described and we decided to mention this differentiation in the tables (Tables 3.1, 3.2, 3.3, and 3.4). To us, this distinction is important to avoid a misdiagnosis when facing a morphologic lesion suspicious of a lymphoma entity non-described in children. In such cases, it is important to eliminate a reactive lymphoid process that may mimic a rare or non-described NHL subtype. For example, primary immune deficiencies such as children with *RAG-1* hypomorphic deficiency might have huge polyclonal expansion of T cells in the spleen or bone marrow mimicking mature peripheral T-cell lymphoma (Fig. 3.1). When dealing with a diagnosis of NHL in children, few histological subtypes cover more than 80–90% of the cases, and these are mainly aggressive lymphoid neoplasms: lymphoblastic B or T, Burkitt and diffuse large B-cell lymphomas, anaplastic large cell lymphomas, and post-transplant or primary immune deficiency-related lymphoproliferations. However, some of these classical pediatric subtypes might present with an unusual histological or clinical pattern, in unusual sites, that can be challenging for diagnosis. This is the case, for example, of a primary central nervous system ALK+ anaplastic large cell lymphoma, presenting with a small cell variant morphology (Fig. 3.2). Nevertheless, other rare subtypes are classically described among children and might be difficult to diagnose requiring full clinical information and ancillary techniques such as double staining combining in situ hybridization and immunophenotyping or molecular techniques.

Table 3.1 2016 WHO classification of non-Hodgkin lymphoma, precursor lymphoid neoplasms

B-lymphoblastic lymphoma/leukemia, NOS
B-lymphoblastic leukemia/lymphoma, NOS with recurrent genetic abnormalities
T-lymphoblastic leukemia/lymphoma
NK-lymphoblastic leukemia/lymphoma[a]

Bold: well described NHL in the pediatric population
[a]Provisional entity according to WHO

Table 3.2 2016 WHO classification of non-Hodgkin lymphoma, mature B-cell lymphomas

Chronic lymphocytic leukemia/small lymphocytic lymphoma, Monoclonal B-cell lymphocytosis, B-cell prolymphocytic leukemia, Splenic marginal zone lymphoma, Hairy cell leukemia, Splenic B-cell lymphoma/leukemia, unclassifiable[a]*, Splenic diffuse red pulp small B-cell lymphoma*[a]*, Hairy cell leukemia-variant*[a]*, Lymphoplasmacytic lymphoma (Waldenstrom macroglobulinemia), Mu heavy chain disease, Gamma heavy chain disease*
Alpha heavy chain disease
Extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue (MALT lymphoma)
Nodal marginal zone lymphoma**, Pediatric nodal marginal zone lymphoma**[a]
Follicular lymphoma, In situ follicular neoplasia, *Duodenal-type follicular lymphoma*
Pediatric-type follicular lymphoma
Large B-cell lymphoma with IRF4 rearrangement[a]
Primary cutaneous follicle center lymphoma, Mantle cell lymphoma, In situ *mantle cell neoplasia*
Diffuse large B-cell lymphoma (DLBCL), NOS Germinal center B-cell type
Diffuse large B-cell lymphoma (DLBCL), NOS Activated B-cell type, *T-cell/histiocyte-rich large B-cell lymphoma*, Primary DLBCL of the CNS, *Primary cutaneous DLBCL, leg type*, EBV-positive DLBCL, NOS, EBV+ Mucocutaneous ulcer[a]*, DLBCL associated with chronic inflammation, Intravascular large B-cell lymphoma, Primary effusion lymphoma*, HHV8-positive DLBCL, NOS
Lymphomatoid granulomatosis
Primary mediastinal (thymic) large B-cell lymphoma
ALK-positive large B-cell lymphoma, Plasmablastic lymphoma
Burkitt lymphoma
Burkitt-like lymphoma with 11q aberration[a]
High-grade B-cell lymphoma, with MYC and BCL2 and/or BCL6 rearrangements, High-grade B-cell lymphoma, NOS
B-cell lymphoma, unclassifiable, with features intermediate between DLBCL and classical Hodgkin lymphoma

Bold: well described NHL in the pediatric population; Italic: classically not described below 18 years; Non-Bold Non-Italic: rarely described in children
[a]Provisional entity according to WHO

This is the case, for example, of an optimal diagnosis of chronic active Epstein–Barr virus (EBV) infection of T and NK cell type, systemic form, recently individualized in the revised WHO classification in which it is crucial to demonstrate that EBV-positive cells are of the T lineage (Fig. 3.3). It might be important at a national and/or international level such as the EICNHL (European Intergroup for Children NHL, with Japan and Hong Kong) and the North American Children's Oncology Group (COG) to have validated database of these rare NHL subtypes requiring both the analysis of those cases by a panel of pathologists (with detailed phenotypical and molecular analyses of the case) and very precise clinical information such as clinical Case Report Form (CRF) created by a panel of clinicians interested in rare subtypes. For example, the differentiation of chronic active

Table 3.3 2016 WHO classification of non-Hodgkin lymphoma, mature T and NK cell lymphomas

T-cell prolymphocytic leukemia, T-cell large granular lymphocytic leukemia, *Chronic lymphoproliferative disorder of NK cells*[a], Aggressive NK cell leukemia
Systemic EBV+ T-cell Lymphoma of childhood
Chronic active EBV infection of T- and NK-cell type, systemic form
Hydroa vacciniforme-like lymphoproliferative disorder
Extranodal NK/T-cell lymphoma, nasal type
Adult T-cell leukemia/lymphoma, Enteropathy-associated T-cell lymphoma, Monomorphic epitheliotropic intestinal T-cell lymphoma, Indolent T-cell lymphoproliferative disorder of the GI tract[a]
Hepatosplenic T-cell lymphoma
Subcutaneous panniculitis-like T-cell lymphoma
Mycosis fungoides, *Sezary syndrome*
Primary cutaneous CD30-positive T-cell lymphoproliferative disorders
Lymphomatoid papulosis
Primary cutaneous anaplastic large cell lymphoma, Primary cutaneous gamma-delta T-cell lymphoma, Primary cutaneous CD8-positive aggressive epidermotropic cytotoxic T-cell lymphoma[a], *Primary cutaneous acral CD8-positive T-cell lymphoma*[a], Primary cutaneous CD4-positive small/medium T-cell lymphoproliferative disorder[a]
Peripheral T-cell lymphoma, NOS
Angioimmunoblastic T-cell lymphoma, Follicular T-cell lymphoma, Nodal peripheral T-cell lymphoma with TFH phenotype
Anaplastic large cell lymphoma, ALK-positive
Anaplastic large cell lymphoma, ALK-negative
Breast implant-associated anaplastic large cell lymphoma[a]

Bold: well-described NHL in the pediatric population; Italic: classically not described below 18 years; Non-Bold Non-Italic: rarely described in children

[a]Provisional entity according to WHO

Table 3.4 2016 WHO classification of non-Hodgkin lymphoma, post-transplant lymphoproliferative disorders (PTLD)

Plasmacytic hyperplasia PTLD
Infectious mononucleosis PTLD
Florid follicular hyperplasia PTLD
Polymorphic PTLD
Monomorphic PTLD (B- and T/NK-cell types)
Classical Hodgkin lymphoma PTLD

Bold: well described NHL in the pediatric population

EBV infection of T-cell type, systemic form, from the systemic EBV-positive T-cell lymphoma of childhood is very complex and could be better validated in the future by an international pathology and clinical study. Rather than using ICD-9 or ICD-10 codes for the creation of the database [6], it seems important to follow the ICD-O codes related to the WHO 2016 classification and to review old cases taking into account this new classification.

Cytology, Needle Biopsy, or Open Wedge Biopsy for Diagnosis?

A dogma for NHL diagnosis is that we need histopathology and therefore a tissue biopsy for the diagnosis. However, in children, in very few selected cases, with a typical clinical presentation, cytology with phenotype by flow cytometry and genetic profiling allows an adapted clinical management. This is the case for Burkitt lymphoma when the clinical presentation, the flow phenotype, and the FISH for MYC translocation are all typical and for lymphoblastic B-cell or T-cell lymphoma when the leukemic phase or marrow involvement (too low to be considered as acute lymphoblastic leukemia) is present at diagnosis and considered sufficient for the diagnostic laboratory for cytology, immunophenotyping, and molecular evaluation. What is really important in this cytological approach is that any unusual feature (clinical, cytological, phenotypical, or molecular) should lead to a histological evaluation.

The use of core needle biopsy for histological diagnosis of pediatric tumors is increasingly performed as it is the case in adult tumors, although at a lower frequency. A recent prospective study for diagnosis on non-nephroblastoma solid intraabdominal tumors has clearly shown the significant advantage of open wedge biopsy over needle biopsy for diagnosis [7]. However, a single-center retrospective study of 396 image-guided percutaneaous needle biopsies performed in children for pediatric tumors showed a diagnostic accuracy of 91%, underlining however the importance of 4 passes, and of the ability to freeze at least one core for molecular diagnostic tools [8]. Concerning lymphoma diagnosis suspicion, the ideal management of tissue sample requires touch imprints (Cytology, FISH), flow cytometry, freezing tissue for molecular techniques, and of course large amount of tissue for histopathological and immunohistochemical studies. Since lymphoma can arise as a primary tumor in any tissue (i.e., bone, skin), it might be useful to stain one imprint and to send a fresh sample for flow cytometry when lymphoma cannot be ruled out (i.e., small cell round tumor). These requirements highlight the importance of large amount of tissue samples for diagnosis and explain why an open wedge biopsy is preferred over a needle biopsy by most pathologists. Nevertheless, a diagnosis of NHL can be made on most needle biopsies if all the morphological and immunohistochemical criteria of a well-defined entity are present, but the pathologist should not make a definite diagnosis in all other cases. In addition, it is very difficult to exclude a diagnosis of lymphoma on a needle biopsy if the biopsied tissue is normal (i.e., normal architecture of lymph node and normal distribution of B and T cells) and if clinical suspicion is strong. Therefore, core needle biopsies performed by an interventional radiologist could be a

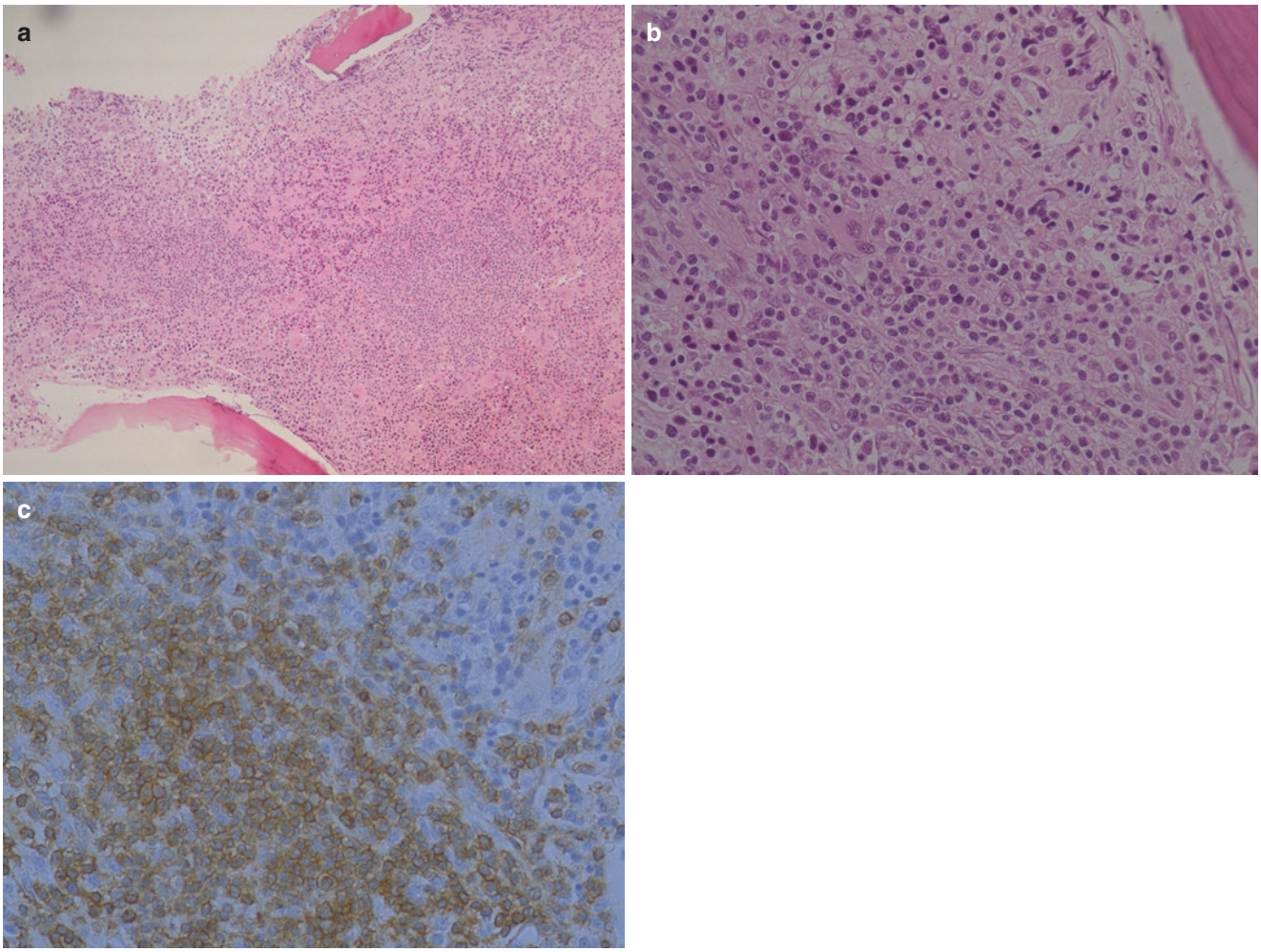

Fig. 3.1 Polyclonal Gamma delta T-cell expansion in the bone marrow of a 13-year-old *RAG1* hypomorphic patient with pancytopenia mimicking lymphoma. (**a**) at low magnification, infiltrate of small lymphoid cells destroying the normal architecture of the bone marrow (H &E stain). (**b**) at higher magnification this dense infiltrate is made of small lymphoid cells. (**c**) Most of these lymphoid cells are CD3-positive T cells

reasonable first diagnostic procedure in close collaboration with the pathologists and clinicians; however parents need to be informed that in a minority of cases, an open wedge biopsy might be needed for an optimal diagnosis. Open wedge biopsy is often the rule when dealing with chronic superficial adenopathies suspicious of a rare indolent pediatric lymphoma subtype (such as follicular lymphoma pediatric type) or in the context of possible primary immunodeficiency disease associated with a lymphoproliferation.

Ancillary Techniques

When dealing with the possible diagnosis of lymphoma, in addition to the classical paraffin-embedded analysis of histopathology, immunohistochemistry, and immunophenotyping, other techniques are currently required to classify the NHL according to 2016 WHO Classification. The immunohistochemical profile performed should stick to the phenotype described for each entity. When dealing with an undifferentiated small−/medium-sized blue cell tumors, in addition to the classical panel for non-lymphoid tumors, a first screen comprising CD79a, CD3, TdT, and CD30 will help to diagnose the most frequent lymphoid neoplasms. EBER in situ hybridization to detect the presence of EBV in tumor cells is widely used in the case of suspicious immune deficiency-related lymphoproliferations or EBV-associated lymphomas. The setup by the pathology laboratory of colorimetric double stain CD3/EBER, CD79a/EBER, CD20/EBER, and CD8/EBER to demonstrate the lymphoid lineage affected by EBV is crucial, as there is often B- and T-cell expansion in EBV-associated lymphoproliferations. Clonality tests by PCR to look for *IgH*, *Kappa*, *Lambda*, *TCR gamma*, and *TCR beta* rearrangements are very useful in difficult cases of B- or T-cell lymphoma or when discussing lymphoid expansions mimicking lymphoma; in most

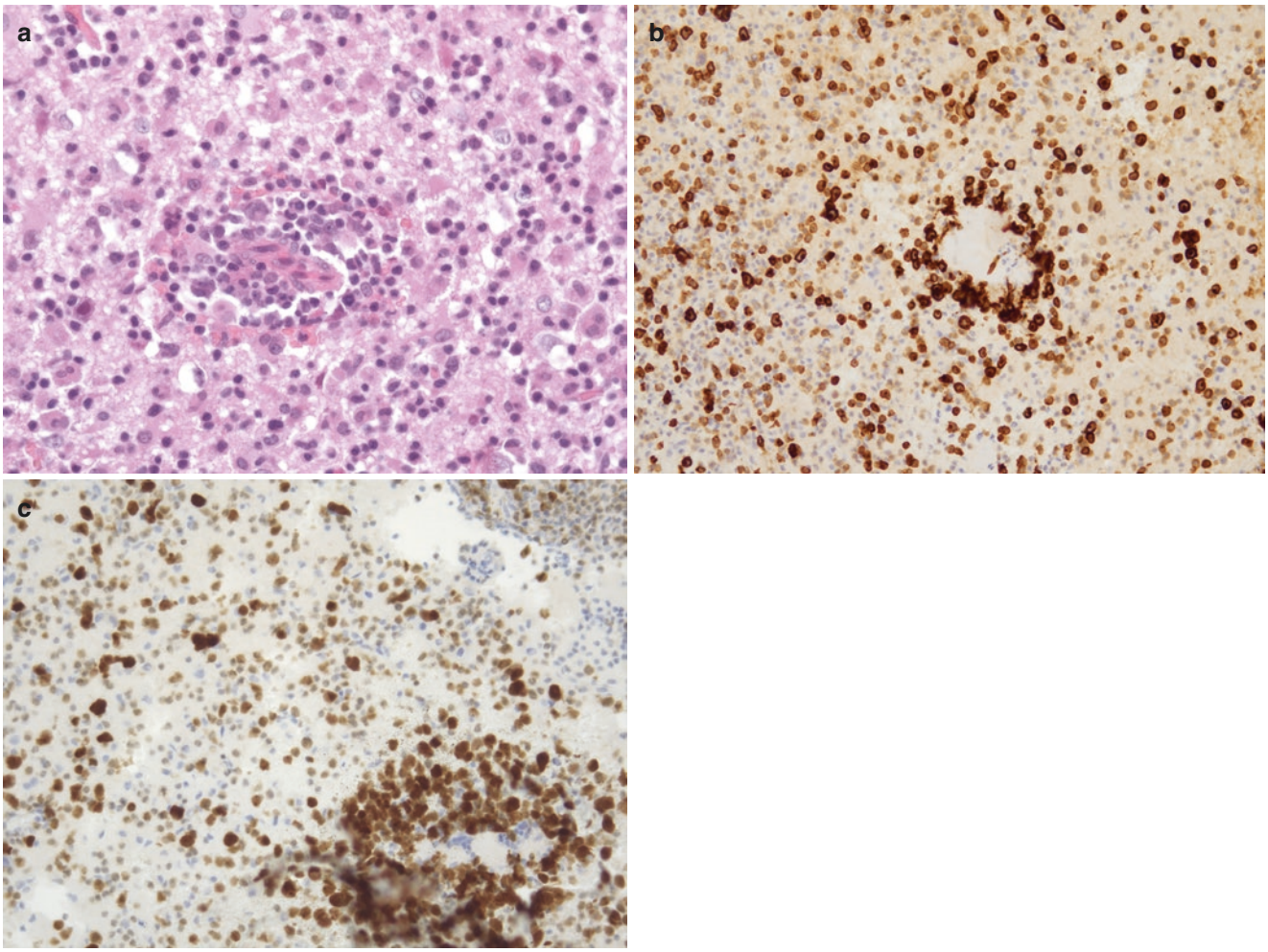

Fig. 3.2 Anaplastic large cell lymphoma, small cell variant occurring at presentation in the central nervous system (CNS) in a 5-year-old child. (**a**) Small lymphoid cell infiltration in a perivascular predominant topography of the central nervous associated with an interstitial infiltrate. (**b**) Strong expression of CD30 by small lymphoid cells. (**c**) Nuclear expression of ALK by tumor cells

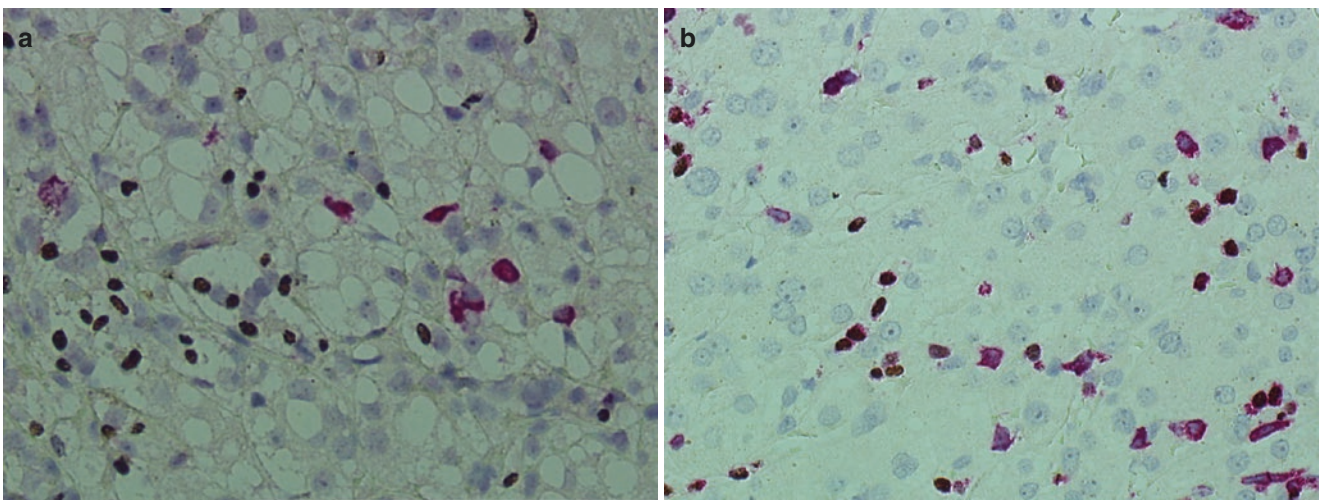

Fig. 3.3 Chronic active EBV infection of the T-cell type, Systemic, liver biopsy in a 15-year-old child with secondary HLH (hemophagocytosis lymphohistiocytosis). (**a**) Presence of a few CD79a-positive B-cell and plasma cells (red) associated with numerous EBER-positive CD79a-negative cells (black nuclear stain), double staining CD79a/EBER. (**b**) the EBER-positive cells (black nuclei) have all CD3-positive cytoplasmic staining (red) whereas some CD3-positive cells are not EBER-positive, double staining EBER/CD3

cases, formalin fixation allows a good interpretation of B- or T-cell receptor repertoire, although DNA extracted from frozen samples might be important to retrieve in some cases. However, the use of these tests should be very cautious as B-cell or T-cell clones can be present in reactive states and false negatives can occur in true lymphomas. A complex question arising these days concerns the use of karyotype analysis for lymphoma diagnosis. This is a costly and labor-intensive technique with the setup of overnight cultures. The difficulty in predicting clinically a potential lymphoma diagnosis in the approach of a pediatric tumor and the knowledge of the major cytogenetic abnormalities arising in different lymphoma subtypes (i.e., translocations) that can be easily diagnosed by interphase FISH have convinced numerous hematological teams to stop the prescription of karyotype as a systematic first approach (with a few exceptions, for example, when clinical presentation is typical of Burkitt lymphoma) and rather to develop alternative molecular techniques. In this respect, the replacement of karyotype by comparative genomic hybridization (CGH) array to evaluate amplifications, deletions, and the complexity of karyotype combined with interphase FISH to diagnose a recurrent translocation is becoming a very efficient approach. The importance of these techniques in childhood and adolescence NHL is underlined by the fact that two new entities, although provisional, are defined by the presence of a translocation detected by FISH such as large B-cell lymphoma (follicular and/or diffuse) with *IRF4* rearrangement, and by proximal gains and telomeric losses detected by CGH array such as Burkitt-like lymphoma with 11q aberration. The use of flow cytometry from fresh cell suspension from a biopsy is very useful for the diagnosis each time there is a suspicion of lymphoma (lymphoblastic, Burkitt, etc.) as the panel of antigens studied is much larger than immunohistochemistry. Nevertheless, it is highly recommended that the diagnosis should never be performed by flow cytometry alone and be integrated with histopathological diagnosis. For example, a monotypic CD19+ CD10+ BCL2 negative Ki67+ 90%, lymphoid population may correspond to the phenotype of a Burkitt lymphoma but also to the phenotype of a follicular lymphoma pediatric-type. In addition, the study of a panel of mutations by next-generation sequencing (NGS), a targeted expression profiling technique from formalin-fixed paraffin-embedded tissue, could help to better define an entity in difficult cases such as in the differential diagnosis between Burkitt lymphoma and DLBCL [9] and/or between PMBL and DLBCL NOS [10]. Cell-free DNA analysis at diagnosis, through blood liquid biopsy, is a promising technique that has been shown in adult DLBCL to reflect with good sensitivity the main genomic abnormalities of NHL in the absence of a leukemic phase and to allow disease response monitoring through clonal evolution analysis in association with FDG-PET scan data [11].

Overall, all these tools should now be implemented to allow an optimal diagnosis of childhood and adolescence NHL. It underlines the importance of a multidisciplinary laboratory approach for the diagnosis of lymphoma gathering pathologists, biological hematologists, and geneticists ideally working together as a diagnostic unit for the diagnosis of lymphoma.

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References

1. Minard-Colin V, Brugières L, Reiter A, Cairo MS, Gross TG, Woessmann W, Burkhardt B, Sandlund JT, Williams D, Pillon M, Horibe K, Auperin A, Le Deley MC, Zimmerman M, Perkins SL, Raphael M, Lamant L, Klapper W, Mussolin L, Poirel HA, Macintyre E, Damm-Welk C, Rosolen A, Patte C. Non-Hodgkin lymphoma in children and adolescents: progress through effective collaboration, current knowledge, and challenges ahead. J Clin Oncol. 2015;33(27):2963–74.
2. Bollard CM, Lim MS, Gross TG, COG Non-Hodgkin Lymphoma Committee. Children's Oncology Group's 2013 blueprint for research: non-Hodgkin lymphoma. Pediatr Blood Cancer. 2013;60(6):979–84.
3. Sandlund JT, Martin MG. Non-Hodgkin lymphoma across the pediatric and adolescent and young adult age spectrum. Hematology Am Soc Hematol Educ Program. 2016;2016(1):589–97.
4. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, Arber DA, Hasserjian RP, Le Beau MM, Orazi A, Siebert R. WHO classification of tumours of haematopoietic and lymphoid tissues, revised. 4th ed. Lyon: IARC; 2017.
5. Attarbaschi A, Carraro E, Abla O, Barzilai-Birenboim S, Bomken S, Brugieres L, Bubanska E, Burkhardt B, Chiang AK, Csoka M, Fedorova A, Jazbec J, Kabickova E, Krenova Z, Lazic J, Loeffen J, Mann G, Niggli F, Miakova N, Osumi T, Ronceray L, Uyttebroeck A, Williams D, Woessmann W, Wrobel G, Pillon M, European Intergroup for Childhood Non-Hodgkin Lymphoma (EICNHL) and the International Berlin-Frankfur t-Münster (i-BFM) Study Group. Non-Hodgkin lymphoma and pre-existing conditions: spectrum, clinical characteristics and outcome in 213 children and adolescents. Haematologica. 2016;101(12):1581–91.
6. Citrin R, Horowitz JP, Reilly AF, Li Y, Huang YS, Getz KD, Seif AE, Fisher BT, Aplenc R. Creation of a pediatric mature B-cell non-Hodgkin lymphoma cohort within the Pediatric Health Information System Database. PLoS One. 2017;12(10):e0186960.
7. Deeney S, Stewart C, Treece AL, Black JO, Lovell MA, Garrington T, Karrer F, Bruny J. Diagnostic utility of core needle biopsy versus open wedge biopsy for pediatric intraabdominal solid tumors: Results of a prospective clinical study. J Pediatr Surg. 2017;52(12):2042–6.
8. Blondiaux E, Laurent M, Audureau E, Boudjemaa S, Sileo C, Lenoir M, Dainese L, Garel C, Coulomb A, Ducou le Pointe H. Factors influencing the diagnostic yield and accuracy of image-guided percutaneous needle biopsy of pediatric tumors: single-center audit of a 26-year experience. Pediatr Radiol. 2016;46(3):372–82.

9. Rohde M, Bonn BR, Zimmermann M, Lange J, Möricke A, Klapper W, Oschlies I, Szczepanowski M, Nagel I, Schrappe M, MMML-MYC-SYS Project, ICGC MMML-Seq Project, Loeffler M, Siebert R, Reiter A, Burkhardt B. Relevance of ID3-TCF3-CCND3 pathway mutations in pediatric aggressive B-cell lymphoma treated according to the non-Hodgkin Lymphoma Berlin-Frankfurt-Münster protocols. Haematologica. 2017;102(6):1091–8.
10. Bobée V, Ruminy P, Marchand V, Viailly PJ, Abdel Sater A, Veresezan L, Drieux F, Bérard C, Bohers E, Mareschal S, Dubois S, Jais JP, Leroy K, Figeac M, Picquenot JM, Molina TJ, Salles G, Haioun C, Tilly H, Jardin F. Determination of molecular subtypes of diffuse large B-cell lymphoma using a reverse transcriptase multiplex ligation-dependent probe amplification classifier: a CALYM Study. J Mol Diagn. 2017;19(6):892–904.
11. Scherer F, Kurtz DM, Newman AM, Stehr H, Craig AF, Esfahani MS, Lovejoy AF, Chabon JJ, Klass DM, Liu CL, Zhou L, Glover C, Visser BC, Poultsides GA, Advani RH, Maeda LS, Gupta NK, Levy R, Ohgami RS, Kunder CA, Diehn M, Alizadeh AA. Distinct biological subtypes and patterns of genome evolution in lymphoma revealed by circulating tumor DNA. Sci Transl Med. 2016;8(364):364ra155.

Part II

Pathology and Molecular Biology

Pathogenesis of B-Cell Lymphoma

4

Rabea Wagener, Cristina López, and Reiner Siebert

Introduction

The pathogenesis of B-cell lymphomas is assumed to be a multifactorial and multistep process. Known contributing factors include germline predisposition, processes of physiologic B-cell development, environmental factors (e.g. viruses), microenvironmental stimuli, and somatic alterations [1]. Remarkably, these factors do interact on various cellular levels, e.g. germline predisposition to pediatric B-cell lymphoma might cause altered response to viral infection or repair of DNA damage [2, 3]. Moreover, different pathogenetic means can substitute for each other, e.g. essential pathways contributing to lymphomagenesis can be altered by germline or somatic mutations on DNA level or by transcriptional changes on RNA level; the latter may be associated by epigenetic changes [4] which in turn may or may not be induced by external stimuli like viruses [3].

In the following we will outline key principles and general mechanisms underlying the pathogenesis of B-cell non-Hodgkin lymphomas in children and adolescents as far as not reviewed elsewhere in this book (regarding germline predisposition see Chap. 7; for details on the distinct lymphoma entities, see Chaps. 11, 12, 13, 14, 15, 16, 17, and 18). The basis for the understanding of B-cell lymphomagenesis is the normal B-cell development and its underlying cellular and genetic mechanisms, as these physiological processes become hijacked during lymphoma initiation and progression. Accordingly, in the following first the physiologic B-cell development and differentiation are summarized, before we subsequently give an overview on the various mechanisms contributing to B-cell lymphomagenesis.

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Physiologic B-Cell Development

B-cells are key players in the physiological immune response including the humoral immune response as well as the immunological memory [5]. These functions are accomplished by the unique features of B-cells including antigen presentation, immune regulation, and provision of the cellular as well as humoral immune repertoire. The key function of B-cells is to produce immunoglobulins. Besides the signaling through these B-cell receptors, also mechanisms involved in determining immunoglobulin specificity, if mistaken, contribute to lymphomagenesis [6, 7]. Thus, in the following we first review the molecular processes underlying immunoglobulin determination, and then we will provide insights into the physiologic B-cell development.

Immunoglobulins and Immunoglobulin Genes

Immunoglobulin Structure

The immunoglobulins or antibodies are the effector molecules produced by the B-cells mediating the humoral immune response [8]. Furthermore, the membrane-bound form of the antibodies, the B-cell receptor, and its associated cofactors mediate intracellular signals important for B-cell development and differentiation. The gene loci encoding the B-cell receptor undergo complex rearrangements during B-cell development contributing to the antibody diversity [8]. Accordingly, every physiologic B-cell has a unique B-cell receptor harboring an individual specificity for a certain antigen. For a better understanding of the mechanisms leading to this diversity, the structure of the immunoglobulins is described in the following. Immunoglobulins are Y-shaped polypeptides consisting of two heavy and two light chains linked by disulfide bonds. The heavy chain consists of a variable (V_H) and three constant domains (C_H1-3), whereas the light chains are built of a variable (V_L) and one constant domain (C_L) [8]. The variable domains, composed of the vari-

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able domains of the light and heavy chains mediate the specific antigen binding. The constant region, which consists of the constant domains from heavy and light chains, interacts with the effector cells and molecules. The B-cell receptor carries, in contrast to the soluble antibody, a C-terminal polypeptide which anchors the receptor to the cell membrane [9, 10].

The immunoglobulins are encoded by a multi-gene family. Two light-chain types exist. On chromosome 2p11 maps the immunoglobulin κ (IGK) and on chromosome 22q11 the λ (IGL) gene locus. The heavy chain is encoded by a gene locus on chromosome 14q32. These three gene loci contain several coding and non-coding gene segments which are rearranged to form a functional immunoglobulin gene. The light-chain loci have multiple variable (V), joining (J), and constant (C) gene segments. The heavy chain locus harbors in addition diversity (D) gene segments as well as a series of C gene segments: Cμ, Cδ, Cγ, Cα, and Cε. These C gene segments encode the immunoglobulin isotypes: IgM, IgD, IgG, IgA, and IgE, respectively, which confer the effector functions of the respective antibodies [8].

Molecular Processes Remodeling Immunoglobulin Genes

Each B-cell harbors after complex, somatic rearrangements, which contribute to the antibody diversity, a unique immunoglobulin gene (Fig. 4.1). The first step in the generation of this diversity is the VDJ-gene rearrangement which takes place in precursor B-cells within the bone marrow. As outlined above, the immunoglobulin gene locus consists of multiple gene segments which need to be rearranged in order to give rise to a functional coding exon. First, in a random recombination process one of the D gene segments of the heavy chain locus is fused to one J gene segment, followed by a rearrangement of one V gene segment to the already fused DJ segment [11]. Lastly, the VDJ fusion is rearranged to the C gene segment. Due to the multiple gene segments of the heavy chain locus more than 10^4 different VDJ recombinations are possible, contributing to the combinatorial diversity of the antibody repertoire. After successful rearrangement, the heavy chain is expressed on the surface of the precursor B-cell with a surrogate light chain. In a next step, the light-chain locus is rearranged fusing one of the V gene segments to a J segment. The V(D)J recombination is mediated by a V(D)J recombinase complex, including the RAG1 and RAG2 proteins. Those proteins bind to the conserved recombination signal sequences adjacent to each gene segment. Upon binding, the double-stranded DNA is cut and a hairpin structure is built [12]. The hairpin can be removed in different ways leading either to the insertion of non-germline-encoded (N) nucleotides conferred by the terminal deoxynucleotidyl transferase (TdT) or the deletion of single nucleotides conferred by exonucleases at the recombination sites [13]. These sequence alterations confer the junctional diversity of the antibodies.

Other mechanisms contributing to the diversity of the antibodies take place after the B-cell has encountered its antigen primarily during the germinal center (GC) reaction. By somatic hypermutation (SHM) point mutations are introduced within the gene segments encoding the V region of the antibody [14]. This is conducted by the activation-induced deaminase (AID) which upon binding to single-stranded DNA deaminates cytosine to uracil [15, 16]. The binding of AID is heavily dependent on the expression of the respective gene segment. By the introduction of an uracil within the DNA sequence, either the mismatch repair or the base exci-

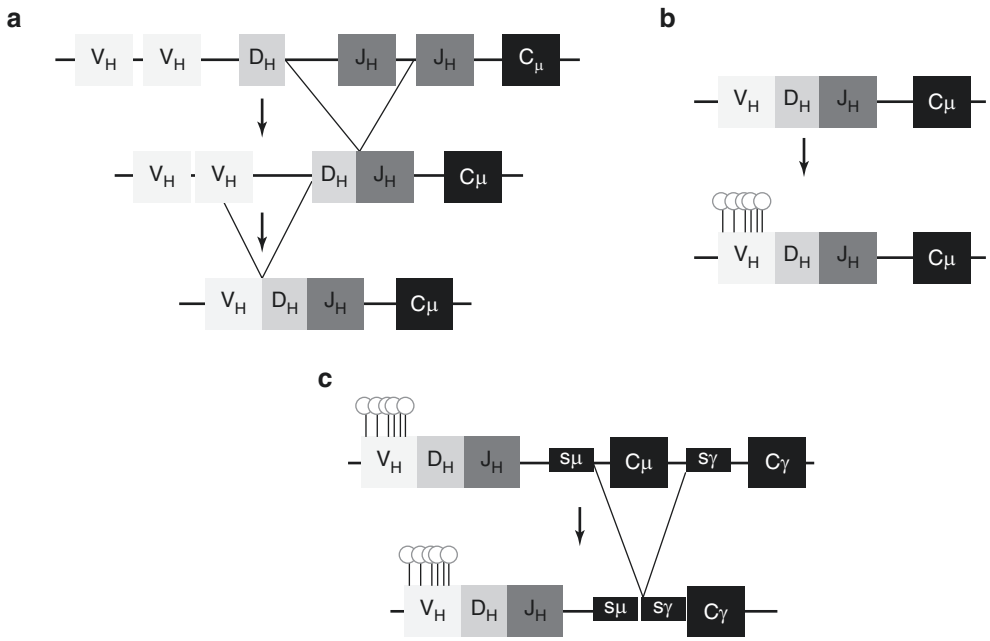

Fig. 4.1 Schematic overview on the molecular processes remodeling the immuglobulin genes, using as example IGH locus. (**a**) Process on the VDJ recombination. First, the D gene segment is assembled to one of the J gene segments. After, one of the V gene segments is associated to the DJ segment. (**b**) Somatic hypermutation process is activated in the germinal center, introducing mutations in the V region of the heavy and the light chain (designated by the lollipops). (**c**) The latest step in the Ig remodeling is the class switch recombination process, which takes place only on the heavy chain locus. Modified from [1]

sion repair mechanism pathways are activated which further introduce alterations within the DNA sequence [14]. Another mechanism taking place in the GC is the class switch recombination (CSR) [17]. By CSR the constant gene segment of the heavy chain locus is switched by an irreversible DNA recombination process including non-homologous DNA recombination [18]. This process is again conferred by the AID which is guided to specific switch regions located within the intron between the J_H and upstream of all C gene segments [19]. Hence, B-cells initially expressing IgM or IgD switch their immunoglobulin isotype in the majority to an IgG but also to an IgA or IgE. This leads on the one hand to a change in the B-cell receptor signaling competence but alters as well the effector function of the antibody.

During all the processes leading to the physiologic shaping of the IGs mistakes can occur which might result in fusion of part of the IG genes with other gene loci. If the latter contain oncogenes, these can be driven by the strong enhancers at the IG loci which physiologically ensure sufficient BCR/IG production in B-cells. Thus, as a consequence of such aberrant IG rearrangements, the oncogenes on the translocation partners are deregulated. Well known examples in pediatric B-cell lymphomas for such IG enhancer hijacking are the oncogenes *MYC* or *IRF4*. The three molecular mechanisms described, V(D)J recombination, CSR, and SHM, involved in the Ig remodeling have all been shown to be implicated in the generation of such aberrant IG rearrangements. Remarkably, the identification of the mechanism leading to an IG translocation provides evidence at which B-cell development stage it took most likely place, as V(D)J recombination usually is restricted to B-cell precursors in the bone marrow whereas CSR and SHM involve different compartments of the germinal center [6]. The IG translocations occurring as a consequence of V(D)J recombination typically have breakpoints that involve RAG recognition sites (RSS) and are directly adjacent to Ig heavy chain J-regions (J_H) gene segments or that are adjacent to regions where the Ig heavy chain D-region (D_H) joins the J-region (D_HJ_H) [20–22]. The presence of N-nucleotides or nucleotides removed in the junctional sequence are typical features of this molecular mechanism. In contrast, the typical features of IG translocations due to aberrant SHM are that the breakpoints are located within or adjacent to rearranged V(D)J genes and that mutations in the V regions are present. Breakpoints in the IGH constant genes particularly affect switch regions and indicate that they derived from mistaken class switching.

B-Cell Differentiation

Early B-Cell Development

The development of B-cells, initiated in fetal liver, relocates to the bone marrow during the maturation of the embryo. B-cells derive from hematopoietic stem cells which give rise to cells of the myeloid as well as lymphoid lineage, the latter including the B- and T-cell progeny (Fig. 4.2). The commitment to the B-cell lineage is mediated through different transcription factors including EBF1, E2A, and PAX5 [23]. The early precursor B-cells differentiate within the bone marrow in which the B-cell receptor gene is formed by the V(D)J recombination process which is a central process for the generation of mature B-cells [24]. After rearrangement of the heavy chain locus, the pro B-cells express a precursor B-cell receptor on its surface together with a surrogate light chain [25, 26]. In case the rearrangement was non-functional, the second allele can be rearranged or the cells undergo apoptosis [27]. If the rearrangement was functional, the second heavy chain allele is suppressed (allelic exclusion) and the light chain is rearranged starting at the IGK locus [28, 29]. In case this rearrangement is non-functional, the IGL locus can be rearranged [30, 31]. Accordingly, due to isotype exclusion B-cells express usually either the Igκ or Igλ light chains. A functional rearrangement of both, the heavy and light chains leads to the expression of a B-cell receptor of the IgM isotype on the cell surface in now immature B-cells. These B-cells are counter-selected for autoreactivity [24]. In case the B-cells recognize self-antigens, cells either undergo apoptosis, the receptor can get edited using the non-rearranged allele or the cells enter a stage of immunological tolerance. The self-tolerant B-cells leave the bone marrow and pass through the spleen for further negative selection. The now mature naïve B-cells either reside in the spleen within the marginal zone or in the majority of cases circulate through the peripheral blood, lymph, and secondary lymphoid organs until they die or encounter their cognate antigen.

The Germinal Center Reaction

The term germinal center describes a histological structure located within secondary lymphoid tissue in which B-cells selected for production of high-affinity antibodies reside. When the naïve B-cell encounters its cognate antigen, these cells migrate into the T-cell zone. The interaction with the T-cells stimulates the proliferation leading to the formation of primary foci as an early GC reaction. The GC initiation relies on the induction of several transcriptional modulators. Among those modulators is BCL6, which is expressed as soon as the activated, naïve B-cells interact with a T-cell in the T-cell zone. BCL6 functions as a transcriptional repressor regulating the GC formation and maintenance including silencing of the anti-apoptotic BCL2 protein [32]. Downregulation of BCL2 ensures that the B-cells maintain in a proliferative state during SHM-based introduction of mutations [33]. In addition, it is important for maintenance of the GC B-cell state, as it downregulates factors including BLIMP1 which is a master regulator for termi-

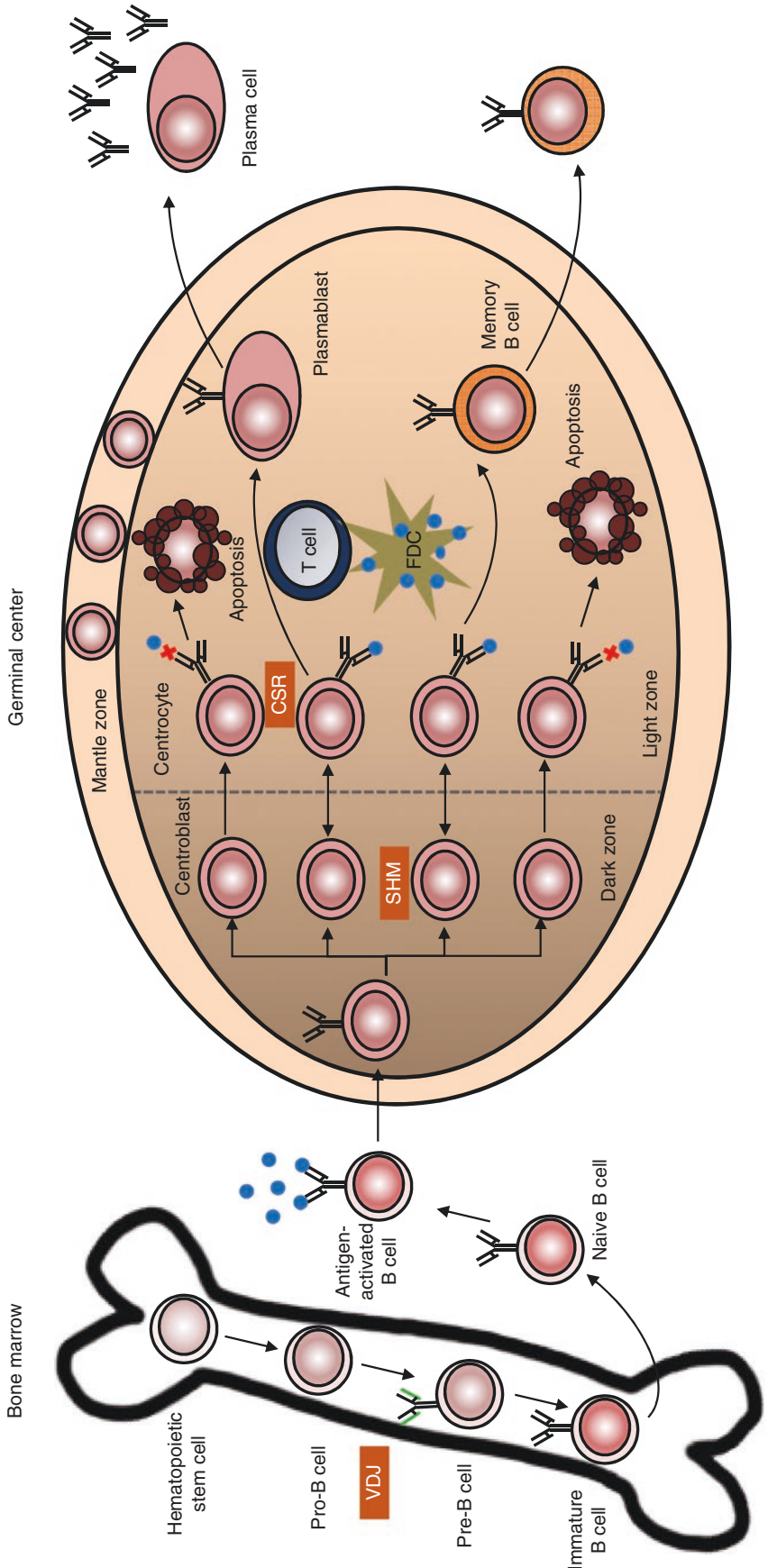

Fig. 4.2 Overview of B-cell development. Precursor B-cells develop and mature in the bone marrow, developing to naïve B-cells. After exposure of the naive B-cells with their antigen and blast transformation, they evolve into short-lived plasma cells or enter the germinal center (GC). Centroblasts can undergo apoptosis or develop into centrocytes. Post-GC cells comprise plasma cells and memory/marginal zone B-cells. Modified from [1]

nal differentiation [34]. Other important modulators for GC formation include MYC and MEF2B. After a couple of days the full germinal center structure has formed consisting of a dark zone, which is a cell dense zone consisting mainly of highly proliferative centroblasts harboring a dense nucleus. Furthermore, a light zone can be observed, in which centrocytes interact with follicular dendritic cells (FDC) as well as T-helper (T_H) cells. These zones are surrounded by a mantle zone which consists of locally, resting B-cells which were not activated by their antigen [24, 35]. The polarization of the germinal center B-cells into the dark and light zone relies on chemokine gradients of CXCL13 and CXCL12, whose receptors are upregulated in light zone (CXCR5) and dark zone (CXCR4) B-cells [36, 37]. Initially, the centroblasts within the dark zone proliferate and accumulate mutations which are inserted within the first 1–2 kb downstream of the V gene segment transcriptional start [24, 38, 39]. Mutations within this site, encoding for the variable domain of the antibody which interacts with the antigen, might increase the affinity to the cognate antigen. After a couple of proliferative cycles, the cells enter the light zone in which the affinity of the now modified B-cell receptor is tested by FDCs and T_H-cells [24, 39]. Accordingly, the centrocytes compete with others for the most specific antigen recognition. Only those with the highest affinity receive pro-survival signals by which they either undergo further rounds of proliferation within the dark zone or undergo the terminal differentiation steps [40]. One of the factors regulating the reentry into the dark zone is MYC. MYC, being repressed by BCL6 after the GC initiation, becomes reactivated in a subset of light zone B-cells that reenter the dark zone for further cycles of proliferation and SHM [41, 42]. The iterative process of the cyclic reentry back and forth from dark to light zone is a stepwise process further improving the affinity of the B-cell receptor to its antigen. Cells which are not positively selected for undergo apoptosis. During the time the centrocytes reside within the light zone, CSR takes place further diversifying the immunoglobulin repertoire.

Post-germinal Center B-Cell Differentiation

After the final steps of affinity maturation of B-cells, the GC B-cells leave the GC as either differentiated memory B-cells or plasma cells. Both cell types play important roles in the adaptive immunity, as they produce high-affinity antibodies and confer the humoral immunological memory which is the first-line immune reaction upon reencounter of the antigen. The factors leading to the terminal differentiation of GC B-cells is yet not fully understood. Current theories convey that the strength of the B-cell receptor interaction with the antigen is a determinant, another, that a developmental switch in the GC reactions exists or certain cytokines stimuli promote the cell fate.

A prerequisite for the differentiation from a GC B-cell to a plasma cell is the termination of the GC transcription program. This includes the inactivation of PAX5 which is an essential maintenance factor of mature B-cell identity. After suppression of PAX5, cells differentiate into preplasmablast cells, which secrete low amounts of antibodies. Furthermore, the downregulation of PAX5 orchestrates the regulation of other factors important for terminal differentiation [43]. Additionally, B-cell receptor and CD40 signaling triggers the activation of BLIMP1/PRDM1, which promotes the plasma cell fate [34]. Parallel to the BLIMP1 activation or even upstream, IRF4 is activated which suppresses the expression of BCL6 which is the key GC identity factor. Hence, taken together, low PAX5 and BCL6 but high IRF4 and BLIMP1 expression switch off genes required for proliferation as well as affinity maturation and promote the reprogramming to the plasma cell transcription program [34, 44, 45]. Some plasma cells migrate to the bone marrow, where these long-lived cells produce high-affinity antibodies [46]. Other plasma cells enter the medullary cords of the lymph nodes or spleen, where they express high titers of antibodies, but are depleted within 2 weeks after infection.

Memory B-cells are long-lived B-cells which can divide if at all very slowly and circle through the blood or reside in the bone marrow or spleen. They represent the initial phase of secondary immune response [47]. Upon antigen encounter, the memory B-cells become a proliferative burst and rapidly differentiate into plasma cells producing high-affinity antibodies as the first line against pathogens. In contrast to plasma cells, the factors leading to differentiation into memory B-cells are unclear. A role for phosphorylated STAT5 and BCL6 has been proposed. In addition, a CD40 stimulation in the centrocytes directing the memory B-cell differentiation has been described [48].

Extra-germinal Center Plasma and Memory B-Cell Differentiation

Within the recent years, several publications have shown that plasma cells as well as memory B-cells do not all derive from mature B-cells that have went through the GC reaction. Instead, plasma cells can as well derive from naïve marginal zone B-cells and mature, naïve B-cells circulating through blood and lymph system. Which of the cells differentiate into plasma cells depends on the nature, dose, and form of the antigen as well as the location at which the antigen was encountered. The differentiation into a plasma cell is also dependent on the interaction with a T-cell, hence if the activation of the B-cells is T-cell dependent (TD) or independent (TI) [49]. The TD immune response leads usually to the differentiation to plasma cells via the GC reaction whereas the TI independent immune response is independent from this. The TI

immune response can be induced by antigens which activate conserved pattern recognition receptors as, for example, the toll-like receptors leading to a polyclonal B-cell response (TI-1) or by antigens which have a repetitive structure as bacterial capsules which active the B-cells by B-cell receptor cross-linking (TI-2) [50]. Generally, plasma cells generated by a TI immune response are in comparison to plasma cells induced by a TD response short lived [51]. Moreover, the affinity of their B-cell receptor to the antigen is lower than from TD plasma cells, as the cells are not selected for higher affinity. Interestingly, it has been reported that class switch recombination can, although the cells did not undergo the germinal center reaction, take place, mainly switching the B-cell receptor isotype to IgG2.

The earliest antibody response to a couple of pathogens already takes place in the fetal liver. At this time point B1 cells, which are located within the peritoneal and pleural cavities, as well as, the lamina propria of the gut, can produce natural antibodies as response to some pathogens. The B-cell receptor repertoire is skewed toward antigens which induce a TI-2 immune response. When the B1 encounter such a pathogen, they migrate into the spleen or gut and produce, as the earliest plasma cells, natural IgM antibody [52].

Another source for an early plasma cell response are marginal zone B-cells. These B-cells, localized within the marginal zone of the spleen are the first B-cells to respond to pathogens and differentiate into plasma cells [53]. Mostly, the marginal zone B-cells recognize TI-2 antigens, which circulate through the blood. Hence, due to the permanent localization within the marginal zone of the spleen, the B-cells respond more rapidly than naïve B-cells to antigens localized within the blood. Activated B-cells move to the red pulp of the spleen where they undergo massive proliferation while in parallel differentiating into plasmablasts secreting immunoglobulins. In contrast to mature, naïve B-cells, they react faster to the presence of antigens and have an increased responsiveness [54].

Lastly, early plasma cell response can be conducted by mature naïve B-cells which have encountered their antigen in a TD response. But instead of inducing a GC reaction, these cells can proliferate and form extra-follicular foci of plasmablasts and plasma cells at the periphery of peri-arteriolar lymphoid sheaths. Within these foci the activated cells differentiate to plasmablasts and plasma cells producing antibodies. These cells are in contrast to post-GC short lived as the foci disappear 8 days after antigen encounter [55].

Taken together, plasma cells generated from marginal zone or mature naïve B-cells are short lived, respond rapidly as initial immune response, and have B-cell receptors which are somatically not mutated, leading to production of low-affinity antibodies to their cognate antigen.

In line with the extra germinal center plasma cell differentiation, several studies reported on the existence of memory B-cells which express IgM or have somatically unmutated B-cell receptor which indicates that they do not derive from germinal center B-cells. Hence, memory B-cells can derive early after immunization from naïve B-cells which become activated by their cognate antigen in a T-cell-dependent manner and differentiate outside the germinal center [56]. In principle the GC-independent memory B-cells harbor the same features as the GC-dependent memory B-cells including a long life, rapid proliferation capacity, high sensitivity to low-dose antibody, as well as the capability to differentiate fast into plasma cells during second immune response. In contrast, the memory quality is not as good as in GC-dependent memory B-cells, as there are a fewer isotype-switched memory B-cells, less somatic mutations, and no affinity maturation. But the latter is not a disadvantage, since GC-independent memory B-cells are already preselected, giving rise in a secondary immune response to progeny which acquire mutations and, hence, can act quicker in a second pathogen encounter.

Assumed Cell of Origin of B-Cell Lymphomas in Children and Adolescents

Different stages of B-cell differentiation are characterized by the specific structure of the BCR, the transcriptional and epigenetic profiles, as well as, the expression pattern of differentiation markers. When B-cells go through malignant transformation, they usually keep the features of the respective differentiation stages. Nevertheless, the initial transforming event might occur on a different B-cell maturation stage than the one the tumor cells are ultimately frozen in. The best known example is follicular lymphoma in adults, where the pathognomonic IGH-*BCL2* translocation derives from a mislead V(D)J rearrangement in bone marrow B-cell precursor cells, whereas the actual tumor cells clearly show morphologic, transcriptional, and immunologic features of germinal center B-cells with ongoing somatic hypermutation of IG genes. Despite such discrepancies, the supposed cell of origin or the frozen maturation state are used to determine the origin of the human B-cell lymphomas [1, 57].

Taking these pitfalls and limitations into account, B-cell lymphomas can be divided into the following groups (Fig. 4.3):

- *B-cell precursor neoplasms*: B-cell lymphoblastic lymphoma in many aspects resembles precursor B-cell lymphoblastic leukemia. The cell of origin based on the marker profile (particularly TdT) and immune gene analysis is supposed to be a B-cell precursor in the bone marrow. There might be some heterogeneity regarding the different pro- and pre-B-cell stages.

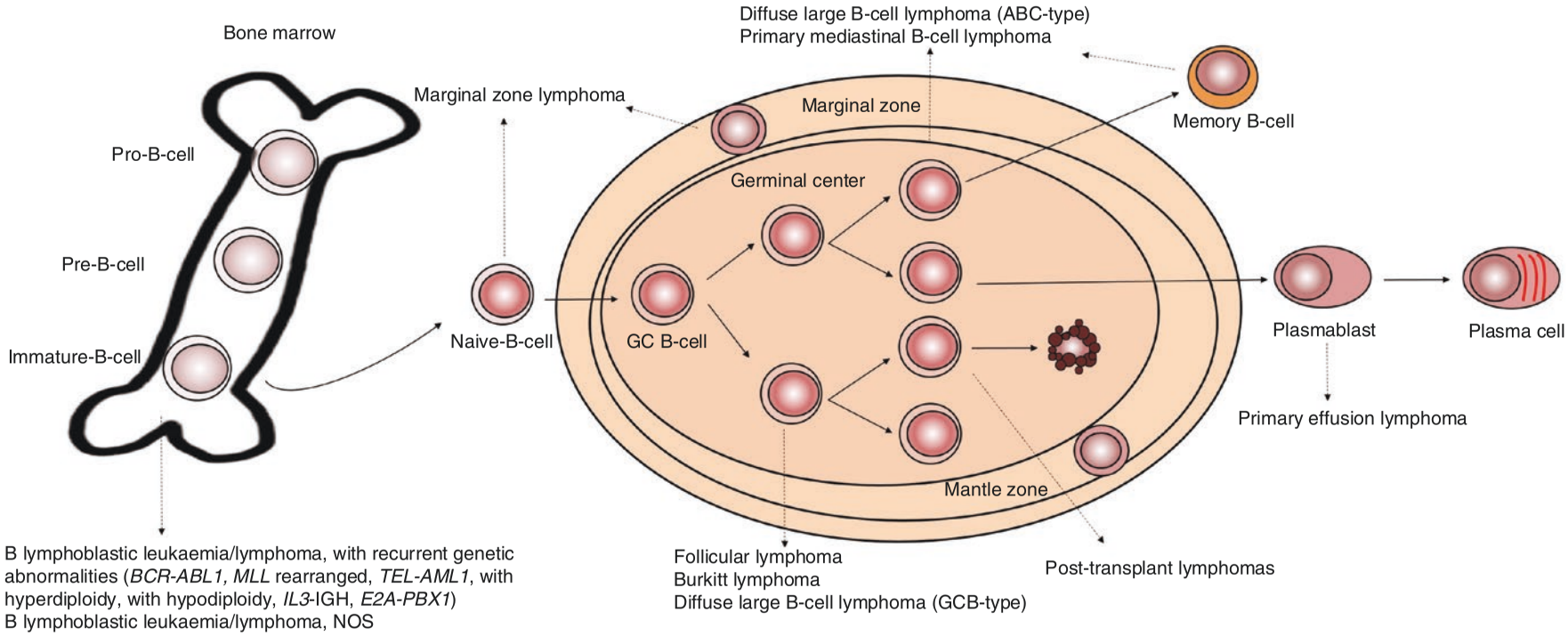

Fig. 4.3 Diagrammatic representation of cellular origin of the most common B-cell lymphomas in children and adolescents. B-cell tumors develop from stages of B-cell maturation. Most lymphomas are derived from germinal center (GC) B-cells or from B-cells that went through the GC. Nevertheless, precursor B-lymphoblastic leukemia and lymphoma are derived from precursor B-cell from the bone marrow compartment

- *Germinal center-derived B-cell lymphomas:* The vast majority of pediatric and adolescent lymphomas derive from cells which had entered the germinal center. Both endemic and sporadic Burkitt lymphomas (BL) show several features of dark zone cells (centroblasts). At least sporadic BL might be derived from B-cells poised to become IgA-expressing cells and a link to a primary immune response is discussed. Diffuse large B-cell lymphoma based on gene expression data are divided into those with similarity to GC B-cells (GCB-DLBCL) or to in vitro-activated B-cells (ABC-DLBCL). ABC-DLBCL are rare in young patients, their incidence increases with age. Thus, most pediatric and adolescent DLBCL are of GCB type. Primary mediastinal and central nervous system B-cell lymphomas are also derived from cells with contact to the germinal center but might be more advanced in the GC reaction than typical GCB.
- *Marginal zone B-cell lymphomas:* Nodal marginal zone B-cell lymphomas (NMZL) derive from marginal zone B-cells, a subset of those might also derive from naïve B-cells [58].

Mechanisms Contributing to the Transformation of B-Cells

Somatic Genetic Alterations

The theory of cancer development postulates that a normal cell requires several "hits" that change its normal functions. Cellular functions typically altered by such hits are cell cycle control, proliferation and apoptosis, B-cell inherent signaling pathways, and epigenetic modifiers. Various mutational mechanisms can contribute to the generation of such mutations. Besides the B-cell inherent mechanisms outlined above, mutational signature analyses have pointed to the role of aging or antiviral responses triggered by the APOBEC family [59, 60]. These hits can occur either in cancer drivers, which are typically divided into oncogenes and tumor suppressor genes, but also in passengers, which only have a minor importance in the transformation or clonal evolution. Cells with genetic abnormalities are selected based on the fitness and survival and have the opportunity to acquire further aberrations. Different genomic aberrations, like chromosomal translocations, single nucleotide variants, or copy number changes lead to oncogene activation and tumor suppressor gene inactivation.

Oncogene Activation via Chromosomal Translocations

Chromosomal translocations usually activate oncogenes either via enhancer hijacking or via the production of fusion transcripts.

In case of enhancer hijacking, the complete (coding part) of a gene is usually brought in the vicinity of an enhancer of another gene expressed in the (cell of origin of the) tumor cells. Typical events associated with enhancer hijacking in pediatric and adolescent B-cell lymphomas are chromosomal translocations affecting the IGH locus on 14q32.33 or the IGK and IGL light-chain loci on 2p12 and 22q11, respectively. The intact oncogenes, encoding, e.g. *MYC* or *IRF4*, are juxtaposed to the various enhancer elements of the IG loci resulting in deregulated expression of the oncoproteins [61] (Table 4.1). Besides, the SHM machinery might further activate the oncogenes through mutations, e.g. in the MYC boxes after the translocation, as the machinery "jumps" over from the IG locus to the partner chromosome after the juxtaposition. The probably most promiscuous oncogene in B-cell lymphomas, which can be activated by hijacking of enhancers from a wide set of genes, is *BCL6*, which encodes a key regulator of germinal center B-cells. Remarkably, the set of genes deregulated by enhancer hijacking in mature B-cell lymphomas shows some dependence of age: whereas *BCL2* deregulation through translocation to the IGH locus is present in around 85% of follicular lymphomas and 30% of diffuse large B-cell lymphoma of adulthood, this change is rare in the same disease in the adolescent population and almost completely absent below the age of 18 years (with the notable exception of IG-*MYC* translocated Burkitt like lymphoma with precursor phenotype). Similarly, the frequency of *BCL6* translocations is lower in children than in (elderly) adults, which might be associated with the age-associated changes in cell of origin of these neo-

Table 4.1 Chromosomal translocations involving the IG loci in mature B-cell neoplasm and children and adolescents

Disease	Translocation	Partner gene
Burkitt lymphoma	t(2;8) (p12;q24)	*MYC*
Large B-cell lymphoma (LBCL) with *IRF4* rearrangement	t(2;6) (p12;p25)	*IRF4*
Diffuse large B-cell lymphoma, Follicular lymphoma	t(3;14) (q27;q32)[a]	*BCL6*
Large B-cell lymphoma (LBCL) with *IRF4* rearrangement	t(6;14) (p25;q32)	*IRF4*
Burkitt lymphoma, Diffuse large B-cell lymphoma	t(8;14) (q24;q32)	*MYC*
Diffuse large B-cell lymphoma	t(9;14) (p13;q32)	*PAX5*
Burkitt lymphoma, Aggressive B-cell lymphoma	t(14;16) (q32;q24)	*CBFA2T3*
Follicular lymphoma, Diffuse large B-cell lymphoma	t(14;18) (q32;q21)[a]	*BCL2*
Burkitt lymphoma	t(8;22) (q24;q11)	*MYC*
Large B-cell lymphoma (LBCL) with *IRF4* rearrangement	t(6;22) (p25;q11)	*IRF4*

[a]Variants with IG light-chain loci in 2p12 and 22q11 have been reported

plasms. In contrast, IG-*IRF4* translocations are more frequent in younger patients; the same seems to hold true for Burkitt lymphoma with IG-*MYC* translocation in general, which might point to an age-dependent susceptibility to these diseases. It is intriguing to speculate that the latter is associated with the frequency of primary immune response particularly those driven by enteric microbiota.

Whereas enhancer hijacking is quite common in mature B-cell lymphoma and associated with many of the hallmark chromosomal alterations, the development of fusion transcripts from two separate genes seems to be rather rare in mature B-cell neoplasms [61]. In precursor B-cell neoplasms, i.e., lymphoblastic leukemia/lymphoma, a set of fusion genes has been described, including e.g. *BCR/ABL, ETV6/RUNX1,* or *MLL*-fusions, to name the most common. In mature B-cell lymphomas, the probably best known are *ALK*-fusion genes. ALK-positive large B-cell lymphoma is a very rare lymphoma, characterized by the chromosomal translocation t(2;17)(p23;q23), involving *Clathrin* (*CLTCL*) gene in 17q23 and the *ALK* gene in 2p23, generating a CLTCL-ALK fusion protein [62, 63]. Some cases are associated with the t(2;5)(p23;q35), generating the NPM1-ALK fusion protein. Fusion transcripts encode fusion proteins containing parts of both involved proteins. The part of the partner protein changes the protein function of the oncoprotein, e.g. in case of ALK-fusion leads to constitutive activation of kinase activity due to (aberrant) homodimerization.

Though it is usually assumed that chromosomal translocations lead to oncogene activation, it needs to be emphasized that tumor suppressor gene inactivation due to gene disruption is in part associated with intron retention in fusion transcripts and seems to be a rather much more common consequence of these structural chromosomal aberrations.

Tumor Suppressor Inactivation

Tumor suppressor gene inactivation usually occurs via biallelic deletion and/or mutation. An alternative mechanism to deletion is copy neutral loss of heterozygosity (CNN-LOH), sometimes described as (partial) uniparental (iso) disomy. In addition, tumor suppressor gene function might also be altered by mono-allelic inactivation and haploinsufficiency or by expression of a mutant dominant-negative protein form.

Tumor suppressor genes commonly altered in many tumors including various subtypes of pediatric and adolescent B-cell lymphomas are *TP53* and *CDNK2A*. Mostly, changes in these genes are secondary or even late events in clonal evolution and particularly *TP53* inactivation has been frequently linked to unfavorable prognosis. In case of Li Fraumeni syndrome, monoallelic inactivation can predispose to lymphomas, similarly to *ATM* inactivation in Ataxia telangiectasia. Whereas both latter genes function particularly in DNA repair, another class of tumor suppressors commonly hit in mature B-cell lymphomas both in younger and older patients are genes involved in epigenetic modifications, like KMT2D *(formerly known as MLL2*), encoding a histone methyltransferase, *CREBBP*, encoding a histone acetyltransferase, and *SMARCA4*, being a member of the SWI/SNF chromatin remodeling complex.

Other tumor suppressor genes recurrently targeted in particular subtypes of pediatric and adolescent B-cell lymphomas are *TNFRSF14A*, frequently targeted in pediatric follicular lymphoma, and *ID3*, mutated in around 60–70% of sporadic Burkitt lymphomas [64, 65]. The *ID3* mutations free its binding partner, namely, TCF3, from the heterodimeric complex and allow TCF3 to bind the DNA and activate its targets. Remarkably, activating mutations of the *TCF3* oncogene do have the same effect showing the strong interplay of tumor suppressors and oncogenes.

Oncogene Activation

As shown above oncogene activation cannot only occur though chromosomal translocations as detailed before, but also like in case of *TCF3* through activating mutations. Similarly, a high proportion of FL particularly in older patients carry activating mutations of the methyltransferase gene *EZH2*. Additional means of oncogene activation are copy number gains up to amplifications. The oncogenes outlined above, like *MYC* or *IRF4*, are also recurrently hit by such copy number gains. There are several other examples of oncogene activation described in B-cell lymphoma, e.g. gains in *REL*, a component of the NF-κB complex [66, 67]. Moreover, gains in the 9p region, typically detected in primary mediastinal B-cell lymphoma (PMBL), lead to activation of *JAK2, PDL1*, and *PDL2*. The prior is involved in activation of the JAK/STAT pathway, the latter two, also involved in chromosomal translocations with various partners, play a role in the immune detection of the tumor cells. Besides coding genes, also non-coding genes can be targeted by activating oncogenic mechanisms. Probably the best known is gain of oncogenic miR-17-92 cluster, which is a transcriptional target of *MYC* [68] frequently altered in BL.

Oncogenic activation through mutations can also be a side effect of SHM and CSR at non-Ig genes [69]. Such off-target activity of SHM produces point mutations in proto-oncogenes like *BCL6* and *CD95* which are also mutated in a considerable fraction of normal GC and memory B-cells.

Signaling Through the B-Cell Receptor

The selection of cells expressing a BCR is also a common feature in malignant B-cells. The majority of B-cell lymphomas express a BCR, however sometimes at low levels [1, 70–72]. In case of an IG translocation, this usually affects a non-productive IG locus and leaves the productive IG locus intact [73, 74] indicating that at least when the IG transloca-

tion occurred, the BCR signaling has been necessary for B-cell survival and development of the B-cell neoplasm. Moreover, several types of B-cell lymphomas show ongoing mutations in the IGH V-region during tumor clone progression [78–80]. Though such mutations if deleterious could prevent functional heavy and light-chain pairing, those tumors still express the BCR indicating selection against damaging mutations [64, 66]. In fact, the low frequency of BCR loss in subtypes of B-cell lymphomas with ongoing somatic hypermutation, like FL and MALT lymphoma, shows the importance of BCR expression in these B-cell lymphomas. Consequently, the survival signals mediated by the BCR expression in normal B-cells likely also play a role in the survival of at least a subset of B-cell lymphoma cells.

BCR-Dependent Lymphomas

Some B-cell lymphomas use the IgM constant regions to form their BCR; however, the majority of B-cell lymphomas derives from germinal center cells that usually switch their BCRs from IgM to IgG. Notably, IgM and IgG-associated BCRs are linked to different downstream signaling: IgM-BCR signaling promotes the survival and proliferation of B-cells by activating pathways like the NF-$_{\kappa}$B pathway; in contrast, IgG-BCR signaling promotes plasmacytic differentiation by the activation of ERK and MAPK pathways [75–77].

A prototype of lymphomas retaining IgM-BCR expression is FL [78]. This lymphoma is genetically characterized by a t(14;18) translocation, involving the IGH locus and the *BCL2* gene. In FL, the productive IGH allele is never translocated to *BCL2* and assures expression of IgM. In contrast, the non-productive allele is translocated and undergoes CSR to IgG, showing the selective pressure on the cell to retain IgM expression [79]. Another example is DLBCL, where the GCB subtype usually expresses IgG-BCR but does not require BCR signaling for survival [80], whereas the ABC subtype retains the IgM-BCR [81] in part due to deletions within the IGH "switch" regions (Sμ and Sɣ), needed for class switch recombination [82]. These deletions take place on the productive IGH allele, and blocking class switch recombination [83].

There are two forms of pathological BCR signaling in B-cell malignancies: Chronic active BCR signaling involves diverse downstream pathways, including MAPK, PI3K, NFAT, and NF-kB pathways. This form is typical for ABC-DLBCL, where the presence of mutations in *CD79A* and *CD79B* is reported in over 20% of the cases. Functional analyses showed that knockdown of proximal BCR subunits, including IgM, IgK, CD79A, and CD79B, is lethal for ABC-DLBCL [81]. On the other hand, tonic BCR signaling activates only the PI3K pathway, like in BL. As outlined above, the majority of BL cases show activating mutations in *TCF3* or inactivating mutations in its negative regulator, *ID3*. TCF3 has been reported as a factor required for expression of all immunoglobulin genes. In addition, TCF3 represses the expression of SHP1, a negative regulator of BCR signaling. Hence, TCF3 enhances BCR signaling by two different ways. Correspondingly, knockdown of TCF3 decreased PI3K activity in BL cell lines and was lethal. Moreover, HSP90 induces apoptosis in BL cells due to the disruption of tonic BCR signaling. HSP90 impairs SYK kinase which is required for the efficient activation of BCR complex [84].

An atypical form of BCR signaling is observed in follicular lymphoma caused by the presence of N-linked glycosylation acceptor sites in the V region induced by SHM [85]. Due to these, BCRs are modified by high-mannose oligosaccharides, which can interact with mannose-binding lectins present on the stromal cells in the tumor microenvironment, leading to cross-linking of the BCR and initiation of the BCR signaling [86, 87].

BCR-Independent Lymphomas

Despite the essential role of BCR expression and signaling in many B-cell lymphomas, there are a considerable number of exceptions. For example, inactivating IGH V-region gene mutations have been described in at least 10–20% of the cases of post-transplant lymphomas [88–90]. As these are EBV-positive lymphomas, it is assumed that expression of the EBV-encoded latent membrane protein 2A (LMP2A) can replace for the BCR expression and signaling. More controversial is the role of BCR function in primary mediastinal B-cell lymphomas. These lymphomas usually lack expression of a BCR, and the components of the BCR signaling cascade are downregulated. Nevertheless, these lymphomas are usually not associated with inactivating IGH V-region gene mutations [91–93].

The Role of Pathogens in Pediatric and Adolescent B-Cell Lymphomas

Some viruses clearly seem to contribute to the pathogenesis of pediatric and adolescent B-cell lymphomas. The best documented examples are two members of the ɣ herpes virus family, human herpes virus (HHV) 4, better known as EBV, and human herpes virus 8 (HHV8) [3, 94].

EBV has been shown to contribute to the pathogenesis of Burkitt lymphoma (BL) and post-transplant lymphoproliferative disorders (PTLD). Of BL, three epidemiologic variants are described: endemic, sporadic, and immunodeficiency-associated, with the frequency of EBV infection differing significantly between the variants. The endemic BL variant shows EBV infection in the tumor cells in nearly all of the cases [95]. In contrast, EBV infection of the tumor cells is only described in 25–40% of patients with

immunodeficiency-associated BL [96, 97] and even less frequently in sporadic BL, where it can be detected in less than 20% of the cases. Importantly, EBV seronegativity at the time of transplantation is the most important risk factor for EBV-driven PTLD. About 60% of the PTLDs are EBV-positive. Nevertheless, EBV-negative PTLDs are also reported in around 20–40% of the cases and more common in adults.

With regard to EBV, three latency types specific to individual EBV-associated tumors have been described. Latency type I is associated with BL and shows restricted expression of EBV-encoded nuclear antigen 1 (EBNA1), the EBV-encoded small RNAs (EBERs), and BAMHI A rightward transcripts (BARTs) [98–100]. Latency type II infected cells typically express EBNA1, EBERs, BARTS, and the latent membrane proteins (LMP1, LMP2a, and LMP2B). This latency type is associated with Hodgkin lymphoma. LMP1 and LMP2a mimic an active CD40 receptor and BCR, respectively [101]. Both latent membrane proteins provide survival signals for B-cells in the GC. Latency type III, which is associated with PTLD and EBV-transformed lymphoblastoid cell lines (LCLs), expresses both transcripts and all the EBV latent proteins, containing the six nuclear antigens (EBNA1, EBNA2, EBNA3A, EBNA3B, EBNA3C, and EBNA-LP), and the three membrane proteins (LMP1, LMP2A, and LMP2B). EBNA2 is important to drive the proliferation of the transformed B-cells [3]. On the other hand, EBERs are expressed by all EBV-infected cells, which is used for the EBV detection by means of in situ hybridization for these transcripts.

HHV8 is detected in few DLBCL, not otherwise specified, and in primary effusion lymphoma (PEL). PEL is a very rare lymphoma mainly found in acquired immune deficiency syndrome (AIDS) patients. HHV8 establishes a latent infection in B-cells and encodes several homologues to human proteins including cytokines (interleukin-6, macrophage inflammatory proteins, interferon-regulatory factors) and regulatory genes (cyclin D, G-protein coupled receptor, etc.) [94, 102].

In addition to HHV4 and HHV8, a pathogenetic role of other viruses like hepatitis C virus (HCV) [103] and HHV6 is being discussed for lymphomagenesis [104]. Future studies using next-generation sequencing (NGS) will probably provide further insights into the role of pathogens in B-cell lymphomagenesis.

Besides viruses, also other pathogens have been linked to the development of B-cell lymphomas. In particular, subtypes of marginal zone lymphomas of MALT type have been associated with infections with *H. pylori*, *C. psittaci*, or *B. burgdorferi*. The best evidence of a pathogenic role for foreign antigens in these lymphomas derives from MALT lymphomas of the stomach [105]. The vast majority of MALT lymphoma patients are infected by H. pylori, where the antibiotic treatment targeted to the pathogen can cure these patients [106, 107]. Besides, some patients with DLBCL of the stomach infected by H. pylori can be also cured using antibiotic treatment [108]. Whether such pathogens also play a role in pediatric marginal zone lymphomas needs to be investigated.

On a more general level, various subtypes of lymphomas are assumed to be polymicrobial diseases. A clear link to malaria infections caused by P. falciparum has been established for endemic BL where an immune stimulatory and probably AID-mediated DNA damaging role of the parasite infection is discussed. In sporadic BL the localization of the tumors in the ileoceacal region, a cell of origin being a B-cell poised to IgA expression and an incidence curve with intriguing parallels to the IgA-expression and Peyer patch development might indicate a role of the primary immune response to the microbiome colonialization of the gut. In analogy, also B-cell lymphomas predominately presenting in the Waldeyer's ring like *IRF4*-rearrangement positive LBCL might be triggered by pathogenic infection. Antigenic stimulation seems also to be involved in splenic marginal zone lymphoma (SMZL). Some patients with a SMZL with villous lymphocytes are infected with hepatitis C virus (HCV), and treatment against this virus using interferon-α (IFN-α) abolishes the lymphoma in around 75% of these patients [109]. In contrast, IFN-α has no effect in other patients with disease but without HCV infection. Thus, the overall role of pathogens besides direct oncogenic function but rather as promoting factor in B-cell lymphomas needs further investigation, particularly in the light of BCR signaling and primary immune responses like in children, young adults, and after transplantation.

The Role of the Microenvironment in the Pathogenesis of B-Cell Lymphomas

The interaction of tumor cells with cells in the tumoral microenvironment can also affect survival and proliferation of the malignant B-cells in various lymphomas. For example, follicular lymphoma cells proliferate in follicular structures associated with T-helper cells and follicular dendritic cells, resembling normal GC B-cells. In vitro studies have shown that follicular lymphoma cells, among others, receive stimulation via the CD40 receptor from the microenvironment. Expression and signaling via CD40 is a main survival signal also for normal GC B-cells [110, 111].

In addition, the macrophage (MP) infiltration has been described as pathogenetic factor in various lymphomas even linked to prognosis in several studies [112, 113]. Macrophages are heterogeneous, multifunctional, myeloid-derived leukocytes that are part of the innate immune system. Tumor-associated MPs (TAMs) are MPs with specific M2 phenotype that play a central role in the pathophysiology of tumors [114].

Related to B- and T-cell neoplasms, TAMs are involved in tumor progression, often associated with poor prognosis owing to the secretion of chemokines and cytokines. Different active proteases stimulate tumor growth, angiogenesis, metastasis, and immunosuppression [115]. In DLBCL, the implication of MPs has been related to the ability of DLBCL cells to escape the immune surveillance of tumor-specific cytotoxic T-cells recruiting M2 TAMs that highly express immune checkpoint molecules, such as PD-L1 and PD-L2, on their surfaces. These interact with PD-1 receptors expressed on intratumoral T-cells and provide inhibitory signals. This could be an explanation for the effective therapy with anti-PD-/PD-L1 in some cases of DLBCL [116].

Epigenetic Alterations Leading to Tumorigenesis

In addition to the genomic alterations contributing to the initiation and progression of B-cell lymphoma and leukemia, the importance of epigenetic alterations in the pathogenesis of these neoplasias has been acknowledged over the last years. The best studied epigenetic marks in B-cell neoplasias are DNA methylation and histone modifications, which are involved in basic biological mechanisms including regulation of gene expression, replication, and DNA repair (as reviewed in [117, 118]).

Among the histone modifiers most frequently altered in B-cell lymphomas are EZH2, CREBBP, and EP300 as well as KMT2D. The methyltransferase EZH2 is part of the Polycomb Repression Complex 2 (PRC2), which methylates histone 3 lysine 27 (H3K27) a marker for repressed chromatin (heterochromatin). In B-cells, EZH2 is expressed during early B-cell development where it plays a role in VDJ recombination [119]. In naïve B-cells, the expression of EZH2 is downregulated. During the GC reaction, EZH2 is upregulated and establishes repressive H3K27 marks at promoters of genes which are involved in differentiation and cell cycle regulation [120, 121]. *EZH2* mutations are frequently detected in DLBCL as well as FL, with the most recurrent alteration affecting a tyrosine in the functional SET domain of the protein (Tyr641) being mutated in 21.7% of DLBCL and 7.2% of FL [122]. This mutation is predicted to affect the enzymatic activity by shifting the efficiency to methylate H3K27 to trimethylation instead of mono- or demethylation [123, 124].

The Mixed Lineage Leukemia (MLL) gene family belongs also to the histone methyltransferases. In contrast to the PRC2 complex, members of this family methylate histone 3 lysine 4 (H3K4) which is a marker for transcriptional activation [125]. Diverse alterations affecting these modifiers have been reported. Accordingly, *KMT2A* (formerly known as MLL1) is translocated in about 70% of infant leukemias [126]. The catalytic SET domain conducting the methyltransferase activity is frequently lost due to the translocation. Nevertheless, it is believed that the MLL1 alterations are associated with aberrant histone methylation and hence, with the overexpression of target genes. Truncating and frameshift mutations of *KMT2D* have been reported to occur in up to ~90% of FL [66, 127] and ~30% of DLBCL [128, 129]. These mutations affecting the SET domain lead to a loss of function and, hence, to a deficiency of H3K4 methylation, suggesting a tumor-suppressive function for KMT2D [130].

In addition to histone methylation, frequent alterations in B-cell neoplasia affect histone acetylation via changes in histone acetyltransferases (HAT). Histone acetylation mediates an open chromatin structure allowing bromodomain proteins to be recruited which induce the transcriptional activation. The most recurrently altered HAT in malignant B-cell lymphomas are CBP (encoded by *CREBBP*) and p300 (encoded by *EP300*) which are described to have tumor-suppressive functions in B-cell lymphoma [67, 131]. About ~30% of DLBCL harbor alterations leading to a loss of CREBBP HAT domain function, with GCB-DLBCL being more frequently affected than ABC-DLBCL [67]. The HAT domain is also inactivated in ~30% of FL [67] and 18% of relapsed pediatric B-ALL [131]. The inactivation of CBP leads to an expansion of the GC B-cell compartment, downregulates MHC class II expression, and promotes tumor cell growth [132]. Mutations disrupting the HAT domain of p300 were reported in ~10% of DLBCL [67, 133] and FL [67]. P300 inactivation confers resistance against BCL6 inhibitors [133].

Apart from the changes affecting the histone modification, changes in DNA methylation take place during B-cell development and contribute to the physiological processes linked to the differentiation. Hence, each developmental stage of a B-cell is composed of an unique epigenetic pattern [4, 134]. B-cell neoplasias maintain a certain degree of similarity to their assumed normal B-cell counterpart; thus, the DNA methylation pattern can be used for the determination of the cell of origin and for classification purposes [134, 135]. On the other hand, the neoplastic B-cells are characterized by a number of DNA methylation changes which in part interact with genomic and transcriptional changes in the deregulation of key transforming processes [135–140].

The establishment of DNA methylation patterns involves the DNA methyltransferases (DNMT) DNMT1, DNMT3A, and DNMT3B. DNMT1 has been shown to be significantly upregulated in GC B-cells suggesting a role in GC reaction and differentiation [141]. Indeed, experiments with *Dnmt1* hypomorphic mice have shown that the GC formation upon immunization is impaired [141]. In line, DNMT1 as well as DNMT3B overexpression has been described in 69% and 86% of BL, respectively [142].

Pathogenetic Hallmarks of Common Subtypes of B-Cell Lymphomas in Children and Adolescents

Burkitt Lymphoma and Burkitt-Like Lymphoma with 11q Aberration

The hallmark genetic aberration as well as the assumed primary event in all three epidemiologic subtypes of BL is t(8;14)(q24;q32) or its variants t(2;8)(p12;q24) and t(8;22)(q24;q11). All these changes lead to deregulation of the *MYC* oncogene by its juxtaposition next to one of the enhancer elements in the IGH (14q32), IGK (2p12), or IGL (22q11) locus (Table 4.2).

BL is characterized by a low genomic complexity. Cytogenetically, an IG-*MYC* translocation is detected as sole abnormality in 40% of cases. The most frequent secondary alterations are structural aberrations involving chromosome 1 (>30% of the patients), especially of the long arm, and often resulting in a partial trisomy 1q. Aberrations affecting the chromosomal region 13q31, mainly involving the mir-17-92 miRNA cluster [68], have been reported in 15% of the cases [143]. Moreover, gains of chromosomes 7 and 12 and deletions in 6q and 17p are common. Besides the secondary chromosomal imbalances, recent genomic sequencing studies have identified recurrent somatic mutations in *MYC*, *ID3*, *TCF3*, *CCND3*, *SMARCA4*, *TP53, FBXO11, ARID1A, DDX3X* in both sporadic and endemic BL [64, 135, 144, 145] (Table 4.2).

The existence of BLs without an IG-*MYC* translocation has been subject to controversial discussion. A provisional entity of *MYC*-negative "Burkitt-like lymphoma with 11q aberration" has been recently included in the new WHO lymphoma classification. Those cases resemble BL based on gene expression profile and pathological characteristics but importantly lack a *MYC* translocation. Instead, 11q aberrations with proximal gains and telomeric losses are typical [146] (Table 4.2).

Table 4.2 Overview of the frequent chromosomal aberrations in pathogenetic hallmark of B-cell lymphomas in children and adolescents

Disease	Cytogenetic aberration/ genes involved	Mutations
Burkitt lymphoma	t(8;14)(q24;q32)[a]/*MYC* 1q aberrations Trisomy 7, trisomy 12 dup(13q)/mir-17-92	*ID3, TCF3, CCND3, TP53, SMARCA4, FBXO11, ARID1A, DDX3X*
Burkitt-like lymphoma with 11q aberration	Gain 11q23.2-q23.3/ *PAFAH1B2* Loss 11q24/ *ETS1* Gains 7q34-qter, 12pter-p12.2, 18q21.2, 19pter-p13.2 Loss 6q14.3-q22.2	
Diffuse large B-cell lymphoma	t(3;14)(q27;q32)[a]/*BCL6* t(8;14)(q24;q32)[a]/*MYC* t(14;18)(q32;q21)[a]/*BCL2* *+3/3q, +18/18q, +19q, del(6q), del(9p)/ CDKN2A +1q, +2p13-p16, +7, +11q, +12/12q*	*BCL6, EZH2, KMT2D, CREBBP, PRDM1, TNFRSF14, CARD11, GNA13, CD79B, MYD88*
ALK+ large B-cell lymphoma	t(2;17)(p23;q23)/*CLTC-ALK* t(2;5)(p23;q35)/*NPM1-ALK*	
Primary mediastinal large B-cell lymphoma	t(3;14)(q27;q32)[a]/*BCL6* t(14;16)(q32;p13)/*CIITA* *+9/9p23-p24/ JAK2, PDL1, PDL2* *+2p13-p16/ REL and BCL11A*	*STAT6, PTPN1, ITPKB, MFHAS1, XPO1*

[a]variants with IG light-chain loci in 2p12 and 22q11 have been reported

Diffuse Large B-Cell Lymphoma (Including ALK+ Large B-Cell Lymphoma)

DLBCL is a heterogeneous group of diseases with varying morphologic, immunophenotypic, and molecular features. Several of these features change with age [147]. DLBCL in children and adolescents, in contrast to those occurring at older age, are enriched for GCB-type cases and depleted for *BCL2* and *BCL6* translocations. The mutational landscape of DLBCL in children and young adults warrants further investigation.

A special subgroup of large B-cell lymphoma enriched in young patients is ALK+ large B-cell lymphoma (ALK+ LBCL). It is a rare and an aggressive neoplasm, accounting for <1% of DLBCLs. The key player of this lymphoma is the overexpression of ALK protein, as a result of fusion protein generated by a translocation of the *ALK* gene on chromosome 2, as described above. Typically these translocations are associated with complex karyotypes. The STAT3 pathway is constitutively activated in ALK+ LBCLs and the tumors respond to ALK inhibitors (Table 4.2).

Primary Mediastinal Large B-Cell Lymphoma

Primary mediastinal large B-cell lymphoma (PMBL) is a mature aggressive large B-cell lymphoma (LBCL). It affects mainly young adults with a predominance of females and a median age at diagnosis of 35 years. PMBL display a specific gene expression profile which is different from GCB or ABC DLBCLs but shows similarities to Hodgkin lymphoma. Chromosomal aberrations affecting the *BCL6*, *MYC*, and *BCL2* loci are absent or rarely detected. Nevertheless, translocations involving *CIITA* locus in 16p13.3 have been described in more than 50% of cases. The partner genes of translocations are in the majority of cases *PDL1* or *PDL2* but these are not the only partners described. In addition, gains

in the region of 9p, containing the genes *JAK2*, *PDL1*, *PDL2*, and gains in 2p16.1, including the genes *REL* and *BCL11A*, have been described as recurrent genetic events in PMBL [148, 149] (Table 4.2).

PMBL are characterized by a constitutively activated JAK/STAT signaling pathway. Furthermore, mutations in *STAT6* and *PTPN1*, a negative regulator of JAK/STAT signaling, are detected in 72% and 25% of PMBL cases, respectively. Additional mutations affect *ITPKB*, *MFHAS1*, and *XPO1* [148] (Table 4.2).

Large B-Cell Lymphoma with *IRF4* Rearrangement

Large B-cell lymphoma (LBCL) with *IRF4* rearrangement is an uncommon lymphoma, comprising for 0.05% of diffuse LBCLs. This neoplasm shows a decreasing incidence in older age groups.

Besides expression of germinal center markers, the immunophenotype is characterized by the strong expression of IRF4/MUM1. Moreover, BCL6 is co-expressed, while PRDM1 is frequently negative [148].

The key genomic event in this lymphoma is an often cryptic rearrangement of *IRF4* with an IG locus, *BCL6* rearrangement have also been detected in LBCL with *IRF4* breaks, whereas *MYC* and *BCL2* breaks were absent in the reported cases. In addition to *IRF4* rearrangements, the genomic profile of this entity shows a complex pattern of genetic changes including *TP53* deletions [148, 150] (Table 4.2). Nevertheless, patients have favorable outcome after treatment.

Pediatric-Type Follicular Lymphoma

Pediatric-type follicular lymphoma (PTFL) is an infrequent nodal follicular lymphoma (FL) that appears in children and young adults, but can also occur in the older population. This neoplasm is characterized by the lack of genomic rearrangements involving the *BCL2*, *BCL6*, or *IRF4* locus. Moreover, the frequent mutations affecting KMT2D, *CREBBP*, and *EZH2* described in adult FL, are absent in PTFL. Instead, deletions in the1p36 chromosomal region or mutations involving *TNFRSF14* are the most common genetic aberrations in PTFL. Moreover, mutations in *MAP2K1* have been reported in 40–50% of the cases [148] (Table 4.2).

Conclusions and Outlook

As detailed above and exemplified for B-cell lymphomas common in young patients, the pathogenesis of B-cell lymphomas is based on multifactorial grounds. The complexity, interdependency, and timely order of pathogenetic processes ultimately leading to clinically overt B-cell lymphomas are yet by far not completely understood. Even the distinction between driver and passenger events is still challenging, though some key lymphoma-initiating events like the Burkitt translocation t(8;14) have been known since more than 40 years. The reception of the above detailed mechanisms involved in the pathogenesis of B-cell lymphomas has to take into account that most current models of B-cell lymphomagenesis rely on simple, mostly monodimensional data and assumptions. Moreover, a linear and directional evolution of a B-cell lymphoma from a non-neoplastic precursor is usually assumed. Nevertheless, in fact to capture the overall complexity of B-cell lymphomagenesis, each neoplastic cell of a probably heterogenic tumor would have to be mapped at a single point in a space of n-dimensions, with several of the factors outlined above but also all different OMIC layers being separate dimensions. Moreover, the age of the patient and time since tumor initiation have to be considered as dimensions in this space. It needs to be clearly stated, that most of the concepts outlined above do not take such a "spacial" approach. Moreover, in many instances the experimental or observational procedures providing the data for the models likely modify features which appear different in the native host of the tumor, i.e., the patient. All this has to be taken into account if such observations are translated into clinics. Clearly, no diagnostic test yet captures the biologic complexity in its entirety (and if so probably best morphology which in essence provides a birds-eye view on the tumoral processes). Indeed, given the number of deregulated processes, altered genes, and changed signals, it seems rather surprising that the tumor takes advantage rather than disadvantage from those. Likely, this is due to the fact that many processes show a high grade of redundancy and alternatives. Thus, it will remain a challenge for the future to identify in the n-dimensional space of pathogenesis those events, which are indeed key to the pathogenesis and, thus, can be subject of novel treatment strategies.

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References

1. Kuppers R. Mechanisms of B-cell lymphoma pathogenesis. Nat Rev Cancer. 2005;5(4):251–62. nrc1589 [pii].
2. Moriyama T, Metzger ML, Wu G, et al. Germline genetic variation in ETV6 and risk of childhood acute lymphoblastic leukaemia: a systematic genetic study. Lancet Oncol. 2015;16(16):1659–66. https://doi.org/10.1016/S1470-2045(15)00369-1.
3. Kuppers R. B cells under influence: transformation of B cells by Epstein-Barr virus. Nat Rev Immunol. 2003;3(10):801–12. https://doi.org/10.1038/nri1201.

4. Kulis M, Heath S, Bibikova M, et al. Epigenomic analysis detects widespread gene-body DNA hypomethylation in chronic lymphocytic leukemia. Nat Genet. 2012;44(11):1236–42. https://doi.org/10.1038/ng.2443.
5. Moss DJ, Lutzky VP. EBV-specific immune response: early research and personal reminiscences. Curr Top Microbiol Immunol. 2015;390(Pt 1):23–42. https://doi.org/10.1007/978-3-319-22822-8_3.
6. Kuppers R, Dalla-Favera R. Mechanisms of chromosomal translocations in B cell lymphomas. Oncogene. 2001;20(40):5580–94. https://doi.org/10.1038/sj.onc.1204640.
7. Lossos IS, Alizadeh AA, Eisen MB, et al. Ongoing immunoglobulin somatic mutation in germinal center B cell-like but not in activated B cell-like diffuse large cell lymphomas. Proc Natl Acad Sci U S A. 2000;97(18):10209–13. https://doi.org/10.1073/pnas.180316097.
8. Schroeder HW Jr, Cavacini L. Structure and function of immunoglobulins. J Allergy Clin Immunol. 2010;125(2 Suppl 2):S41–52. https://doi.org/10.1016/j.jaci.2009.09.046.
9. De Groot C, Kapsenberg ML, Leene W. Observations on transmembrane structures of surface immunoglobulin in the plasma membrane of B lymphocytes. Biochim Biophys Acta. 1982;689(2):275–82. 0005-2736(82)90260-7 [pii].
10. Kehry M, Ewald S, Douglas R, et al. The immunoglobulin mu chains of membrane-bound and secreted IgM molecules differ in their C-terminal segments. Cell. 1980;21(2):393–406. 0092-8674(80)90476-6 [pii].
11. Lieber MR, Hesse JE, Mizuuchi K, et al. Lymphoid V(D)J recombination: nucleotide insertion at signal joints as well as coding joints. Proc Natl Acad Sci U S A. 1988;85(22):8588–92.
12. van Gent DC, Ramsden DA, Gellert M. The RAG1 and RAG2 proteins establish the 12/23 rule in V(D)J recombination. Cell. 1996;85(1):107–13. S0092-8674(00)81086-7 [pii].
13. Tonegawa S. Somatic generation of antibody diversity. Nature. 1983;302(5909):575–81.
14. Levy NS, Malipiero UV, Lebecque SG, et al. Early onset of somatic mutation in immunoglobulin VH genes during the primary immune response. J Exp Med. 1989;169(6):2007–19.
15. Muramatsu M, Sankaranand VS, Anant S, et al. Specific expression of activation-induced cytidine deaminase (AID), a novel member of the RNA-editing deaminase family in germinal center B cells. J Biol Chem. 1999;274(26):18470–6.
16. Pasqualucci L, Guglielmino R, Houldsworth J, et al. Expression of the AID protein in normal and neoplastic B cells. Blood. 2004;104(10):3318–25. https://doi.org/10.1182/blood-2004-04-1558.
17. Lieber MR, Yu K, Raghavan SC. Roles of nonhomologous DNA end joining, V(D)J recombination, and class switch recombination in chromosomal translocations. DNA Repair (Amst). 2006;5(9–10):1234–45. S1568-7864(06)00154-6 [pii].
18. Yan CT, Boboila C, Souza EK, et al. IgH class switching and translocations use a robust non-classical end-joining pathway. Nature. 2007;449(7161):478–82. nature06020 [pii].
19. Honjo T, Kinoshita K, Muramatsu M. Molecular mechanism of class switch recombination: linkage with somatic hypermutation. Annu Rev Immunol. 2002;20:165–96. https://doi.org/10.1146/annurev.immunol.20.090501.112049.
20. Tsujimoto Y, Louie E, Bashir MM, et al. The reciprocal partners of both the t(14; 18) and the t(11; 14) translocations involved in B-cell neoplasms are rearranged by the same mechanism. Oncogene. 1988;2(4):347–51.
21. Tsujimoto Y, Gorham J, Cossman J, et al. The t(14;18) chromosome translocations involved in B-cell neoplasms result from mistakes in VDJ joining. Science. 1985;229(4720):1390–3.
22. Jager U, Bocskor S, Le T, et al. Follicular lymphomas' BCL-2/IgH junctions contain templated nucleotide insertions: novel insights into the mechanism of t(14;18) translocation. Blood. 2000;95(11):3520–9.
23. Medina KL, Singh H. Genetic networks that regulate B lymphopoiesis. Curr Opin Hematol. 2005;12(3):203–9. 00062752-200505000-00002 [pii].
24. Rajewsky K. Clonal selection and learning in the antibody system. Nature. 1996;381(6585):751–8. https://doi.org/10.1038/381751a0.
25. Corbett SJ, Tomlinson IM, Sonnhammer EL, et al. Sequence of the human immunoglobulin diversity (D) segment locus: a systematic analysis provides no evidence for the use of DIR segments, inverted D segments, "minor" D segments or D-D recombination. J Mol Biol. 1997;270(4):587–97. S0022-2836(97)91141-4 [pii].
26. Ravetch JV, Siebenlist U, Korsmeyer S, et al. Structure of the human immunoglobulin mu locus: characterization of embryonic and rearranged J and D genes. Cell. 1981;27(3 Pt 2):583–91. 0092-8674(81)90400-1 [pii].
27. Tiegs SL, Russell DM, Nemazee D. Receptor editing in self-reactive bone marrow B cells. J Exp Med. 1993;177(4):1009–20.
28. Hieter PA, Maizel JV Jr, Leder P. Evolution of human immunoglobulin kappa J region genes. J Biol Chem. 1982;257(3):1516–22.
29. Schable KF, Zachau HG. The variable genes of the human immunoglobulin kappa locus. Biol Chem Hoppe Seyler. 1993;374(11):1001–22.
30. Kawasaki K, Minoshima S, Nakato E, et al. One-megabase sequence analysis of the human immunoglobulin lambda gene locus. Genome Res. 1997;7(3):250–61.
31. Vasicek TJ, Leder P. Structure and expression of the human immunoglobulin lambda genes. J Exp Med. 1990;172(2):609–20.
32. Liu YJ, Zhang J, Lane PJ, et al. Sites of specific B cell activation in primary and secondary responses to T cell-dependent and T cell-independent antigens. Eur J Immunol. 1991;21(12):2951–62. https://doi.org/10.1002/eji.1830211209.
33. Reed JC. Bcl-2-family proteins and hematologic malignancies: history and future prospects. Blood. 2008;111(7):3322–30. https://doi.org/10.1182/blood-2007-09-078162.
34. Shaffer AL, Yu X, He Y, et al. BCL-6 represses genes that function in lymphocyte differentiation, inflammation, and cell cycle control. Immunity. 2000;13(2):199–212. S1074-7613(00)00020-0 [pii].
35. MacLennan IC. Germinal centers. Annu Rev Immunol. 1994;12:117–39. https://doi.org/10.1146/annurev.iy.12.040194.001001.
36. Allen CD, Ansel KM, Low C, et al. Germinal center dark and light zone organization is mediated by CXCR4 and CXCR5. Nat Immunol. 2004;5(9):943–52. https://doi.org/10.1038/ni1100.
37. Victora GD, Schwickert TA, Fooksman DR, et al. Germinal center dynamics revealed by multiphoton microscopy with a photoactivatable fluorescent reporter. Cell. 2010;143(4):592–605. https://doi.org/10.1016/j.cell.2010.10.032.
38. Goossens T, Klein U, Kuppers R. Frequent occurrence of deletions and duplications during somatic hypermutation: implications for oncogene translocations and heavy chain disease. Proc Natl Acad Sci U S A. 1998;95(5):2463–8.
39. Kuppers R, Zhao M, Hansmann ML, et al. Tracing B cell development in human germinal centres by molecular analysis of single cells picked from histological sections. EMBO J. 1993;12(13):4955–67.
40. Kosco-Vilbois MH. Are follcicular dendritic cells really good for nothing? Nat Rev Immunol. 2003;3:764–9.
41. Calado DP, Sasaki Y, Godinho SA, et al. The cell-cycle regulator c-Myc is essential for the formation and maintenance of germinal centers. Nat Immunol. 2012;13(11):1092–100. https://doi.org/10.1038/ni.2418.
42. Dominguez-Sola D, Victora GD, Ying CY, et al. The proto-oncogene MYC is required for selection in the germinal center and cyclic reentry. Nat Immunol. 2012;13(11):1083–91. https://doi.org/10.1038/ni.2428.
43. Cobaleda C, Schebesta A, Delogu A, et al. Pax5: the guardian of B cell identity and function. Nat Immunol. 2007;8(5):463–70. ni1454 [pii].

44. Falini B, Fizzotti M, Pucciarini A, et al. A monoclonal antibody (MUM1p) detects expression of the MUM1/IRF4 protein in a subset of germinal center B cells, plasma cells, and activated T cells. Blood. 2000;95(6):2084–92.
45. Angelin-Duclos C, Cattoretti G, Lin KI, et al. Commitment of B lymphocytes to a plasma cell fate is associated with Blimp-1 expression in vivo. J Immunol. 2000;165(10):5462–71.
46. Manz RA, Hauser AE, Hiepe F, et al. Maintenance of serum antibody levels. Annu Rev Immunol. 2005;23:367–86. https://doi.org/10.1146/annurev.immunol.23.021704.115723.
47. McHeyzer-Williams M, Okitsu S, Wang N, et al. Molecular programming of B cell memory. Nat Rev Immunol. 2011;12(1):24–34. https://doi.org/10.1038/nri3128.
48. Casamayor-Palleja M, Feuillard J, Ball J, et al. Centrocytes rapidly adopt a memory B cell phenotype on co-culture with autologous germinal centre T cell-enriched preparations. Int Immunol. 1996;8(5):737–44.
49. Han JH, Akira S, Calame K, et al. Class switch recombination and somatic hypermutation in early mouse B cells are mediated by B cell and Toll-like receptors. Immunity. 2007;27(1):64–75. S1074-7613(07)00330-5 [pii].
50. Mond JJ, Lees A, Snapper CM. T cell-independent antigens type 2. Annu Rev Immunol. 1995;13:655–92. https://doi.org/10.1146/annurev.iy.13.040195.003255.
51. Toellner KM, Jenkinson WE, Taylor DR, et al. Low-level hypermutation in T cell-independent germinal centers compared with high mutation rates associated with T cell-dependent germinal centers. J Exp Med. 2002;195(3):383–9.
52. Martin F, Kearney JF. B1 cells: similarities and differences with other B cell subsets. Curr Opin Immunol. 2001;13(2):195–201. S0952-7915(00)00204-1 [pii].
53. Lopes-Carvalho T, Kearney JF. Development and selection of marginal zone B cells. Immunol Rev. 2004;197:192–205. 112 [pii].
54. Oliver AM, Martin F, Gartland GL, et al. Marginal zone B cells exhibit unique activation, proliferative and immunoglobulin secretory responses. Eur J Immunol. 1997;27(9):2366–74. https://doi.org/10.1002/eji.1830270935.
55. Jacob J, Kassir R, Kelsoe G. In situ studies of the primary immune response to (4-hydroxy-3-nitrophenyl)acetyl. I. The architecture and dynamics of responding cell populations. J Exp Med. 1991;173(5):1165–75.
56. Kaji T, Ishige A, Hikida M, et al. Distinct cellular pathways select germline-encoded and somatically mutated antibodies into immunological memory. J Exp Med. 2012;209(11):2079–97. https://doi.org/10.1084/jem.20120127.
57. Klein U, Dalla-Favera R. Germinal centres: role in B-cell physiology and malignancy. Nat Rev Immunol. 2008;8(1):22–33. nri2217 [pii].
58. Traverse-Glehen A, Verney A, Baseggio L, et al. Analysis of BCL-6, CD95, PIM1, RHO/TTF and PAX5 mutations in splenic and nodal marginal zone B-cell lymphomas suggests a particular B-cell origin. Leukemia. 2007;21(8):1821–4. 2404706 [pii].
59. Alexandrov LB, Nik-Zainal S, Wedge DC, et al. Signatures of mutational processes in human cancer. Nature. 2013;500(7463):415–21. https://doi.org/10.1038/nature12477.
60. Wagener R, Alexandrov LB, Montesinos-Rongen M, et al. Analysis of mutational signatures in exomes from B-cell lymphoma cell lines suggest APOBEC3 family members to be involved in the pathogenesis of primary effusion lymphoma. Leukemia. 2015;29(7):1612–5. https://doi.org/10.1038/leu.2015.22.
61. Willis TG, Dyer MJ. The role of immunoglobulin translocations in the pathogenesis of B-cell malignancies. Blood. 2000;96(3):808–22.
62. Gascoyne RD, Lamant L, Martin-Subero JI, et al. ALK-positive diffuse large B-cell lymphoma is associated with Clathrin-ALK rearrangements: report of 6 cases. Blood. 2003;102(7):2568–73. https://doi.org/10.1182/blood-2003-03-0786.
63. Gesk S, Gascoyne RD, Schnitzer B, et al. ALK-positive diffuse large B-cell lymphoma with ALK-Clathrin fusion belongs to the spectrum of pediatric lymphomas. Leukemia. 2005;19(10):1839–40. 2403921 [pii].
64. Richter J, Schlesner M, Hoffmann S, et al. Recurrent mutation of the ID3 gene in Burkitt lymphoma identified by integrated genome, exome and transcriptome sequencing. Nat Genet. 2012;44(12):1316–20. https://doi.org/10.1038/ng.2469.
65. Schmidt J, Ramis-Zaldivar JE, Nadeu F, et al. Mutations of MAP2K1 are frequent in pediatric-type follicular lymphoma and result in ERK pathway activation. Blood. 2017;130(3):323–7. https://doi.org/10.1182/blood-2017-03-776278.
66. Morin RD, Mendez-Lago M, Mungall AJ, et al. Frequent mutation of histone-modifying genes in non-Hodgkin lymphoma. Nature. 2011;476(7360):298–303. https://doi.org/10.1038/nature10351.
67. Pasqualucci L, Dominguez-Sola D, Chiarenza A, et al. Inactivating mutations of acetyltransferase genes in B-cell lymphoma. Nature. 2011;471(7337):189–95. https://doi.org/10.1038/nature09730.
68. Oduor CI, Kaymaz Y, Chelimo K, et al. Integrative microRNA and mRNA deep-sequencing expression profiling in endemic Burkitt lymphoma. BMC Cancer. 2017;17(1):761-017-3711-9. https://doi.org/10.1186/s12885-017-3711-9.
69. Pasqualucci L, Neumeister P, Goossens T, et al. Hypermutation of multiple proto-oncogenes in B-cell diffuse large-cell lymphomas. Nature. 2001;412(6844):341–6. https://doi.org/10.1038/35085588.
70. Yano T, van Krieken JH, Magrath IT, et al. Histogenetic correlations between subcategories of small noncleaved cell lymphomas. Blood. 1992;79(5):1282–90.
71. Segal GH, Edinger MG, Owen M, et al. Concomitant delineation of surface Ig, B-cell differentiation antigens, and HLADR on lymphoid proliferations using three-color immunocytometry. Cytometry. 1991;12(4):350–9. https://doi.org/10.1002/cyto.990120410.
72. Gunven P, Klein G, Klein E, et al. Surface immunoglobulins on Burkitt's lymphoma biopsy cells from 91 patients. Int J Cancer. 1980;25(6):711–9.
73. Nussenzweig A, Nussenzweig MC. Origin of chromosomal translocations in lymphoid cancer. Cell. 2010;141(1):27–38. https://doi.org/10.1016/j.cell.2010.03.016.
74. de Jong D, Voetdijk BM, Van Ommen GJ, et al. Translocation t(14;18) in B cell lymphomas as a cause for defective immunoglobulin production. J Exp Med. 1989;169(3):613–24.
75. Horikawa K, Martin SW, Pogue SL, et al. Enhancement and suppression of signaling by the conserved tail of IgG memory-type B cell antigen receptors. J Exp Med. 2007;204(4):759–69. jem.20061923 [pii].
76. Martin SW, Goodnow CC. Burst-enhancing role of the IgG membrane tail as a molecular determinant of memory. Nat Immunol. 2002;3(2):182–8. https://doi.org/10.1038/ni752.
77. Dogan I, Bertocci B, Vilmont V, et al. Multiple layers of B cell memory with different effector functions. Nat Immunol. 2009;10(12):1292–9. https://doi.org/10.1038/ni.1814.
78. Staudt LM. A closer look at follicular lymphoma. N Engl J Med. 2007;356(7):741–2. 356/7/741 [pii].
79. Vaandrager JW, Schuuring E, Kluin-Nelemans HC, et al. DNA fiber fluorescence in situ hybridization analysis of immunoglobulin class switching in B-cell neoplasia: aberrant CH gene rearrangements in follicle center-cell lymphoma. Blood. 1998;92(8):2871–8.
80. Lenz G, Nagel I, Siebert R, et al. Aberrant immunoglobulin class switch recombination and switch translocations in activated B cell-like diffuse large B cell lymphoma. J Exp Med. 2007;204(3):633–43. jem.20062041 [pii].
81. Davis RE, Ngo VN, Lenz G, et al. Chronic active B-cell-receptor signalling in diffuse large B-cell lymphoma. Nature. 2010;463(7277):88–92. https://doi.org/10.1038/nature08638.
82. Chaby R, Szabo L. 3-Deoxy-2-octulosonic acid 5-phosphate: a component of the endotoxin of Bordetella pertussis. Eur J Biochem. 1975;59(1):277–80.

83. Ruminy P, Etancelin P, Couronne L, et al. The isotype of the BCR as a surrogate for the GCB and ABC molecular subtypes in diffuse large B-cell lymphoma. Leukemia. 2011;25(4):681–8. https://doi.org/10.1038/leu.2010.302.
84. Walter R, Pan KT, Doebele C, et al. HSP90 promotes Burkitt lymphoma cell survival by maintaining tonic B-cell receptor signaling. Blood. 2017;129(5):598–608. https://doi.org/10.1182/blood-2016-06-721423.
85. Zhu D, Ottensmeier CH, Du MQ, et al. Incidence of potential glycosylation sites in immunoglobulin variable regions distinguishes between subsets of Burkitt's lymphoma and mucosa-associated lymphoid tissue lymphoma. Br J Haematol. 2003;120(2):217–22. 4064 [pii].
86. Coelho V, Krysov S, Ghaemmaghami AM, et al. Glycosylation of surface Ig creates a functional bridge between human follicular lymphoma and microenvironmental lectins. Proc Natl Acad Sci U S A. 2010;107(43):18587–92. https://doi.org/10.1073/pnas.1009388107.
87. Radcliffe CM, Arnold JN, Suter DM, et al. Human follicular lymphoma cells contain oligomannose glycans in the antigen-binding site of the B-cell receptor. J Biol Chem. 2007;282(10):7405–15. M602690200 [pii].
88. Brauninger A, Spieker T, Mottok A, et al. Epstein-Barr virus (EBV)-positive lymphoproliferations in post-transplant patients show immunoglobulin V gene mutation patterns suggesting interference of EBV with normal B cell differentiation processes. Eur J Immunol. 2003;33(6):1593–602. https://doi.org/10.1002/eji.200323765.
89. Timms JM, Bell A, Flavell JR, et al. Target cells of Epstein-Barr-virus (EBV)-positive post-transplant lymphoproliferative disease: similarities to EBV-positive Hodgkin's lymphoma. Lancet. 2003;361(9353):217–23. S0140-6736(03)12271-4 [pii].
90. Capello D, Cerri M, Muti G, et al. Molecular histogenesis of posttransplantation lymphoproliferative disorders. Blood. 2003;102(10):3775–85. https://doi.org/10.1182/blood-2003-05-1683.
91. Ritz O, Leithauser F, Hasel C, et al. Downregulation of internal enhancer activity contributes to abnormally low immunoglobulin expression in the MedB-1 mediastinal B-cell lymphoma cell line. J Pathol. 2005;205(3):336–48. https://doi.org/10.1002/path.1688.
92. Pileri SA, Gaidano G, Zinzani PL, et al. Primary mediastinal B-cell lymphoma: high frequency of BCL-6 mutations and consistent expression of the transcription factors OCT-2, BOB.1, and PU.1 in the absence of immunoglobulins. Am J Pathol. 2003;162(1):243–53. S0002-9440(10)63815-1 [pii].
93. Leithauser F, Bauerle M, Huynh MQ, et al. Isotype-switched immunoglobulin genes with a high load of somatic hypermutation and lack of ongoing mutational activity are prevalent in mediastinal B-cell lymphoma. Blood. 2001;98(9):2762–70.
94. Carbone A, Gloghini A. KSHV/HHV8-associated lymphomas. Br J Haematol. 2008;140(1):13–24. BJH6879 [pii].
95. Burkitt DP. Classics in oncology. A sarcoma involving the jaws in African children. CA Cancer J Clin. 1972;22(6):345–55.
96. Zech L, Haglund U, Nilsson K, et al. Characteristic chromosomal abnormalities in biopsies and lymphoid-cell lines from patients with Burkitt and non-Burkitt lymphomas. Int J Cancer. 1976;17(1):47–56.
97. Gabarre J, Raphael M, Lepage E, et al. Human immunodeficiency virus-related lymphoma: relation between clinical features and histologic subtypes. Am J Med. 2001;111(9):704–11. S0002934301010208 [pii].
98. Young LS, Dawson CW, Eliopoulos AG. The expression and function of Epstein-Barr virus encoded latent genes. Mol Pathol. 2000;53(5):238–47.
99. Szeles A, Falk KI, Imreh S, et al. Visualization of alternative Epstein-Barr virus expression programs by fluorescent in situ hybridization at the cell level. J Virol. 1999;73(6):5064–9.
100. Crawford DH. Biology and disease associations of Epstein-Barr virus. Philos Trans R Soc Lond Ser B Biol Sci. 2001;356(1408):461–73. https://doi.org/10.1098/rstb.2000.0783.
101. Kilger E, Kieser A, Baumann M, et al. Epstein-Barr virus-mediated B-cell proliferation is dependent upon latent membrane protein 1, which simulates an activated CD40 receptor. EMBO J. 1998;17(6):1700–9. https://doi.org/10.1093/emboj/17.6.1700.
102. Narkhede M, Arora S, Ujjani C. Primary effusion lymphoma: current perspectives. Onco Targets Ther. 2018;11:3747–54. https://doi.org/10.2147/OTT.S167392.
103. Machida K, Cheng KT, Sung VM, et al. Hepatitis C virus induces a mutator phenotype: enhanced mutations of immunoglobulin and protooncogenes. Proc Natl Acad Sci U S A. 2004;101(12):4262–7. https://doi.org/10.1073/pnas.0303971101.
104. Lacroix A, Collot-Teixeira S, Mardivirin L, et al. Involvement of human herpesvirus-6 variant B in classic Hodgkin's lymphoma via DR7 oncoprotein. Clin Cancer Res. 2010;16(19):4711–21. https://doi.org/10.1158/1078-0432.CCR-10-0470.
105. Cavalli F, Isaacson PG, Gascoyne RD, et al. MALT lymphomas. Hematology Am Soc Hematol Educ Program. 2001:1:241–58.
106. Bayerdorffer E, Neubauer A, Rudolph B, et al. Regression of primary gastric lymphoma of mucosa-associated lymphoid tissue type after cure of Helicobacter pylori infection. MALT Lymphoma Study Group. Lancet. 1995;345(8965):1591–4. S0140-6736(95)90113-2 [pii].
107. Wotherspoon AC, Doglioni C, Diss TC, et al. Regression of primary low-grade B-cell gastric lymphoma of mucosa-associated lymphoid tissue type after eradication of Helicobacter pylori. Lancet. 1993;342(8871):575–7. 0140-6736(93)91409-F [pii].
108. Morgner A, Miehlke S, Fischbach W, et al. Complete remission of primary high-grade B-cell gastric lymphoma after cure of Helicobacter pylori infection. J Clin Oncol. 2001;19(7):2041–8. https://doi.org/10.1200/JCO.2001.19.7.2041.
109. Hermine O, Lefrere F, Bronowicki JP, et al. Regression of splenic lymphoma with villous lymphocytes after treatment of hepatitis C virus infection. N Engl J Med. 2002;347(2):89–94. https://doi.org/10.1056/NEJMoa013376.
110. Umetsu DT, Esserman L, Donlon TA, et al. Induction of proliferation of human follicular (B type) lymphoma cells by cognate interaction with CD4+ T cell clones. J Immunol. 1990;144(7):2550–7.
111. Johnson PW, Watt SM, Betts DR, et al. Isolated follicular lymphoma cells are resistant to apoptosis and can be grown in vitro in the CD40/stromal cell system. Blood. 1993;82(6):1848–57.
112. Relander T, Johnson NA, Farinha P, et al. Prognostic factors in follicular lymphoma. J Clin Oncol. 2010;28(17):2902–13. https://doi.org/10.1200/JCO.2009.26.1693.
113. Dave SS, Wright G, Tan B, et al. Prediction of survival in follicular lymphoma based on molecular features of tumor-infiltrating immune cells. N Engl J Med. 2004;351(21):2159–69. 351/21/2159 [pii].
114. Woo SR, Corrales L, Gajewski TF. Innate immune recognition of cancer. Annu Rev Immunol. 2015;33:445–74. https://doi.org/10.1146/annurev-immunol-032414-112043.
115. Chistiakov DA, Myasoedova VA, Revin VV, et al. The impact of interferon-regulatory factors to macrophage differentiation and polarization into M1 and M2. Immunobiology. 2018;223(1):101–11. S0171-2985(17)30140-7 [pii].
116. Horlad H, Ma C, Yano H, et al. An IL-27/Stat3 axis induces expression of programmed cell death 1 ligands (PD-L1/2) on infiltrating macrophages in lymphoma. Cancer Sci. 2016;107(11):1696–704. https://doi.org/10.1111/cas.13065.
117. Bannister AJ, Kouzarides T. Regulation of chromatin by histone modifications. Cell Res. 2011;21(3):381–95. https://doi.org/10.1038/cr.2011.22.
118. Kouzarides T. Chromatin modifications and their function. Cell. 2007;128(4):693–705. S0092-8674(07)00184-5 [pii].
119. Su IH, Basavaraj A, Krutchinsky AN, et al. Ezh2 controls B cell development through histone H3 methylation and Igh rearrangement. Nat Immunol. 2003;4(2):124–31. https://doi.org/10.1038/ni876.

120. Velichutina I, Shaknovich R, Geng H, et al. EZH2-mediated epigenetic silencing in germinal center B cells contributes to proliferation and lymphomagenesis. Blood. 2010;116(24):5247–55. https://doi.org/10.1182/blood-2010-04-280149.
121. Raaphorst FM, van Kemenade FJ, Fieret E, et al. Cutting edge: polycomb gene expression patterns reflect distinct B cell differentiation stages in human germinal centers. J Immunol. 2000;164(1):1–4. ji_v164n1p1 [pii].
122. Morin RD, Johnson NA, Severson TM, et al. Somatic mutations altering EZH2 (Tyr641) in follicular and diffuse large B-cell lymphomas of germinal-center origin. Nat Genet. 2010;42(2):181–5. https://doi.org/10.1038/ng.518.
123. Sneeringer CJ, Scott MP, Kuntz KW, et al. Coordinated activities of wild-type plus mutant EZH2 drive tumor-associated hypertrimethylation of lysine 27 on histone H3 (H3K27) in human B-cell lymphomas. Proc Natl Acad Sci U S A. 2010;107(49):20980–5. https://doi.org/10.1073/pnas.1012525107.
124. Yap DB, Chu J, Berg T, et al. Somatic mutations at EZH2 Y641 act dominantly through a mechanism of selectively altered PRC2 catalytic activity, to increase H3K27 trimethylation. Blood. 2011;117(8):2451–9. https://doi.org/10.1182/blood-2010-11-321208.
125. Mills AA. Throwing the cancer switch: reciprocal roles of polycomb and trithorax proteins. Nat Rev Cancer. 2010;10(10):669–82. https://doi.org/10.1038/nrc2931.
126. Arai S, Yoshimi A, Shimabe M, et al. Evi-1 is a transcriptional target of mixed-lineage leukemia oncoproteins in hematopoietic stem cells. Blood. 2011;117(23):6304–14. https://doi.org/10.1182/blood-2009-07-234310.
127. Okosun J, Bodor C, Wang J, et al. Integrated genomic analysis identifies recurrent mutations and evolution patterns driving the initiation and progression of follicular lymphoma. Nat Genet. 2014;46(2):176–81. https://doi.org/10.1038/ng.2856.
128. Pasqualucci L, Trifonov V, Fabbri G, et al. Analysis of the coding genome of diffuse large B-cell lymphoma. Nat Genet. 2011;43(9):830–7. https://doi.org/10.1038/ng.892.
129. Lohr JG, Stojanov P, Lawrence MS, et al. Discovery and prioritization of somatic mutations in diffuse large B-cell lymphoma (DLBCL) by whole-exome sequencing. Proc Natl Acad Sci U S A. 2012;109(10):3879–84. https://doi.org/10.1073/pnas.1121343109.
130. Zhang J, Dominguez-Sola D, Hussein S, et al. Disruption of KMT2D perturbs germinal center B cell development and promotes lymphomagenesis. Nat Med. 2015;21(10):1190–8. https://doi.org/10.1038/nm.3940.
131. Mullighan CG, Zhang J, Kasper LH, et al. CREBBP mutations in relapsed acute lymphoblastic leukaemia. Nature. 2011;471(7337):235–9. https://doi.org/10.1038/nature09727.
132. Hashwah H, Schmid CA, Kasser S, et al. Inactivation of CREBBP expands the germinal center B cell compartment, down-regulates MHCII expression and promotes DLBCL growth. Proc Natl Acad Sci U S A. 2017;114(36):9701–6. https://doi.org/10.1073/pnas.1619555114.
133. Cerchietti LC, Hatzi K, Caldas-Lopes E, et al. BCL6 repression of EP300 in human diffuse large B cell lymphoma cells provides a basis for rational combinatorial therapy. J Clin Invest. 2010;120(12):4569–82. https://doi.org/10.1172/JCI42869.
134. Oakes CC, Seifert M, Assenov Y, et al. DNA methylation dynamics during B cell maturation underlie a continuum of disease phenotypes in chronic lymphocytic leukemia. Nat Genet. 2016;48(3):253–64. https://doi.org/10.1038/ng.3488.
135. Kretzmer H, Bernhart SH, Wang W, et al. DNA methylome analysis in Burkitt and follicular lymphomas identifies differentially methylated regions linked to somatic mutation and transcriptional control. Nat Genet. 2015;47(11):1316–25. https://doi.org/10.1038/ng.3413.
136. O'Riain C, O'Shea DM, Yang Y, et al. Array-based DNA methylation profiling in follicular lymphoma. Leukemia. 2009;23(10):1858–66. https://doi.org/10.1038/leu.2009.114.
137. Bennett LB, Schnabel JL, Kelchen JM, et al. DNA hypermethylation accompanied by transcriptional repression in follicular lymphoma. Genes Chromosomes Cancer. 2009;48(9):828–41. https://doi.org/10.1002/gcc.20687.
138. Martin-Subero JI, Kreuz M, Bibikova M, et al. New insights into the biology and origin of mature aggressive B-cell lymphomas by combined epigenomic, genomic, and transcriptional profiling. Blood. 2009;113(11):2488–97. https://doi.org/10.1182/blood-2008-04-152900.
139. Leshchenko VV, Kuo PY, Shaknovich R, et al. Genomewide DNA methylation analysis reveals novel targets for drug development in mantle cell lymphoma. Blood. 2010;116(7):1025–34. https://doi.org/10.1182/blood-2009-12-257485.
140. Berdasco M, Esteller M. Aberrant epigenetic landscape in cancer: how cellular identity goes awry. Dev Cell. 2010;19(5):698–711. https://doi.org/10.1016/j.devcel.2010.10.005.
141. Shaknovich R, Cerchietti L, Tsikitas L, et al. DNA methyltransferase 1 and DNA methylation patterning contribute to germinal center B-cell differentiation. Blood. 2011;118(13):3559–69. https://doi.org/10.1182/blood-2011-06-357996.
142. Robaina MC, Mazzoccoli L, Arruda VO, et al. Deregulation of DNMT1, DNMT3B and miR-29s in Burkitt lymphoma suggests novel contribution for disease pathogenesis. Exp Mol Pathol. 2015;98(2):200–7. https://doi.org/10.1016/j.yexmp.2015.03.006.
143. Johansson B, Mertens F, Mitelman F. Cytogenetic evolution patterns in non-Hodgkin's lymphoma. Blood. 1995;86(10):3905–14. PMID: 7579360.
144. Kaymaz Y, Oduor CI, Yu H, et al. Comprehensive transcriptome and mutational profiling of endemic Burkitt lymphoma reveals EBV type-specific differences. Mol Cancer Res. 2017;15(5):563–76. https://doi.org/10.1158/1541-7786.MCR-16-0305.
145. Love C, Sun Z, Jima D, et al. The genetic landscape of mutations in Burkitt lymphoma. Nat Genet. 2012;44(12):1321–5. https://doi.org/10.1038/ng.2468.
146. Salaverria I, Martin-Guerrero I, Wagener R, Kreuz M, Kohler CW, Richter J, Pienkowska-Grela B, Adam P, Burkhardt B, Claviez A, Damm-Welk C, Drexler HG, Hummel M, Jaffe ES, Küppers R, Lefebvre C, Lisfeld J, Löffler M, Macleod RA, Nagel I, Oschlies I, Rosolowski M, Russell RB, Rymkiewicz G, Schindler D, Schlesner M, Scholtysik R, Schwaenen C, Spang R, Szczepanowski M, Trümper L, Vater I, Wessendorf S, Klapper W, Siebert R, Molecular Mechanisms in Malignant Lymphoma Network Project, Berlin-Frankfurt-Münster Non-Hodgkin Lymphoma Group. A recurrent 11q aberration pattern characterizes a subset of MYC-negative high-grade B-cell lymphomas resembling Burkitt lymphoma. Blood. 2014;123(8):1187–98. https://doi.org/10.1182/blood-2013-06-507996. Epub 2014 Jan 7. PMID:24398325.
147. Klapper W, Kreuz M, Kohler CW, et al. Patient age at diagnosis is associated with the molecular characteristics of diffuse large B-cell lymphoma. Blood. 2012;119(8):1882–7. https://doi.org/10.1182/blood-2011-10-388470.
148. Swerdlow S, Campo E, Harris N, et al. WHO classification of tumours of haematopoietic and lymphoid tissues. 4th ed. Lyon: International Agency for Research on Cancer; 2017.
149. Wessendorf S, Barth TF, Viardot A, et al. Further delineation of chromosomal consensus regions in primary mediastinal B-cell lymphomas: an analysis of 37 tumor samples using high-resolution genomic profiling (array-CGH). Leukemia. 2007;21(12):2463–9. 2404919 [pii].
150. Salaverria I, Martin-Guerrero I, Burkhardt B, et al. High resolution copy number analysis of IRF4 translocation-positive diffuse large B-cell and follicular lymphomas. Genes Chromosomes Cancer. 2013;52(2):150–5. https://doi.org/10.1002/gcc.22014.

Pathogenesis of T-Non-Hodgkin's Lymphoma

5

Jonathan Bond and Owen Patrick Smith

T-Lymphoblastic Lymphoma

T-lymphoblastic lymphoma (T-LBL) accounts for one fifth of all childhood and adolescent NHL and frequently presents with a mediastinal mass and advanced stage III/IV disease (Fig. 5.1) [1, 2]. Reported median age at diagnosis ranges from 7.0 to 10.5 years, with a male/female ratio of about 2.5:1 [3]. The current World Health Organization (WHO) classification lists T-acute lymphoblastic leukemia (T-ALL)/T-LBL as a single pathological category, with T-LBL diagnosis requiring that bone marrow infiltration by immature CD3-positive lymphoblasts is less than 25% [4]. Although there is ongoing debate as to whether T-ALL and T-LBL are truly distinct entities, or in fact represent heterogeneous clinical presentation of the same disease, the treatment of T-LBL has developed in parallel with childhood ALL strategies [5, 6].

Practical aspects of tissue accessibility mean that the molecular characterization of T-ALL is comparatively much more extensive, and discussion of T-LBL oncogenesis is therefore heavily informed by reference to its leukemic counterpart. In this section, we will describe the spectrum of genetic alterations in T-LBL, highlighting any known differences with T-ALL. These alterations can broadly be divided into structural abnormalities (including recurrent translocations and whole or partial chromosomal gain or loss), and somatically acquired mutations that typically affect T-lymphoid signaling pathways. We will also discuss published attempts to categorize T-LBL based on either T-receptor gene rearrangement status or transcriptional profiling, and detail the current attempts at defining a genetic risk classifier for pediatric T-LBL.

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Genetic Alterations in T-LBL

Translocations As in T-ALL, oncogene activation can occur as a result of rearrangements involving T-receptor genes that are located on chromosomes 7q34 (*TRB*), 7p14 (*TRG*), and 14q11 (*TRA/TRD*). T-receptor gene translocations are reported to occur in 18–44% of T-LBL [6–9], leading to aberrant expression of well-described T-lymphoid oncogenes, including loci that encode for homeobox-containing proteins (*HOXA9*, *TLX1*, *TLX3*) or T-specific transcription factors (*LYL1*, *LMO2*, *TAL1*) [10, 11]. It is believed that the cell-inappropriate activity of these molecules is directly linked to the oncogenic differentiation block that is found in both T-ALL and T-LBL. For example, TLX proteins have been shown to cause aberrant recruitment of ETS1 to the *TRA* enhancer, thereby inhibiting maturation beyond the thymic cortical developmental stage [12].

Translocations of chromosome 9q34 are more common in T-LBL than in T-ALL, and the t(9;17) (q34;q22–23) alteration has been reported in 2–15% of cases [7–9]. The 9q34 region contains several oncogenes that are known to be pathologically important in T-lymphoid malignancy, including *NOTCH1*, *ABL1*, *SET*, and *NUP214* [13–15], and the identity of the major molecular actor in this rearrangement is therefore not clear. Translocations that generate T-ALL-associated fusion transcripts such as *PICALM-MLLT10* and *NUP214-ABL1* have also been described in T-LBL. These exhibit similar correlation with phenotypic maturity as is seen in leukemia cases [11].

Deletions and loss of heterozygosity (LOH) Chromosome 6q LOH is found in both T-ALL and T-LBL, but the most

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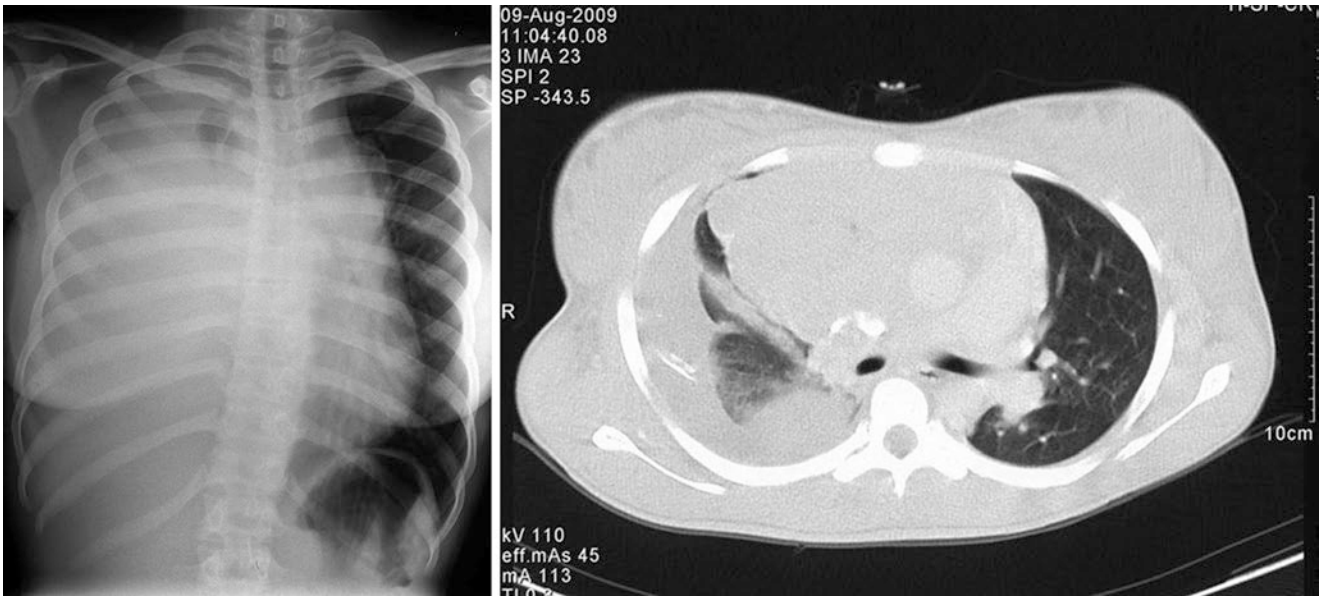

Fig. 5.1 Large mediastinal mass at T-lymphoblastic lymphoma presentation. Chest radiograph (left panel) and CT scan (right panel) showing a large lymphomatous mass with mediastinal shift, pericardial effusion, and right pleural effusion

commonly affected region differs between the diseases. In T-ALL, the 6q14–15 region is more frequently altered, whereas LOH of 6q16 is more common in T-LBL. Chromosome 6q LOH occurs in 12–19% of pediatric T-LBL and has been shown to correlate with poor outcome in several studies [16–18]. This region contains the *CASP8AP2* gene (also known as *FLASH*) that is believed to mediate glucocorticoid signaling [19, 20], and the prognostic effects of this alteration are therefore thought to be linked to impaired steroid treatment response.

Chromosome 9p deletions and LOH are reported in 11–47% of T-LBL [6, 21–23], which is marginally lower than the rates found in T-ALL [24]. This region contains the *CDKN2A* and *CDKN2B* genes that encode the cyclin-dependent kinases $p16^{INK4A}$ and $p15^{INK4B}$ and the p53-stabilizing protein $p14^{ARF}$. These loci are frequently altered in human cancers and are traditionally considered to contribute to oncogenesis by affecting the retinoblastoma and p53 pathways [25]. In a T-lymphoid context, *CDKN2A* inactivation has also been described to co-operate with both NOTCH1 signaling [26] and chromatin remodeling [27] to promote leukemogenesis.

Altered chromosome number This is also relatively common, being seen in 55–69% of pediatric and adolescent T-LBL [5–9]. Pseudodiploidy (25–44%) and hyperdiploidy (22–25%) are the most frequently reported numerical alterations, while hypodiploidy (3–5%) is rare.

NOTCH1 pathway activation Mutations in components of the NOTCH1 pathway are extremely common in acute T-lymphoid malignancies [28]. Genetic alterations cause constitutive NOTCH1 pathway signaling either through activating mutation of *NOTCH1* [13] or reduced function of the FBXW7 ubiquitin ligase that normally mediates NOTCH1 degradation [29]. *NOTCH1* mutations are found in 43–66% of pediatric T-LBL, and loss-of-function *FBXW7* alterations, which may or may not be concomitant, are reported in 18–21% of cases [11, 17, 18, 30]. While *NOTCH1* mutation with or without *FBXW7* mutation appears to be associated with good outcome, isolated *FBXW7* alteration has not to date been reported to alter prognosis [17, 18, 30].

Kinase signaling pathway abnormalities Activation of the PI3K-AKT signaling cascade can be caused by mutation in *PIK3R1*, *PIK3CA*, or *PTEN* [31, 32]. Of these, only *PTEN* alteration has been described to affect prognosis, although this effect was outweighed by the presence of concomitant *NOTCH1* mutation [32]. Mutations in *NRAS* or *KRAS* occur in about 10% of pediatric T-LBL, and unlike in T-ALL, have not to date been reported to correlate with outcome [32]. Activation of JAK-STAT signaling by *TEL-JAK2* translocation and *JAK2* mutations has also been described [33].

Other alterations Other rare structural genetic alterations include LOH of *ATM* and *TP53* [22] and localized deletions of chromosomes 12p13 or 17q11 [23]. The *ETV6-NCOA2* translocation that was previously described in early thymic precursor and biphenotypic ALL [34] was also reported in a single case of immature T-LBL [35]. Whole-exome profiling of five pediatric T-LBL samples identified multiple candidate mutations, many of which differed from those reported in T-ALL [17], and it is likely that future next-generation sequencing studies will identify further recurrent alterations.

Other Approaches to Categorization of T-LBL

T-receptor gene rearrangement status Immunophenotypic profiling and evaluation of the rearrangement status of T-receptor (TR) genes allows categorization of both T-ALL and T-LBL by resemblance to normal T-lymphoid ontogeny [11, 36]. Normal TR gene recombination is highly ordered, sequentially involving the *TRD*, *TRG*, *TRB*, and *TRA* loci [37, 38]. TR-based classification allows categorization of T-LBL into immature (cytoplasmic TCRβ-), intermediate (cytoplasmic TCRβ+ surface TCR−/+), and mature (surface TCR+ biallelic *TRD*-deleted) groups that correlates with expression of specific oncogenes [11]. For example, *TAL1* positivity co-segregates with mature T-LBL, whereas intermediate cases are more likely to express homeobox-containing genes such as *HOXA9* and *TLX1*. The intermediate T-LBL group has been suggested to have a relatively favorable prognosis, albeit in a series that included both pediatric and adult cases, where the latter comprised a high proportion of immature T-LBLs that had reduced survival [11].

Ontogenic immaturity as determined by absence of biallelic deletion (ABD) of the *TRG* locus has also been linked to T-LBL outcome. *TRG* ABD was originally described to predict poor prognosis in pediatric T-ALL [39], although subsequent implementation of minimal residual disease (MRD)-based treatment strategies and improved outcomes in treatment-resistant cases [40, 41] means that this prognostic link probably no longer pertains. The link between *TRG* deletion status with outcome in pediatric and adolescent T-LBL has been evaluated in one series. Although ABD was rare (4 of 53 cases), this was associated with a statistically significantly reduced survival [18].

Transcriptional profiling Microarray studies have identified recurrent patterns of gene expression in pediatric T-ALL, allowing reproducible categorization of leukemias according to a limited number of transcriptional profiles [42–44]. Comparative data in T-LBL is scarce, although efforts have been made to identify differentially expressed transcripts between T-ALL and T-LBL [6, 23, 45]. These studies have identified genes that are implicated in a diverse range of cellular function, including adhesion, caspase-mediated apoptosis, immune response genes, and regulation of transcription and protein biosynthesis. T-LBLs were reported to have increased expression of *KMT2A* and reduced expression of *CD47* when compared with T-ALL [45]. MicroRNA expression studies have also been performed, with miR223 being identified as a potential poor prognostic marker in one study [46].

Toward a Genetic Classifier for Risk Group Stratification in T-LBL

Children with T-LBL who fail front line chemotherapy have a dismal prognosis and those who relapse after treatment cessation can only be rescued by allogeneic stem cell transplantation. This has raised the question over the past three decades as to whether patients can be "risk-stratified" at diagnosis and thus allocate them to a specific treatment regimen whose intensity is modulated according to the risk of relapse, such that children who are predicted to have favorable outcomes receive lesser intensity regimens, sparing unwanted toxicities, while those with higher risk for treatment failure receive more intensive +/− experimental therapy.

Until recently there has been a paucity of molecular/genetic and prognostic factors in T-LBL, mainly due to the lack of suitable lymphoma material for such analysis. A small number of molecular retrospective studies in pediatric T-LBL have been published showing a correlation of clinical outcome with LOH6q, ABD, and mutations in *NOTCH1*, *FBXW7*, and *PTEN* (Table 5.1). More recently an international cooperative group comprising the Italian (AIEOP), the French (SFCE), and the German (BFM-D) study groups evaluated the potential of using genetic markers for T-LBL risk stratification. A consensus was reached that the mutational status of *NOTCH1* and *FBXW7* would be used in a new stratification system for prospective validation in an international cooperative treatment protocol for children and adolescents with lymphoblastic lymphoma [EudraCT number: 2017–001691-39], LBL 2018 trial. Patients with mutations in *NOTCH1* and/or *FBXW7* are stratified into the standard-risk group and those with *NOTCH1* and *FBXW7* germline status are stratified into the high-risk group. Patients without information on the mutational status of *NOTCH1* and *FBXW7* are stratified in the standard-risk group. Other molecular markers such as *PTEN*, ABD, LOH6q, *NRAS*, *KRAS*, *PIK3CA*, *PIK3R1*, and *FLASH* that have shown some prognostic relevance in children and adult T-LBL will be validated prospectively in the LBL 2018 trial.

Table 5.1 Prognostic genetic markers in pediatric T-LBL

Genetic marker	% of Cases	Prognostic impact	References
NOTCH1^{Mut} +/− *FBXW7*^{Mut}	50–60	Good	[11, 18, 23, 30, 47]
LOH 6q	10–15	Poor	[7, 16, 47]
PTEN altered	15	Poor	[32]
ABD	7	Poor	[18]

LOH loss of heterozygosity, *ABD* absence of biallelic deletion of the *TRG* locus

Peripheral T-Cell Lymphoma

The rare cases of non-lymphoblastic T-cell lymphomas in children and adolescents are categorized as peripheral T-cell lymphomas (PTCL), which is effectively a diagnostic umbrella that comprises 21 separate T-NHL subtypes in the latest WHO classification [4]. The rarity and heterogeneity of pediatric PTCL mean that meaningful epidemiological and pathological data are understandably scarce, although several groups have published retrospective analyses of national and international registry data [48–52]. Historical reports should be interpreted with caution in the light of evolutions in the diagnosis and molecular understanding of other disease subgroups. For example, modern pathological re-evaluation resulted in many cases originally diagnosed as PTCL to be re-categorized as either anaplastic large cell lymphoma (ALCL) or autoimmune lymphoproliferative syndrome (ALPS) [48].

The two largest published series on pediatric PTCL [48, 51] reported a median age at diagnosis of 11.1–12.6 years, with a male predominance of approximately 60%. As in adults, PTCL not otherwise specified (NOS) was the most frequent PTCL subgroup, comprising 42.0–47.4% of cases. Extranodal NK/T-cell lymphoma (14.7–23.7%), subcutaneous panniculitis-like T-cell lymphoma (13.2–14.0%), and hepatosplenic T-cell lymphoma (13.2–14.0%) were the next most common subsets, while primary cutaneous gamma-delta T-cell lymphoma (0.7–2.6%), angioimmunoblastic T-cell lymphoma (AITL) 0–2.8%, and mycosis fungoides (0–4.9%) were all extremely rare.

Of note, in the series reported by a European Intergroup for Childhood NHL (EICNHL) and international Berlin-Frankfurt-Münster (i-BFM) collaboration [51], a quarter of childhood and adolescent PTCL patients had pre-existing morbidity. This included a significant proportion who had received either hematopoietic stem cell or solid organ transplantation prior to PTCL diagnosis. Constitutional genetic disorders such as Nijmegen breakage syndrome, CATCH22 syndrome, and Trisomy 21 were also seen. This report also noted that these cases were more likely to have an unfavorable prognosis compared with the remainder of the pediatric PTCL cohort.

Comprehensive analysis of the molecular pathology of pediatric PTCL has also been hindered by disease rarity in this age group. PTCL in both adults and children has been shown to frequently harbor a complex karyotype [53, 54]. Reports of recurrent specific abnormalities are however lacking, although Isochromosome 7 and/or Trisomy 8 have been described to occur in most pediatric hepatosplenic T-cell lymphomas [48].

Advances in high-throughput sequencing technologies have permitted identification of a plethora of novel genetic alterations in PTCL in recent years, and this will hopefully lead to improved molecular classification of these diseases. Genes found to be affected by mutations include the guanine exchange factors *RHOA* [55–57] and *VAV1* [58, 59]; epigenetic factors *DNMT3A* [55, 57, 60], *TET2* [61], and *IDH2* [62]; and molecules involved in TCR signaling, including *CD28* [57, 63, 64]. In addition, RNA sequencing has revealed potentially therapeutically targetable kinase fusions [59], while recurrent rearrangements involving P53-related factors have also been detected [65]. The cohorts analyzed in these studies were overwhelmingly adult in nature, and any extrapolation to pediatric and adolescent PTCL should be made with caution.

References

1. You MJ, Medeiros LJ, Hsi ED. T-lymphoblastic leukemia/lymphoma. Am J Clin Pathol. 2015;144(3):411–22.
2. Smock KJ, Nelson M, Tripp SR, Sanger WG, Abromowitch M, Cairo MS, et al. Characterization of childhood precursor T-lymphoblastic lymphoma by immunophenotyping and fluorescent in situ hybridization: a report from the Children's Oncology Group. Pediatr Blood Cancer. 2008;51(4):489–94.
3. Burkhardt B, Mueller S, Khanam T, Perkins SL. Current status and future directions of T-lymphoblastic lymphoma in children and adolescents. Br J Haematol. 2016;173(4):545–59.
4. Swerdlow SH. Cancer IAfRo, organization WH. International Agency for Research on Cancer: WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues; 2008.
5. Burkhardt B. Paediatric lymphoblastic T-cell leukaemia and lymphoma: one or two diseases? Br J Haematol. 2010;149(5):653–68.
6. Uyttebroeck A, Vanhentenrijk V, Hagemeijer A, Boeckx N, Renard M, Wlodarska I, et al. Is there a difference in childhood T-cell acute lymphoblastic leukaemia and T-cell lymphoblastic lymphoma? Leuk Lymphoma. 2007;48(9):1745–54.
7. Burkhardt B, Bruch J, Zimmermann M, Strauch K, Parwaresch R, Ludwig WD, et al. Loss of heterozygosity on chromosome 6q14-q24 is associated with poor outcome in children and adolescents with T-cell lymphoblastic lymphoma. Leukemia. 2006;20(8):1422–9.
8. Lones MA, Heerema NA, Le Beau MM, Sposto R, Perkins SL, Kadin ME, et al. Chromosome abnormalities in advanced stage lymphoblastic lymphoma of children and adolescents: a report from CCG-E08. Cancer Genet Cytogenet. 2007;172(1):1–11.
9. Sekimizu M, Sunami S, Nakazawa A, Hayashi Y, Okimoto Y, Saito AM, et al. Chromosome abnormalities in advanced stage T-cell lymphoblastic lymphoma of children and adolescents: a report from Japanese Paediatric Leukaemia/Lymphoma Study Group (JPLSG) and review of the literature. Br J Haematol. 2011;154(5):612–7.
10. Cave H, Suciu S, Preudhomme C, Poppe B, Robert A, Uyttebroeck A, et al. Clinical significance of HOX11L2 expression linked to t(5;14)(q35;q32), of HOX11 expression, and of SIL-TAL fusion in childhood T-cell malignancies: results of EORTC studies 58881 and 58951. Blood. 2004;103(2):442–50.
11. Baleydier F, Decouvelaere AV, Bergeron J, Gaulard P, Canioni D, Bertrand Y, et al. T cell receptor genotyping and HOXA/TLX1 expression define three T lymphoblastic lymphoma subsets which might affect clinical outcome. Clin Cancer Res. 2008;14(3):692–700.
12. Dadi S, Le Noir S, Payet-Bornet D, Lhermitte L, Zacarias-Cabeza J, Bergeron J, et al. TLX homeodomain oncogenes mediate T cell mat-

uration arrest in T-ALL via interaction with ETS1 and suppression of TCRalpha gene expression. Cancer Cell. 2012;21(4):563–76.
13. Weng AP, Ferrando AA, Lee W, JPt M, Silverman LB, Sanchez-Irizarry C, et al. Activating mutations of NOTCH1 in human T cell acute lymphoblastic leukemia. Science. 2004;306(5694):269–71.
14. Van Vlierberghe P, van Grotel M, Tchinda J, Lee C, Beverloo HB, van der Spek PJ, et al. The recurrent SET-NUP214 fusion as a new HOXA activation mechanism in pediatric T-cell acute lymphoblastic leukemia. Blood. 2008;111(9):4668–80.
15. Graux C, Cools J, Melotte C, Quentmeier H, Ferrando A, Levine R, et al. Fusion of NUP214 to ABL1 on amplified episomes in T-cell acute lymphoblastic leukemia. Nat Genet. 2004;36(10):1084–9.
16. Burkhardt B, Moericke A, Klapper W, Greene F, Salzburg J, Damm-Welk C, et al. Pediatric precursor T lymphoblastic leukemia and lymphoblastic lymphoma: differences in the common regions with loss of heterozygosity at chromosome 6q and their prognostic impact. Leuk Lymphoma. 2008;49(3):451–61.
17. Bonn BR, Huge A, Rohde M, Oschlies I, Klapper W, Voss R, et al. Whole exome sequencing hints at a unique mutational profile of paediatric T-cell lymphoblastic lymphoma. Br J Haematol. 2015;168(2):308–13.
18. Callens C, Baleydier F, Lengline E, Ben Abdelali R, Petit A, Villarese P, et al. Clinical impact of NOTCH1 and/or FBXW7 mutations, FLASH deletion, and TCR status in pediatric T-cell lymphoblastic lymphoma. J Clin Oncol. 2012;30(16):1966–73.
19. Jun JI, Chung CW, Lee HJ, Pyo JO, Lee KN, Kim NS, et al. Role of FLASH in caspase-8-mediated activation of NF-kappaB: dominant-negative function of FLASH mutant in NF-kappaB signaling pathway. Oncogene. 2005;24(4):688–96.
20. Alm-Kristiansen AH, Saether T, Matre V, Gilfillan S, Dahle O, Gabrielsen OS. FLASH acts as a co-activator of the transcription factor c-Myb and localizes to active RNA polymerase II foci. Oncogene. 2008;27(34):4644–56.
21. Okuda T, Shurtleff SA, Valentine MB, Raimondi SC, Head DR, Behm F, et al. Frequent deletion of p16INK4a/MTS1 and p15INK4b/MTS2 in pediatric acute lymphoblastic leukemia. Blood. 1995;85(9):2321–30.
22. Krieger D, Moericke A, Oschlies I, Zimmermann M, Schrappe M, Reiter A, et al. Frequency and clinical relevance of DNA microsatellite alterations of the CDKN2A/B, ATM and p53 gene loci: a comparison between pediatric precursor T-cell lymphoblastic lymphoma and T-cell lymphoblastic leukemia. Haematologica. 2010;95(1):158–62.
23. Basso K, Mussolin L, Lettieri A, Brahmachary M, Lim WK, Califano A, et al. T-cell lymphoblastic lymphoma shows differences and similarities with T-cell acute lymphoblastic leukemia by genomic and gene expression analyses. Genes Chromosomes Cancer. 2011;50(12):1063–75.
24. Sulong S, Moorman AV, Irving JA, Strefford JC, Konn ZJ, Case MC, et al. A comprehensive analysis of the CDKN2A gene in childhood acute lymphoblastic leukemia reveals genomic deletion, copy number neutral loss of heterozygosity, and association with specific cytogenetic subgroups. Blood. 2009;113(1):100–7.
25. Lowe SW, Sherr CJ. Tumor suppression by Ink4a-Arf: progress and puzzles. Curr Opin Genet Dev. 2003;13(1):77–83.
26. Volanakis EJ, Boothby MR, Sherr CJ. Epigenetic regulation of the Ink4a-Arf (Cdkn2a) tumor suppressor locus in the initiation and progression of Notch1-driven T cell acute lymphoblastic leukemia. Exp Hematol. 2013;41(4):377–86.
27. Di Cello F, Dhara S, Hristov AC, Kowalski J, Elbahloul O, Hillion J, et al. Inactivation of the Cdkn2a locus cooperates with HMGA1 to drive T-cell leukemogenesis. Leuk Lymphoma. 2013;54(8):1762–8.
28. Sanchez-Martin M, Ferrando A. The NOTCH1-MYC highway toward T-cell acute lymphoblastic leukemia. Blood. 2017;129(9):1124–33.
29. Thompson BJ, Buonamici S, Sulis ML, Palomero T, Vilimas T, Basso G, et al. The SCFFBW7 ubiquitin ligase complex as a tumor suppressor in T cell leukemia. J Exp Med. 2007;204(8):1825–35.
30. Park MJ, Taki T, Oda M, Watanabe T, Yumura-Yagi K, Kobayashi R, et al. FBXW7 and NOTCH1 mutations in childhood T cell acute lymphoblastic leukaemia and T cell non-Hodgkin lymphoma. Br J Haematol. 2009;145(2):198–206.
31. Palomero T, Dominguez M, Ferrando AA. The role of the PTEN/AKT pathway in NOTCH1-induced leukemia. Cell Cycle. 2008;7(8):965–70.
32. Balbach ST, Makarova O, Bonn BR, Zimmermann M, Rohde M, Oschlies I, et al. Proposal of a genetic classifier for risk group stratification in pediatric T-cell lymphoblastic lymphoma reveals differences from adult T-cell lymphoblastic leukemia. Leukemia. 2016;30(4):970–3.
33. Roncero AM, Lopez-Nieva P, Cobos-Fernandez MA, Villa-Morales M, Gonzalez-Sanchez L, Lopez-Lorenzo JL, et al. Contribution of JAK2 mutations to T-cell lymphoblastic lymphoma development. Leukemia. 2016;30(1):94–103.
34. Strehl S, Nebral K, Konig M, Harbott J, Strobl H, Ratei R, et al. ETV6-NCOA2: a novel fusion gene in acute leukemia associated with coexpression of T-lymphoid and myeloid markers and frequent NOTCH1 mutations. Clin Cancer Res. 2008;14(4):977–83.
35. Bond J, Touzart A, Nadal N, Trinquand A, Thouvenin S, Da Cruz V, et al. Early thymic precursor-like lymphomatous presentation of the ETV6-NCOA2 translocation. Br J Haematol. 2017;181:392–4.
36. Asnafi V, Beldjord K, Boulanger E, Comba B, Le Tutour P, Estienne MH, et al. Analysis of TCR, pT alpha, and RAG-1 in T-acute lymphoblastic leukemias improves understanding of early human T-lymphoid lineage commitment. Blood. 2003;101(7):2693–703.
37. Spits H. Development of alphabeta T cells in the human thymus. Nat Rev Immunol. 2002;2(10):760–72.
38. Blom B, Verschuren MC, Heemskerk MH, Bakker AQ, van Gastel-Mol EJ, Wolvers-Tettero IL, et al. TCR gene rearrangements and expression of the pre-T cell receptor complex during human T-cell differentiation. Blood. 1999;93(9):3033–43.
39. Gutierrez A, Dahlberg SE, Neuberg DS, Zhang J, Grebliunaite R, Sanda T, et al. Absence of biallelic TCRgamma deletion predicts early treatment failure in pediatric T-cell acute lymphoblastic leukemia. J Clin Oncol. 2010;28(24):3816–23.
40. Patrick K, Wade R, Goulden N, Mitchell C, Moorman AV, Rowntree C, et al. Outcome for children and young people with Early T-cell precursor acute lymphoblastic leukaemia treated on a contemporary protocol, UKALL 2003. Br J Haematol. 2014;166(3):421–4.
41. Wood BL, Winter SS, Dunsmore KP, Devidas M, Chen S, Asselin B, et al. T-lymphoblastic leukemia (T-ALL) shows excellent outcome, lack of significance of the early Thymic precursor (ETP) Immunophenotype, and validation of the prognostic value of end-induction minimal residual disease (MRD) in Children's oncology group (COG) Study AALL0434. Blood. 2014;124(21):1.
42. Ferrando AA, Neuberg DS, Staunton J, Loh ML, Huard C, Raimondi SC, et al. Gene expression signatures define novel oncogenic pathways in T cell acute lymphoblastic leukemia. Cancer Cell. 2002;1(1):75–87.
43. Soulier J, Clappier E, Cayuela JM, Regnault A, Garcia-Peydro M, Dombret H, et al. HOXA genes are included in genetic and biologic networks defining human acute T-cell leukemia (T-ALL). Blood. 2005;106(1):274–86.
44. Homminga I, Pieters R, Langerak AW, de Rooi JJ, Stubbs A, Verstegen M, et al. Integrated transcript and genome analyses reveal NKX2-1 and MEF2C as potential oncogenes in T cell acute lymphoblastic leukemia. Cancer Cell. 2011;19(4):484–97.
45. Raetz EA, Perkins SL, Bhojwani D, Smock K, Philip M, Carroll WL, et al. Gene expression profiling reveals intrinsic differences between T-cell acute lymphoblastic leukemia and T-cell lymphoblastic lymphoma. Pediatr Blood Cancer. 2006;47(2):130–40.

46. Mussolin L, Holmes AB, Romualdi C, Sales G, D'Amore ES, Ghisi M, et al. An aberrant microRNA signature in childhood T-cell lymphoblastic lymphoma affecting CDKN1B expression, NOTCH1 and growth factor signaling pathways. Leukemia. 2014;28(9):1909–12.
47. Bonn BR, Rohde M, Zimmermann M, Krieger D, Oschlies I, Niggli F, et al. Incidence and prognostic relevance of genetic variations in T-cell lymphoblastic lymphoma in childhood and adolescence. Blood. 2013;121(16):3153–60.
48. Kontny U, Oschlies I, Woessmann W, Burkhardt B, Lisfeld J, Salzburg J, et al. Non-anaplastic peripheral T-cell lymphoma in children and adolescents--a retrospective analysis of the NHL-BFM study group. Br J Haematol. 2015;168(6):835–44.
49. Kobayashi R, Yamato K, Tanaka F, Takashima Y, Inada H, Kikuchi A, et al. Retrospective analysis of non-anaplastic peripheral T-cell lymphoma in pediatric patients in Japan. Pediatr Blood Cancer. 2010;54(2):212–5.
50. Windsor R, Stiller C, Webb D. Peripheral T-cell lymphoma in childhood: population-based experience in the United Kingdom over 20 years. Pediatr Blood Cancer. 2008;50(4):784–7.
51. Mellgren K, Attarbaschi A, Abla O, Alexander S, Bomken S, Bubanska E, et al. Non-anaplastic peripheral T cell lymphoma in children and adolescents-an international review of 143 cases. Ann Hematol. 2016;95(8):1295–305.
52. Hutchison RE, Laver JH, Chang M, Muzzafar T, Desai S, Murphy S, et al. Non-anaplastic peripheral t-cell lymphoma in childhood and adolescence: a Children's oncology group study. Pediatr Blood Cancer. 2008;51(1):29–33.
53. Lepretre S, Buchonnet G, Stamatoullas A, Lenain P, Duval C, d'Anjou J, et al. Chromosome abnormalities in peripheral T-cell lymphoma. Cancer Genet Cytogenet. 2000;117(1):71–9.
54. Nelson M, Horsman DE, Weisenburger DD, Gascoyne RD, Dave BJ, Loberiza FR, et al. Cytogenetic abnormalities and clinical correlations in peripheral T-cell lymphoma. Br J Haematol. 2008;141(4):461–9.
55. Sakata-Yanagimoto M, Enami T, Yoshida K, Shiraishi Y, Ishii R, Miyake Y, et al. Somatic RHOA mutation in angioimmunoblastic T cell lymphoma. Nat Genet. 2014;46(2):171–5.
56. Yoo HY, Sung MK, Lee SH, Kim S, Lee H, Park S, et al. A recurrent inactivating mutation in RHOA GTPase in angioimmunoblastic T cell lymphoma. Nat Genet. 2014;46(4):371–5.
57. Palomero T, Couronne L, Khiabanian H, Kim MY, Ambesi-Impiombato A, Perez-Garcia A, et al. Recurrent mutations in epigenetic regulators, RHOA and FYN kinase in peripheral T cell lymphomas. Nat Genet. 2014;46(2):166–70.
58. Abate F, da Silva-Almeida AC, Zairis S, Robles-Valero J, Couronne L, Khiabanian H, et al. Activating mutations and translocations in the guanine exchange factor VAV1 in peripheral T-cell lymphomas. Proc Natl Acad Sci U S A. 2017;114(4):764–9.
59. Boddicker RL, Razidlo GL, Dasari S, Zeng Y, Hu G, Knudson RA, et al. Integrated mate-pair and RNA sequencing identifies novel, targetable gene fusions in peripheral T-cell lymphoma. Blood. 2016;128(9):1234–45.
60. Dobay MP, Lemonnier F, Missiaglia E, Bastard C, Vallois D, Jais JP, et al. Integrative clinicopathological and molecular analyses of angioimmunoblastic T-cell lymphoma and other nodal lymphomas of follicular helper T-cell origin. Haematologica. 2017;102(4):e148–e51.
61. Lemonnier F, Couronne L, Parrens M, Jais JP, Travert M, Lamant L, et al. Recurrent TET2 mutations in peripheral T-cell lymphomas correlate with TFH-like features and adverse clinical parameters. Blood. 2012;120(7):1466–9.
62. Cairns RA, Iqbal J, Lemonnier F, Kucuk C, de Leval L, Jais JP, et al. IDH2 mutations are frequent in angioimmunoblastic T-cell lymphoma. Blood. 2012;119(8):1901–3.
63. Rohr J, Guo S, Huo J, Bouska A, Lachel C, Li Y, et al. Recurrent activating mutations of CD28 in peripheral T-cell lymphomas. Leukemia. 2016;30(5):1062–70.
64. Vallois D, Dobay MP, Morin RD, Lemonnier F, Missiaglia E, Juilland M, et al. Activating mutations in genes related to TCR signaling in angioimmunoblastic and other follicular helper T-cell-derived lymphomas. Blood. 2016;128(11):1490–502.
65. Vasmatzis G, Johnson SH, Knudson RA, Ketterling RP, Braggio E, Fonseca R, et al. Genome-wide analysis reveals recurrent structural abnormalities of TP63 and other p53-related genes in peripheral T-cell lymphomas. Blood. 2012;120(11):2280–9.

The Pathogenesis of Anaplastic Large Cell Lymphoma

6

Suzanne Dawn Turner

Introduction

Anaplastic large cell lymphoma (ALCL) is considered a peripheral T cell lymphoma predominantly affecting children and young adults, particularly when associated with the aberrant expression of anaplastic lymphoma kinase (ALK) fusion proteins [1, 2]. *ALK* was an unknown gene until cloned from cases of ALCL and reported in the context of a chromosomal translocation, the t(2;5)(p23;q35) [3]. Many ALK fusion protein variants have since been reported, but the Nucleophosmin (NPM)-ALK resulting from the aforementioned translocation remains the predominant version [4–6]. Rare cases of ALK-negative ALCL have also been reported in children but are more often seen in adults with a comparatively worse prognosis [7]. As well as systemic ALCL, ALK+ or ALK-, other categories of ALCL also exist including cutaneous and breast implant-associated forms although the latter are ALK- and largely affect adults [8–10]. Whether all forms of ALCL share a common origin or are derived from distinct cell types converging on a shared histopathology is unknown.

Cell of Origin

Non-Hodgkin lymphoma (NHL) encompasses a multitude of distinct disease entities largely categorised based on their presumed cellular origin. At its most basic level, whether they are of a B, T or natural killer (NK) cell origin. Within these subclasses exists an array of subtypes largely distinguished according to growth pattern, location, defining genetic abnormalities but most prominently the stage of lymphoid cell ontogeny that they most resemble. The latter in particular applies to anaplastic large cell lymphoma (ALCL), a malignancy classified as a peripheral T cell lymphoma due to systemic involvement and molecular rearrangements of T cell receptor (TCR) genes suggestive of a mature T cell origin [10]. However, whether the cell of origin bears any resemblance to the presenting cell type of the established malignancy or if this final identity was shaped by the pathogenic events driving tumour development is not known.

Immunohistological Features of ALCL Indicative of the Cell of Origin

Whilst at the molecular level, TCR rearrangements can be detected within ALCL, the TCR is not expressed on the cell surface [11, 12]. Instead, a combination of cell surface proteins is identifiable, some in keeping with a T cell identity albeit a ‘confused’ one. For example, expression of CD4 is detected on many ALCL yet often together with the production of cytotoxic proteins such as perforin and granzyme B, the former being indicative of a helper T cell and the latter a cytotoxic one (Fig. 6.1). Whilst we like to compartmentalise cell types, it is likely that there is immense plasticity amongst T cell subsets although expression of either CD4 or CD8 is determined early during thymic development; CD4 and CD8 are both expressed on primitive thymocytes prior to positive selection at which point either CD4 or CD8 is downregulated to generate helper or cytotoxic T cells, respectively [13]. This process is dependent on the recognition of either MHC class II or I presented on thymic epithelial cells in the cortex [13]. As such, the predominant expression of CD4 together with the production of cytotoxic proteins brings into question the cell type most likely to be the precursor of ALCL. One explanation could be that these cells are derived from helper, CD4-expressing T cells but that cytotoxic protein production is an artefact of the transformation process. Indeed, it has been demonstrated that NPM-ALK, the driving event in ALCL, can induce production of these proteins [14].

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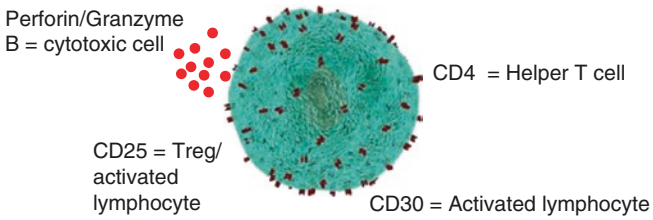

Fig. 6.1 The 'confused' immunophenotype of ALCL. ALCL are pleomorphic presenting with a variety of histological subtypes, but the majority of cells are positive for CD30, which does not distinguish lymphoid lineage but is suggestive of cell activation as is positivity for the epithelial membrane antigen (EMA) and CD25 (although co-expression of CD4 and CD25 with the transcription factor FoxP3 can be indicative of a regulatory T cell). In contrast, CD8 is rarely expressed, yet the majority of tumours produce the cytotoxic proteins perforin and/or granzyme B yet most cases are positive for CD4. CD3 is not expressed in the majority of cases presenting an overall 'confused' cell immuno-identity

As such, in contrast to the accepted dogma of the World Health Organization classification of lymphoid neoplasms, it is perhaps more likely that the normal cell counterpart is a helper rather than a cytotoxic T cell [10]. Other immunological markers include CD30, a protein expressed transiently in the thymus and then on activated lymphocytes, although NPM-ALK activity has also been shown to be responsible for inducing expression of this protein too [15, 16]. Of course, none of these cell surface proteins when considered in isolation can ultimately define the cell or origin. Therefore, apart from molecular TCR rearrangements, there is little certainty in predicting the cell of origin by immunophenotypic markers. An alternative approach is to examine the gene expression profile of tumour cells and to compare this to that of distinct, normal cell counterparts.

Genetic Features of ALCL Indicative of the Cell of Origin

Studies performed comparing the gene expression profile of ALCL tumour cells to clearly defined normal T cell subsets have been unable to assign a specific T cell identity to ALCL (the gene expression profile of ALCL does not cluster with CD4- nor CD8-positive T cells), although more recently gene set enrichment analysis has proposed a Th17 origin [17–19]. However, whether the Th17 gene expression profile is a remnant of the cell of origin or a cell phenotype induced by NPM-ALK is still an open question [18, 20]. Indeed, a regulatory T cell (Treg) origin has also been proposed due to co-expression of CD4 and CD25 on tumour cells together with production of IL10 and TGFβ and expression of FoxP3, although again, this phenotype has been shown to be a consequence of NPM-ALK-induced STAT3 activity particularly with regard to the latter three elements [21] and CD25 can also be indicative of activation status. T helper cells come in many 'flavours' and plasticity between these is dependent on a host of microenvironmental factors, key amongst which is the presence of a distinct profile of cytokines and the cell types that produce them [22] (Table 6.1). For example, interconvertibility between Treg and Th17 cells is particularly evident in autoimmunity and is dependent of the availability of the cytokines IL6 and LIF [23].

Why Is the Identity of the Cell of Origin Important to Know?

Knowing the identity of the cell of origin and its normal physiological function, hypotheses can be constructed as to the pathogenesis and origins of this distinct lymphoma entity. For example, Th1 cells are identified in the context of responses to intracellular pathogens, whereas Th2 cells are present when extracellular pathogens are present in their environment. A Th17 origin would therefore be more indicative of large, extracellular pathogens such as bacteria and/or autoimmune conditions and associated with the presence of neutrophils [24].

Naturally, when a diagnostic pathologist examines a tumour specimen, they are looking at the consequences of an evolutionary process whereby the fittest clone of tumour cells survives and propagates. Hence, the immunophenotype of the established growth may be the consequence of a combination of events occurring during the course of tumour development, with retainment of those properties most conducive to cell survival in their imposed surroundings, presumably those that facilitate acquisition of the so-called hallmarks of cancer [25].

Dissecting Tumour Heterogeneity to Pinpoint the Cell of Origin

It is now accepted that not all cells within a tumour are created equally and that some are 'fitter' than others; whether this be the consequence of clonal evolution whereby in a stochastic process, clones of cells derived from genetically related progenitors acquire properties to improve their survival odds, or a hierarchical process in which much like normal tissue development, a small subset of progenitor cells have the ability to self-renew and differentiate into progeny that later lose this ability. Hence, the phrase cancer stem cell representing a cell from which all tumour cell progeny derive. This terminology elicits ideas of transformed tissue-specific stem cells representing the cells of origin for malignancies, although of course this is not necessarily the case, as any cell that can acquire stemness properties, even if that cell was originally terminally differentiated, could also enact this

Table 6.1 There is immense plasticity in the lymphoid lineage particularly amongst helper T cell subsets whereby cell identity can be affected by the profile of lineage-skewing cytokines to which the cell is exposed in its microenvironment. The result is induced expression of distinct master transcription factors together with production of a profile of effector cytokines ultimately altering the role played by the T cell in an inflammatory response and the target cells with which they interact

Cell type	Tfh	Treg	Tr1	Th1	Th2	Th9	Th17	Th22	ILC3
Early response factor	**STAT3**	STAT5	**STAT3**/5	**STAT1**/4	STAT6	STAT6	**STAT3**	**STAT3**	RORgt
Skewing cytokines	***IL6,*** IL21, IL27	***TGFβ,*** IL2	***IL10,*** IL27	IL12, *IFNγ,* IL18, IL27	IL2, IL4, IL33	IL4, ***TGFβ***	***TGFβ, IL6,*** IL21	**IL6,** IL23, *TNFα*	IL1b, IL23
Effector cytokine	***IL6, IL10,*** IL21	***IL10, TGFβ***, IL35	***IL10, TGFβ***	IL2, *IFNg, TNFa*	IL3, IL4, IL5, ***IL6, IL10,*** IL13, IL25, IL31	***IL9, IL10***	***IL17A, IL17F,*** IL21, *IL22,* **IL26**	IL13, *IL22*	***IL17***, *IL22*
Role/ pathological effects	B cell helps in humoral immunity	Immunosuppression, tolerance	Immunosuppression, tolerance	Antigen presentation, cellular immunity, intracellular pathogens	Humoral immunity, allergies, extracellular parasites	Mucosal immunity, autoimmunity, tissue inflammation, allergies	Tissue inflammation, extracellular pathogens, autoimmunity	Tissue inflammation, allergies	Intestinal homeostasis
Defining transcription factor	Bcl6	**FoxP3**	cMaf, AhR	T-bet	Gata3, Maf1	PU.1, IRF4	**RORγt,** AhR	AhR	**RORγt,** AhR
Target cells	B cells	All other T cells	All other T cells	Macrophages, dendritic cells	Eosinophils, basophils	Mast cells, neutrophils, eosinophils, B cells, T cells	Neutrophils	Epithelial cells, stromal cells	Epithelial cells

Bold, italicised cytokines are those that have been significantly identified in the serum of paediatric ALCL patients, whereas italicisation alone indicates those with a trend towards significance. Transcription factors in bold type are those previously reported to be expressed in ALCL

Tfh follicular helper T cell, *Treg* regulatory T cell, *TR*1 type 1 inducible Treg, *Th* helper T cell, *ILC*3 innate lymphoid cell type 3

role. More preferable is the terminology tumour propagating cell (TPC) better reflective of the perhaps acquired rather than innate properties of stemness a tumour cell may have [26]. With regard to ALCL, the TPC has been identified as sharing gene expression signatures with early thymic progenitors (ETP) or haemopoietic stem cells (HSC) suggestive of a thymic or early haemopoietic origin [27].

The ALCL TPC Is a Primitive Haemopoietic Cell

Tissue-specific stem cells are traditionally identified by their functional properties, i.e. presumed quiescence, ability to give rise to differentiated progeny and the ability to self-renew. More often than not, specific cell surface proteins or Cluster of Differentiation (CD) markers are then assigned to facilitate their identification. As an adaption of this, TPC have been discovered in a number of cancers, most prominently the leukaemias by looking for cells within tumours that express CD proteins associated with stem cells [28]. An alternative approach when cells with TPC properties cannot be identified in this manner is to exploit the functional properties of these cells. In this regard, the side population (SP) technique detects TPC through their relative quiescence and ability to efflux dyes through ABCG2 transporters [29, 30]. Employing this technique, the TPC for ALCL was identified, and whilst no specific CD proteins could be exclusively assigned to these cells, the fact that they gave rise to progeny lacking self-renewal unable to sustain tumour growth in vitro nor in vivo was defined as the TPC for ALCL. Given the lack of expression of cell of origin-defining CD proteins by the ALCL SP cells, gene expression profiling was performed and matched to defined haemopoietic cell subsets by gene set enrichment analysis. These data showed that the ALCL TPC most resembled an ETP or HSC with regard to its gene expression profile [27]. Hence was born the hypothesis that ALCL might initiate in these specific cellular subsets. Given that ALCL is a PTCL with a potential thymic (ETP) or bone marrow (HSC) origin, events in the thymus might also provide clues as to the pathogenesis of this disease. Rather uniquely amongst different cell types, the lymphoid lineage, by nature of its requirement to recognise foreign antigen in the context of self-antigens, an ordered educational process whereby TCR genes are rearranged, provides a timestamp of developmental processes that again may provide further evidence as to the true cell of origin for ALCL.

TCR Gene Rearrangements Provide a History of Tumour Cell Development

TCR gene rearrangement takes place in an ordered manner whereby TCR genes are rearranged in a sequence within developing thymocytes resident in the thymus (Fig. 6.2). This highly coordinated process commences in ETP whereby rearrangement of the TCRδ chain genes precedes TCRγ, followed by TCRβ and ending in TCRα at which point the TCRδ genes are deleted as they are located within the TCRα locus. As this occurs, the thymocytes mature into T cells which exit into the peripheral circulation having survived the processes of β-selection as well as positive and negative selection. As such, it is possible to delineate the history of tumour cells by examining the status of the TCR genes. For ALCL, ALK+, tumour cells carry a range of TCR gene rearrangements not conducive with surviving the thymic environment (Fig. 6.2). For example, many ALCL have major clonal TCRα rearrangements but lack an equivalent TCRβ clone [12]. Thymocytes in this situation would not normally pass the β-selection checkpoint instead undergoing apoptosis and therefore not surviving to rearrange the TCRα chain genes. The fact that tumour cells exist with these abnormal TCR gene rearrangements is indicative of some other activity permissive of surviving the thymic environment to emerge into the periphery as a 'mature' T cell. Indeed, murine models show that NPM-ALK can allow thymocytes to escape β-selection [12]. An alternative explanation would be ongoing VDJ recombination in the periphery although RAG is not expressed in ALCL and this activity would be unable to return VDJ gene segments to their wild-type confirmation [12]. These data also suggest that some of these tumour cells may not have had the capacity to express a cell surface, functional TCR in the first place in keeping with the lack of expression of a TCR on the surface of tumour cells [11]. Assuming these incipient tumour cells escape the thymic environment to enter the peripheral circulation, this then raises the question as to whether events in the periphery contribute to the pathogenesis of this disease.

Unravelling the Pathogenesis of ALCL

Aetiological Associations

Despite a Th17 phenotype being indicative of large extracellular pathogens, there is no documented aetiological association for ALCL. A few case reports have reported systemic ALCL with cutaneous presentation in the context of tick or other insect bites although there is no evidence that these bites led to infections that drove hyper-proliferation and transformation of cells [31, 32]. An alternative explanation would be the homing of tumour cells to these sites of inflammation, mediated by cytokines. However, many children present with B symptoms including fever, but again, whether these are related to the cause of or are a consequence of tumour development is difficult to decipher. Given the differential proposed cytotoxic cell of origin, one might consider viral pathogenesis although viral sequences such as those belonging to EBV have not been detected in ALCL, but one

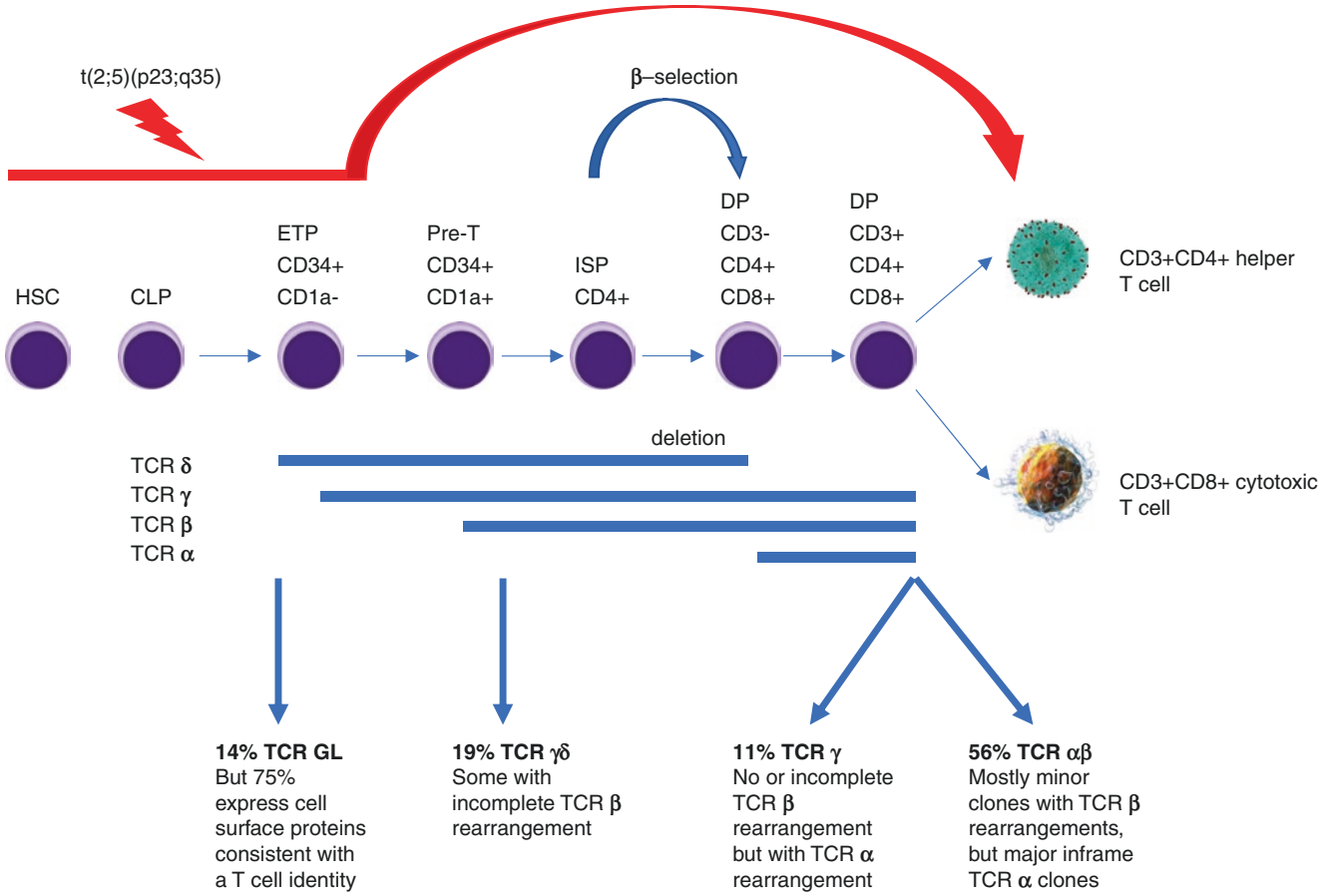

Fig. 6.2 Aberrant thymic events are indicative of a primitive haemopoietic origin for ALCL. The gene expression profile of the TPC driving ALCL is suggestive of an origin in an HSC or ETP. In further support, TCR gene rearrangements are detectable not conducive with thymocyte survival within the harsh thymic environment, whereby selection processes operate to generate functional T cells that recognise non-self but not self-antigens presented with MHC. The t(2;5)(p23;q35)-expressing thymocytes must exit into the periphery to present as a peripheral T cell lymphoma in patients, suggesting that the product of the translocation NPM-ALK facilitates bypass of thymic selection processes as evidenced in murine models. HSC haemopoietic stem cell, CLP common lymphoid progenitor, ETP early thymic precursor, DP double positive, ISP intermediate single positive, TCR T cell receptor

cannot discount a hit-and-run mechanism whereby virally infected B cells drive the initial T cell proliferation during which genetic mutations are acquired that render the incipient tumour cells relatively autonomous for growth [33]. Again, with no evidence observed in the established tumours, this remains supposition at best.

(Epi)genetic/Genomic Instability

It is largely accepted that multiple mutations are required for incipient tumour cells to acquire all the hallmarks of cancer to become established malignancies [25]. These mutations might be incurred as a consequence of an unstable genome, perhaps induced by the presence of the initiating oncogenic event. Whilst NPM-ALK has been shown to have effects on DNA repair capacity within tumour cells, the genome is relatively silent with few consistent genomic or genetic abnormalities [34–41]. These data suggest that either NPM-ALK itself is able to induce the necessary cancer hallmarks and/or that epigenetic events are involved. It has been suggested that the former is the case, as NPM-ALK can indeed activate expression of many proteins that confer the cancer hallmarks as has been demonstrated in multiple publications [16, 42–60]. In addition, NPM-ALK can induce transformation of primary human T cells in a relatively short time frame, although in murine models this is not the case, yet the latter are not true mimics of ALCL [16, 61–64]. Epigenetic alterations are apparent in ALCL, although CpG methylation activity has also been attributed to NPM-ALK- mediated activation of DNMT1 [65, 66]. Hence, rather than requiring a status of (epi)genetic instability, the development of ALCL might instead be dependent on a defined set of 'just right' conditions.

The 'Just Right' Level of Signalling: The Goldilocks Hypothesis

Fortunately, ALCL is a rare cancer suggesting that distinct conditions must be met that are permissive of cellular transformation. Indeed, the presence of the t(2;5)(p23;q35) in as many as 1% of newborn cord blood suggests that not all children born harbouring this translocation will develop ALCL [67]. However, this study did not specifically examine haemopoietic progenitor cells for the presence of the translocation, although cord blood is enriched for these primitive cells. Presuming cells carrying the translocation persist into childhood and maintain its presence as they differentiate, the low prevalence of this cancer suggests that additional events are required for tumorigenesis. Indeed, some of these may be provided by the above-discussed (epi)genetic/genomic alterations characterised in ALCL although as previously mentioned, consistent abnormalities are even rarer and have not been proven as contributing events. An alternative pathway towards tumour development might instead be presented by the inherent activities of T cells in adaptive immunity, although as mentioned previously, established tumours lack cell surface expression of the TCR as well as proximal signalling proteins [11, 68]. However, the T cell compartment has immense plasticity and even in the absence of a functional TCR, cells may behave in an innate manner akin to innate lymphoid cells.

Antigen-Dependent Tumour Growth

In those scenarios where TCR rearrangements are conducive with the expression of a functional TCR, it is possible that antigen-dependent clonal stimulation of incipient tumour cells provides the necessary impetus for tumour growth (Fig. 6.3). In this model, much like in the process of normal T cell activation, on encountering peptide presented in the context of MHC, the affinity of a particular TCR for its ligand might dictate whether certain cells are clonally selected and expanded [69]. This process is a complex and highly regulated one dependent not just on recruitment of co-receptors, key kinases and phosphatases to the engaged TCR complex but also the 'tuning' of the signal intensity by cell surface proteins such as CD5 and CD45. Indeed, a low level of tonic signalling is required under homeostatic conditions to facilitate the survival of circulating naïve T cells, perhaps provided by binding with low affinity to self-peptide:MHC complexes. One might imagine a scenario whereby NPM-ALK might act to fine-tune this activity due to its ability to mimic TCR-induced signal transduction leading to activation of T cells on contact with low-affinity ligand [70]. It has also been demonstrated that differing levels of NPM-ALK expression are detectable in different ALCL tumours adding a further level of complexity whereby the combination of the strength of the NPM-ALK-induced and

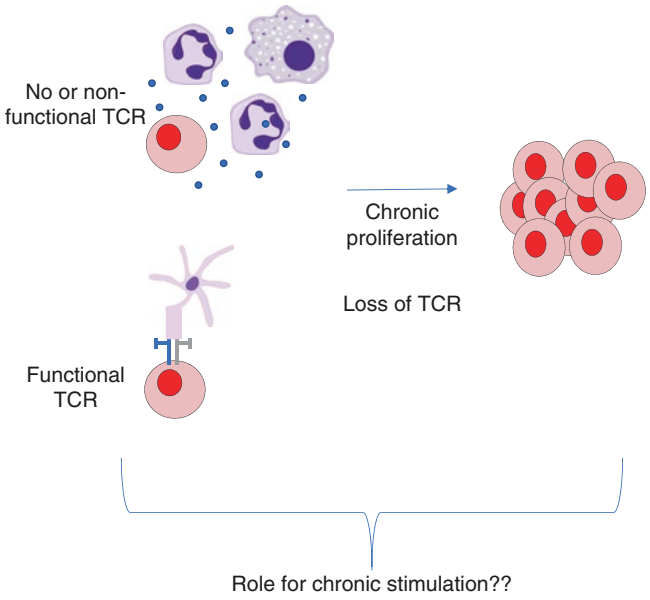

Fig. 6.3 Does chronic stimulation in an antigen-(in)dependent manner facilitate malignant transformation? In the former case, the lack of expression of a cell surface TCR eliminates the ability of cells to respond to antigen presentation and instead may behave more like innate lymphoid cells responding to an inflammatory milieu. In the latter case, the presence of a functional TCR interacting with antigen presented in the context of MHC by antigen-presenting cells may drive chronic proliferation of cells, which due to the presence of NPM-ALK do not reach exhaustion but instead continue to proliferate, in time gaining additional mutational events that are conducive with relatively autonomous growth and establishment of a malignancy. The TCR, if it was present, is downregulated as it becomes surplus to requirements given that NPM-ALK can mimic TCR-induced signalling pathways

TCR-induced signals dictates cell fate [71] (Fig. 6.4). This then begs the question as to why the TCR is no longer expressed on the surface of the established tumour cells, particularly as proteins of proximal TCR-induced signal transduction are silenced by epigenetic means in ALCL, although this may be the very reason given that without proximal T cell signalling proteins, no signal can be transduced through an engaged TCR and therefore its production is purely a waste of resources for the tumour cells [72]. Overall, it is likely that there is a delicate balance of signal transduction required for incipient tumour cells that likely changes on establishment of relatively autonomous growth, and that may differ from one tumour to the next.

Antigen-Independent Tumour Growth

Data published by Malcolm et al. showed that some ALCL have TCR rearrangements that are not consistent with expression of a functional TCR (or lack rearrangements completely) [12]. However, these tumours still express T cell markers such as CD4 suggesting that they derived from the thymic environment. These cells potentially mimic innate lymphoid cells (ILC), a cell type that lacks the ability to recognise distinct antigen and instead is responsive to an inflam-

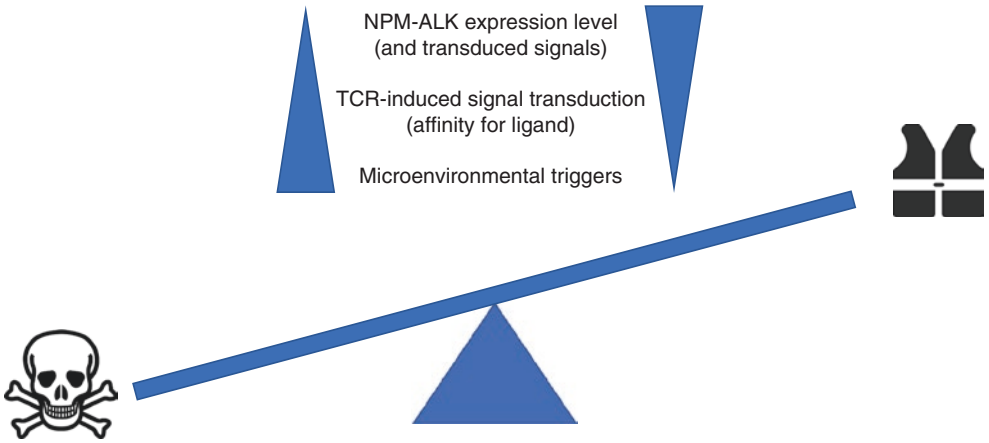

Fig. 6.4 The delicate balance between NPM-ALK expression levels, ligand-engaged TCR-induced signal transduction and outside-in signals provided by microenvironmental cues may dictate incipient tumour cell life or death. A number of 'just right' conditions may be required for transformation of circulating NPM-ALK-expressing T cells into tumour cells dependent on the status of the TCR. In the presence of the TCR, its engagement by ligand combined with the strength of signal transduction dictated by the level of NPM-ALK expression must be of a critical level to be conducive with cell survival and clonal expansion. This may also be influenced by microenvironmental cues which may play a larger role in the transformation of circulating NPM-ALK-expressing T cells that lack a cell surface TCR. Ultimately, the TCR, if present, is downregulated as it becomes redundant or is not compatible with the strength of signals initiated downstream of NPM-ALK and/or the microenvironment

matory milieu [73]. Serum samples taken from paediatric patients with ALCL are enriched with a significant number of cytokines including IL6, IL9, IL10 and IL17A with a trend towards increased levels of IL1ß, IL2, IL22, TNFα and IFNγ, and whilst these may not be truly representative of the cytokines present at the tumour site, it is possible that an inflammatory microenvironment contributes to disease pathogenesis [74]. An inflammatory milieu whereby a multitude of cytokines are produced may instead sustain tumour development rather than antigen-specific reactions of incipient tumour cells. In this scenario, ongoing chronic inflammation provides the impetus for cellular proliferation and transformation (Fig. 6.3).

Conclusions

There are no rules for cancer, and any one malignancy may well differ considerably in its pathogenesis from another of the same subtype. Therefore, whilst histopathologically speaking tumours may appear the same, the pathways they took to reach this status may differ and merge at various steps along the way. Hence, there may be multiple pathways of disease pathogenesis all converging on a similar histopathological presentation. Within ALCL, there are also multiple histopathologies, the most predominant being the common variant which may also reflect the differing pathways towards malignancy within this entity. Ultimately, Darwinian evolution applies whereby the fittest clone survives and constitutes the majority of the established growth. Current evidence suggests an early, primitive origin for ALCL within the thymus or the bone marrow, whereby the presence of the NPM-ALK generating or a variant translocation initiates a process whereby alongside T cell development, the conditions can become 'just right' for cellular transformation and establishment of a malignancy. Whether this process involves antigen-dependent or antigen-independent chronic stimulation remains to be determined although the low-level genomic/genetic instability, as is the case for most paediatric tumours, is suggestive of other contributory events.

References

1. Minard-Colin V, Brugieres L, Reiter A, Cairo MS, Gross TG, Woessmann W, et al. Non-Hodgkin lymphoma in children and adolescents: progress through effective collaboration, current knowledge, and challenges ahead. J Clin Oncol. 2015;33(27):2963–74.
2. Turner SD, Lamant L, Kenner L, Brugieres L. Anaplastic large cell lymphoma in paediatric and young adult patients. Br J Haematol. 2016;173(4):560–72.
3. Morris SW, Kirstein MN, Valentine MB, Dittmer KG, Shapiro DN, Saltman DL, et al. Fusion of a kinase gene, ALK, to a nucleolar protein gene, NPM, in non-Hodgkin's lymphoma. Science. 1994;263(5151):1281–4.
4. Duyster J, Bai RY, Morris SW. Translocations involving anaplastic lymphoma kinase (ALK). Oncogene. 2001;20(40):5623–37.
5. Damm-Welk C, Klapper W, Oschlies I, Gesk S, Rottgers S, Bradtke J, et al. Distribution of NPM1-ALK and X-ALK fusion transcripts in paediatric anaplastic large cell lymphoma: a molecular-histological correlation. Br J Haematol. 2009;146(3):306–9.
6. Perkins SL, Pickering D, Lowe EJ, Zwick D, Abromowitch M, Davenport G, et al. Childhood anaplastic large cell lymphoma has a high incidence of ALK gene rearrangement as determined by immunohistochemical staining and fluorescent in situ hybridi-

sation: a genetic and pathological correlation. Br J Haematol. 2005;131(5):624–7.
7. Hapgood G, Savage KJ. The biology and management of systemic anaplastic large cell lymphoma. Blood. 2015;126(1):17–25.
8. Ye X, Shokrollahi K, Rozen WM, Conyers R, Wright P, Kenner L, et al. Anaplastic large cell lymphoma (ALCL) and breast implants: breaking down the evidence. Mutat Res Rev Mutat Res. 2014;762:123–32.
9. Kinney MC, Kadin ME. The pathologic and clinical spectrum of anaplastic large cell lymphoma and correlation with ALK gene dysregulation. Am J Clin Pathol. 1999;111(1 Suppl 1):S56–67.
10. Swerdlow SH, World Health Organization & International Agency for Research on Cancer. WHO classification of tumours of haematopoietic and lymphoid tissues. Revised 4th ed. Lyon: IARC; 2017.
11. Bonzheim I, Geissinger E, Roth S, Zettl A, Marx A, Rosenwald A, et al. Anaplastic large cell lymphomas lack the expression of T-cell receptor molecules or molecules of proximal T-cell receptor signaling. Blood. 2004;104(10):3358–60.
12. Malcolm TI, Villarese P, Fairbairn CJ, Lamant L, Trinquand A, Hook CE, et al. Anaplastic large cell lymphoma arises in thymocytes and requires transient TCR expression for thymic egress. Nat Commun. 2016;7:10087.
13. Hogquist KA, Jameson SC. The self-obsession of T cells: how TCR signaling thresholds affect fate 'decisions' and effector function. Nat Immunol. 2014;15(9):815–23.
14. Pearson JD, Lee JK, Bacani JT, Lai R, Ingham RJ. NPM-ALK and the JunB transcription factor regulate the expression of cytotoxic molecules in ALK-positive, anaplastic large cell lymphoma. Int J Clin Exp Pathol. 2011;4(2):124–33.
15. Hsu FY, Johnston PB, Burke KA, Zhao Y. The expression of CD30 in anaplastic large cell lymphoma is regulated by nucleophosmin-anaplastic lymphoma kinase-mediated JunB level in a cell type-specific manner. Cancer Res. 2006;66(18):9002–8.
16. Zhang Q, Wei F, Wang HY, Liu X, Roy D, Xiong QB, et al. The potent oncogene NPM-ALK mediates malignant transformation of normal human CD4(+) T lymphocytes. Am J Pathol. 2013;183(6):1971–80.
17. Iqbal J, Weisenburger DD, Greiner TC, Vose JM, McKeithan T, Kucuk C, et al. Molecular signatures to improve diagnosis in peripheral T-cell lymphoma and prognostication in angioimmunoblastic T-cell lymphoma. Blood. 2010;115(5):1026–36.
18. Schleussner N, Merkel O, Costanza M, Liang HC, Hummel F, Romagnani C, et al. The AP-1-BATF and -BATF3 module is essential for growth, survival and TH17/ILC3 skewing of anaplastic large cell lymphoma. Leukemia. 2018;32(9):1994–2007.
19. Eckerle S, Brune V, Doring C, Tiacci E, Bohle V, Sundstrom C, et al. Gene expression profiling of isolated tumour cells from anaplastic large cell lymphomas: insights into its cellular origin, pathogenesis and relation to Hodgkin lymphoma. Leukemia. 2009;23(11):2129–38.
20. Matsuyama H, Suzuki HI, Nishimori H, Noguchi M, Yao T, Komatsu N, et al. miR-135b mediates NPM-ALK-driven oncogenicity and renders IL-17-producing immunophenotype to anaplastic large cell lymphoma. Blood. 2011;118(26):6881–92.
21. Kasprzycka M, Marzec M, Liu X, Zhang Q, Wasik MA. Nucleophosmin/anaplastic lymphoma kinase (NPM/ALK) oncoprotein induces the T regulatory cell phenotype by activating STAT3. Proc Natl Acad Sci U S A. 2006;103(26):9964–9.
22. O'Shea JJ, Paul WE. Mechanisms underlying lineage commitment and plasticity of helper CD4+ T cells. Science. 2010;327(5969):1098–102.
23. Gao W, Thompson L, Zhou Q, Putheti P, Fahmy TM, Strom TB, et al. Treg versus Th17 lymphocyte lineages are cross-regulated by LIF versus IL-6. Cell Cycle. 2009;8(9):1444–50.
24. Tripathi SK, Lahesmaa R. Transcriptional and epigenetic regulation of T-helper lineage specification. Immunol Rev. 2014;261(1):62–83.
25. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell. 2011;144(5):646–74.
26. Visvader JE, Lindeman GJ. Cancer stem cells: current status and evolving complexities. Cell Stem Cell. 2012;10(6):717–28.
27. Moti N, Malcolm T, Hamoudi R, Mian S, Garland G, Hook CE, et al. Anaplastic large cell lymphoma-propagating cells are detectable by side population analysis and possess an expression profile reflective of a primitive origin. Oncogene. 2015;34(14):1843–52.
28. Wang JC, Dick JE. Cancer stem cells: lessons from leukemia. Trends Cell Biol. 2005;15(9):494–501.
29. Vieyra DS, Rosen A, Goodell MA. Identification and characterization of side population cells in embryonic stem cell cultures. Stem Cells Dev. 2009;18(8):1155–66.
30. Wulf GG, Wang RY, Kuehnle I, Weidner D, Marini F, Brenner MK, et al. A leukemic stem cell with intrinsic drug efflux capacity in acute myeloid leukemia. Blood. 2001;98(4):1166–73.
31. Piccaluga PP, Ascani S, Fraternali Orcioni G, Piccioli M, Pileri A Jr, Falini B, et al. Anaplastic lymphoma kinase expression as a marker of malignancy. Application to a case of anaplastic large cell lymphoma with huge granulomatous reaction. Haematologica. 2000;85(9):978–81.
32. Lamant L, Pileri S, Sabattini E, Brugieres L, Jaffe ES, Delsol G. Cutaneous presentation of ALK-positive anaplastic large cell lymphoma following insect bites: evidence for an association in five cases. Haematologica. 2010;95(3):449–55.
33. Skinner GR. Transformation of primary hamster embryo fibroblasts by type 2 simplex virus: evidence for a "hit and run" mechanism. Br J Exp Pathol. 1976;57(4):361–76.
34. Youssif C, Goldenbogen J, Hamoudi R, Carreras J, Viskaduraki M, Cui Y-X, et al. Genomic profiling of pediatric ALK-positive anaplastic large cell lymphoma: a children's cancer and leukaemia group study. Genes Chromosomes Cancer. 2009;48(11):1018–26.
35. Salaverria I, Bea S, Lopez-Guillermo A, Lespinet V, Pinyol M, Burkhardt B, et al. Genomic profiling reveals different genetic aberrations in systemic ALK-positive and ALK-negative anaplastic large cell lymphomas. Br J Haematol. 2008;140(5):516–26.
36. Slupianek A, Poplawski T, Jozwiakowski SK, Cramer K, Pytel D, Stoczynska E, et al. BCR/ABL stimulates WRN to promote survival and genomic instability. Cancer Res. 2011;71(3):842–51.
37. Rink L, Slupianek A, Stoklosa T, Nieborowska-Skorska M, Urbanska K, Seferynska I, et al. Enhanced phosphorylation of Nbs1, a member of DNA repair/checkpoint complex Mre11-RAD50-Nbs1, can be targeted to increase the efficacy of imatinib mesylate against BCR/ABL-positive leukemia cells. Blood. 2007;110(2):651–60.
38. Hoser G, Majsterek I, Romana DL, Slupianek A, Blasiak J, Skorski T. Fusion oncogenic tyrosine kinases alter DNA damage and repair after genotoxic treatment: role in drug resistance? Leuk Res. 2003;27(3):267–73.
39. Slupianek A, Hoser G, Majsterek I, Bronisz A, Malecki M, Blasiak J, et al. Fusion tyrosine kinases induce drug resistance by stimulation of homology-dependent recombination repair, prolongation of G(2)/M phase, and protection from apoptosis. Mol Cell Biol. 2002;22(12):4189–201.
40. Young LC, Bone KM, Wang P, Wu F, Adam BA, Hegazy S, et al. Fusion tyrosine kinase NPM-ALK deregulates MSH2 and suppresses DNA mismatch repair function novel insights into a potent oncoprotein. Am J Pathol. 2011;179(1):411–21.
41. Crescenzo R, Abate F, Lasorsa E, Tabbo F, Gaudiano M, Chiesa N, et al. Convergent mutations and kinase fusions lead to oncogenic STAT3 activation in anaplastic large cell lymphoma. Cancer Cell. 2015;27(4):516–32.
42. Zhang Q, Wang H, Kantekure K, Paterson JC, Liu X, Schaffer A, et al. Oncogenic tyrosine kinase NPM-ALK induces expression of the growth-promoting receptor ICOS. Blood. 2011;118(11):3062–71.

43. Zhang Q, Raghunath PN, Xue L, Majewski M, Carpentieri DF, Odum N, et al. Multilevel dysregulation of STAT3 activation in anaplastic lymphoma kinase-positive T/null-cell lymphoma. J Immunol. 2002;168(1):466–74.
44. Slupianek A, Nieborowska-Skorska M, Hoser G, Morrione A, Majewski M, Xue L, et al. Role of phosphatidylinositol 3-kinase-Akt pathway in nucleophosmin/anaplastic lymphoma kinase-mediated lymphomagenesis. Cancer Res. 2001;61(5):2194–9.
45. Marzec M, Liu X, Wong W, Yang Y, Pasha T, Kantekure K, et al. Oncogenic kinase NPM/ALK induces expression of HIF1alpha mRNA. Oncogene. 2011;30(11):1372–8.
46. Marzec M, Zhang Q, Goradia A, Raghunath PN, Liu X, Paessler M, et al. Oncogenic kinase NPM/ALK induces through STAT3 expression of immunosuppressive protein CD274 (PD-L1, B7-H1). Proc Natl Acad Sci U S A. 2008;105(52):20852–7.
47. Marzec M, Halasa K, Liu X, Wang HY, Cheng M, Baldwin D, et al. Malignant transformation of CD4+ T lymphocytes mediated by oncogenic kinase NPM/ALK recapitulates IL-2-induced cell signaling and gene expression reprogramming. J Immunol. 2013;191(12):6200–7.
48. Anastasov N, Bonzheim I, Rudelius M, Klier M, Dau T, Angermeier D, et al. C/EBPbeta expression in ALK-positive anaplastic large cell lymphomas is required for cell proliferation and is induced by the STAT3 signaling pathway. Haematologica. 2010;95(5):760–7.
49. Bai RY, Dieter P, Peschel C, Morris SW, Duyster J. Nucleophosmin-anaplastic lymphoma kinase of large-cell anaplastic lymphoma is a constitutively active tyrosine kinase that utilizes phospholipase C-gamma to mediate its mitogenicity. Mol Cell Biol. 1998;18(12):6951–61.
50. Bai RY, Ouyang T, Miething C, Morris SW, Peschel C, Duyster J. Nucleophosmin-anaplastic lymphoma kinase associated with anaplastic large-cell lymphoma activates the phosphatidylinositol 3-kinase/Akt antiapoptotic signaling pathway. Blood. 2000;96(13):4319–27.
51. Cussac D, Greenland C, Roche S, Bai RY, Duyster J, Morris SW, et al. Nucleophosmin-anaplastic lymphoma kinase of anaplastic large-cell lymphoma recruits, activates, and uses pp60c-src to mediate its mitogenicity. Blood. 2004;103(4):1464–71.
52. Ambrogio C, Voena C, Manazza AD, Piva R, Riera L, Barberis L, et al. p130Cas mediates the transforming properties of the anaplastic lymphoma kinase. Blood. 2005;106(12):3907–16.
53. Chiarle R, Simmons WJ, Cai H, Dhall G, Zamo A, Raz R, et al. Stat3 is required for ALK-mediated lymphomagenesis and provides a possible therapeutic target. Nat Med. 2005;11(6):623–9.
54. Voena C, Conte C, Ambrogio C, Boeri Erba E, Boccalatte F, Mohammed S, et al. The tyrosine phosphatase Shp2 interacts with NPM-ALK and regulates anaplastic lymphoma cell growth and migration. Cancer Res. 2007;67(9):4278–86.
55. Desjobert C, Renalier MH, Bergalet J, Dejean E, Joseph N, Kruczynski A, et al. MiR-29a down-regulation in ALK-positive anaplastic large cell lymphomas contributes to apoptosis blockade through MCL-1 overexpression. Blood. 2011;117(24):6627–37.
56. Colomba A, Giuriato S, Dejean E, Thornber K, Delsol G, Tronchere H, et al. Inhibition of Rac controls NPM-ALK-dependent lymphoma development and dissemination. Blood Cancer J. 2011;1(6):e21.
57. Dejean E, Foisseau M, Lagarrigue F, Lamant L, Prade N, Marfak A, et al. ALK+ALCLs induce cutaneous, HMGB-1-dependent IL-8/CXCL8 production by keratinocytes through NF-kappaB activation. Blood. 2012;119(20):4698–707.
58. Lim MS, Carlson ML, Crockett DK, Fillmore GC, Abbott DR, Elenitoba-Johnson OF, et al. The proteomic signature of NPM/ALK reveals deregulation of multiple cellular pathways. Blood. 2009;114(8):1585–95.
59. McDonnell SR, Hwang SR, Basrur V, Conlon KP, Fermin D, Wey E, et al. NPM-ALK signals through glycogen synthase kinase 3beta to promote oncogenesis. Oncogene. 2012;31(32):3733–40.
60. McDonnell SR, Hwang SR, Rolland D, Murga-Zamalloa C, Basrur V, Conlon KP, et al. Integrated phosphoproteomic and metabolomic profiling reveals NPM-ALK-mediated phosphorylation of PKM2 and metabolic reprogramming in anaplastic large cell lymphoma. Blood. 2013;122(6):958–68.
61. Turner SD, Merz H, Yeung D, Alexander DR. CD2 promoter regulated nucleophosmin-anaplastic lymphoma kinase in transgenic mice causes B lymphoid malignancy. Anticancer Res. 2006;26(5A):3275–9.
62. Turner SD, Tooze R, Maclennan K, Alexander DR. Vav-promoter regulated oncogenic fusion protein NPM-ALK in transgenic mice causes B-cell lymphomas with hyperactive Jun kinase. Oncogene. 2003;22(49):7750–61.
63. Chiarle R, Gong JZ, Guasparri I, Pesci A, Cai J, Liu J, et al. NPM-ALK transgenic mice spontaneously develop T-cell lymphomas and plasma cell tumors. Blood. 2003;101(5):1919–27.
64. Riera L, Lasorsa E, Bonello L, Sismondi F, Tondat F, Di Bello C, et al. Description of a novel Janus kinase 3 P132A mutation in acute megakaryoblastic leukemia and demonstration of previously reported Janus kinase 3 mutations in normal subjects. Leuk Lymphoma. 2011;52(9):1742–50.
65. Hassler MR, Pulverer W, Lakshminarasimhan R, Redl E, Hacker J, Garland GD, et al. Insights into the pathogenesis of anaplastic large-cell lymphoma through genome-wide DNA methylation profiling. Cell Rep. 2016;17(2):596–608.
66. Zhang Q, Wang HY, Woetmann A, Raghunath PN, Odum N, Wasik MA. STAT3 induces transcription of the DNA methyltransferase 1 gene (DNMT1) in malignant T lymphocytes. Blood. 2006;108(3):1058–64.
67. Laurent C, Lopez C, Desjobert C, Berrebi A, Damm-Welk C, Delsol G, et al. Circulating t(2;5)-positive cells can be detected in cord blood of healthy newborns. Leukemia. 2012;26(1):188–90.
68. Ambrogio C, Voena C, Manazza AD, Martinengo C, Costa C, Kirchhausen T, et al. The anaplastic lymphoma kinase controls cell shape and growth of anaplastic large cell lymphoma through Cdc42 activation. Cancer Res. 2008;68(21):8899–907.
69. Gaud G, Lesourne R, Love PE. Regulatory mechanisms in T cell receptor signalling. Nat Rev Immunol. 2018;18(8):485–97.
70. Turner SD, Yeung D, Hadfield K, Cook SJ, Alexander DR. The NPM-ALK tyrosine kinase mimics TCR signalling pathways, inducing NFAT and AP-1 by RAS-dependent mechanisms. Cell Signal. 2007;19(4):740–7.
71. Pomari E, Basso G, Bresolin S, Pillon M, Carraro E, d'Amore ES, et al. NPM-ALK expression levels identify two distinct subtypes of paediatric anaplastic large cell lymphoma. Leukemia. 2017;31(2):498–501.
72. Ambrogio C, Martinengo C, Voena C, Tondat F, Riera L, di Celle PF, et al. NPM-ALK oncogenic tyrosine kinase controls T-cell identity by transcriptional regulation and epigenetic silencing in lymphoma cells. Cancer Res. 2009;69(22):8611–9.
73. Mjosberg J, Spits H. Human innate lymphoid cells. J Allergy Clin Immunol. 2016;138(5):1265–76.
74. Knorr F, Damm-Welk C, Ruf S, Singh VK, Zimmermann M, Reiter A, et al. Blood cytokine concentrations in pediatric patients with anaplastic lymphoma kinase-positive anaplastic large cell lymphoma. Haematologica. 2018;103(3):477–85.

Pathology

7

Christoph Kornauth, Ana-Iris Schiefer, and Ingrid Simonitsch-Klupp

Introduction

Tumors derived from the lymphoid system comprise neoplastic lesions originated from both precursor and mature B and T/NK cells. The vast majority thereof can be related both genotypically and (immuno-)phenotypically to their physiologically occurring counterparts within diverse lymphoid compartments. The WHO classification of tumors of the hematopoietic and lymphoid tissues [1] has recently been updated (revised 5th edition 2016) and enlists numerous groups and entities of different lymphoid proliferations, which share distinct clinical, morphologic, immunophenotypical, and genetic similarities. The vast majority of B-cell and T-cell non-Hodgkin's lymphomas (NHL) occurring during childhood are aggressive malignancies, whereas indolent lymphomas, on the other hand, are rare in the pediatric population in contrast to the adult population.

In this chapter, however, not all lymphomas will be described; instead, we have focused on those lymphomas occurring most frequently in childhood and young adolescence, as well as try to emphasize the new categories described in the young population in the most recent years.

Precursor Lymphoid Neoplasms

This category includes B-lymphoblastic lymphoma/leukemia (B-LBL/B-ALL) of either the NOS category or with recurrent genetic abnormalities, as well as precursor T-cell neoplasms (T-lymphoblastic lymphoma/leukemia, T-LBL/T-ALL) and NK-cell leukemia/lymphoma.

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LBL is a lymphoma/leukemia of precursor B or T cells with blastoid appearance and immature immunoprofile. When present in the bone marrow (BM) and peripheral blood (PB), it is termed ALL, whereas in case of primary and almost exclusive presentation in lymph nodes or extranodal sites, with only minor BM infiltration (<25%), the term LBL is preferred. However, often both leukemic and lymphomatous components are seen in one patient. LBL represents roughly 30% of lymphomas in childhood, whereas it is infrequently seen in adults [2]. B-LBL more often presents as a leukemic disease, whereas T-LBL is usually associated with a lymphomatous disease, thereby often presenting as a mediastinal mass [3]. In case of lymphomatous B-LBL, one singular peripheral lymph node or extranodal tissues such as the skin, the gonads, soft tissue, or tonsils are the primary sites [4].

Morphology and Immunohistology (Fig. 7.1)

LBL of both B-cell and T-cell immunophenotype present with a characteristic diffuse infiltration with effacement of the underlying tissue. Lymph nodes affected by T-LBL often show a marked interfollicular infiltration with sparing of residual B-cell follicles. In many cases, a diffuse and streaky or single-file infiltration of the perinodal fatty tissue and adjacent tissue can be detected. Tumors represent rapidly dividing neoplasms with numerous mitotic figures and also frequent apoptosis, sometimes with abundant starry-sky macrophages intermingled. Cytologically, LBL tumors are relatively monotonous infiltrations composed of small lymphoid cells with round to oval, slightly irregular nuclear contours and narrow cytoplasms. In most cases, nucleoli are rather small, but may sometimes be prominent. The chromatin is usually finely dispersed. By pure morphology, a distinction of B-LBL and T-LBL is not conceivable.

In smears or touch preparations, small- to medium-sized blasts are seen, with variations in cytoplasmic ranges varying

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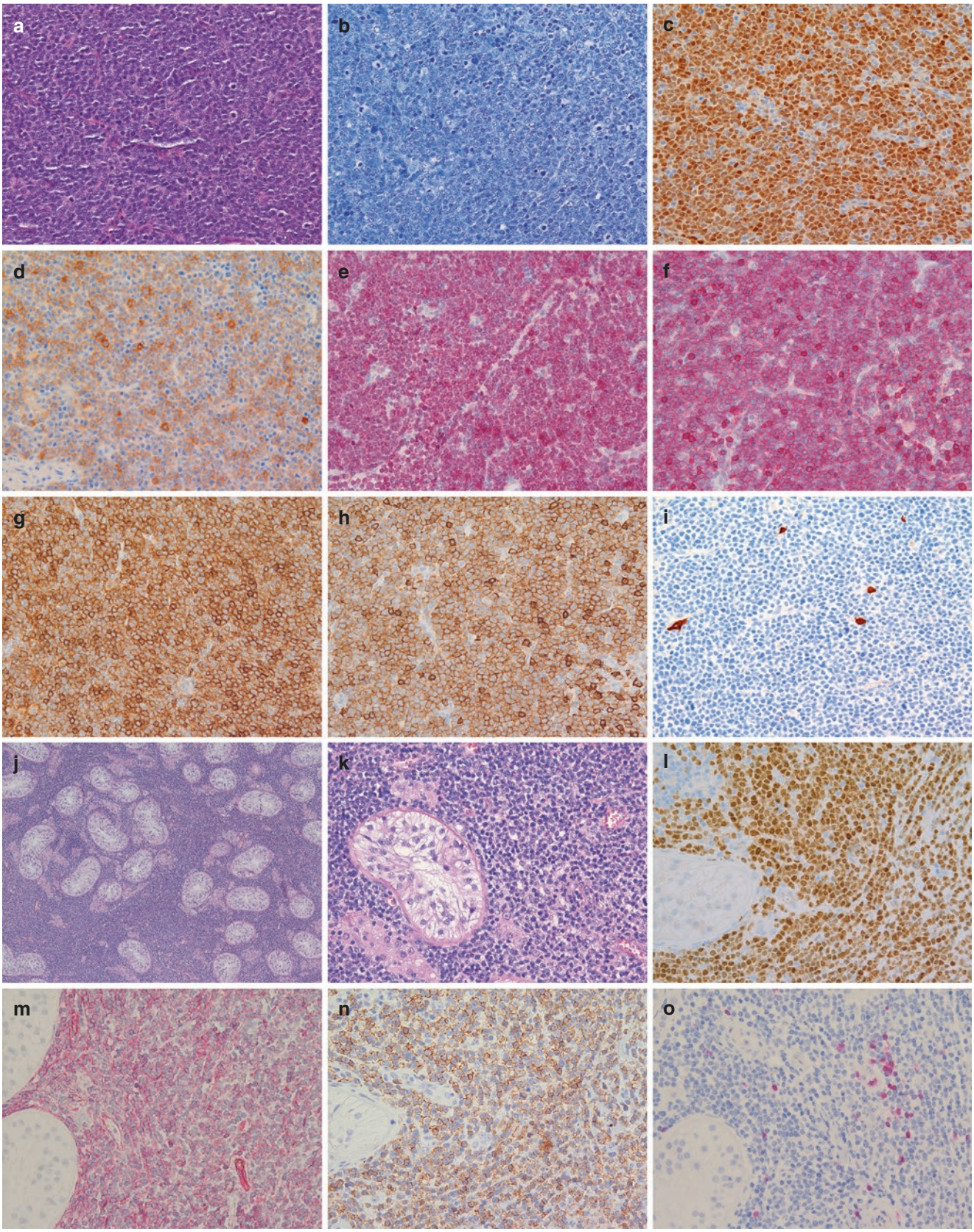

Fig. 7.1 Lymphoblastic lymphoma. Lymphoblastic lymphoma/leukemia of T (**a**–**i**)- and B (**j**–**o**)-cell origin: Morphologically indistinguishable, both neoplasms are composed of small- to medium-sized monomorphic blasts (**a**, **j** + **k** with testicular involvement). TdT is positive in both subtypes (**c**, **l**), whereas CD3 (**e**, **o**) and CD5 (**f**) are present in T-LBL. CD1a (**d**) positivity and CD4 (**g**)-CD8 (**h**) co-expression are indicative of a late cortical T-cell phenotype. CD34 (**m**) is variably positive in both subtypes, whereas CD117 (**i**) is generally absent. CD19 (**n**) is indicative of B-cell origin

from narrow to moderate sizes, sometimes with small vacuoles. In about 10% of cases, coarse azurophilic granules may be noticed, which may stain with Sudan Black B on smears or touch preparations [5]. The nuclei are either roundish or slightly convoluted, and contain dispersed chromatin with a variable amount of nucleoli [6]. Cytochemical analysis shows negativity of blasts for MPO and NSE, but may show reactivity in the PAS stain [1].

Immunophenotype of B-LBL/B-ALL

The blasts are positive for the B-cell markers CD19, cyCD79a, and cyCD22 and are usually positive for PAX-5 and TdT, whereas expression of CD20 and CD34 is variable [7]. Positivity of myeloid markers such as CD13 and CD33 may be noted, whereas MPO expression is usually absent, although weak expression may be seen with immunohistochemistry, especially after antigen retrieval [7, 8]. Care must be taken in cases of strong MPO expression, because most MPO+ cases belong to the AML category or represent mixed phenotype B/myeloid leukemia [1, 7]. Based on the stage of B-blast-differentiation [9], antigen expression varies, and basically four stages are discerned: in the earliest stage (pro-B ALL or early precursor B-ALL), blasts are positive for CD19, CD79a, CD22, and TdT. In the second stage (common B-ALL) the blasts show additional expression of CD10; the two most mature stages comprise pre-B-ALL with expression of cytoplasmic μ-chains and the transitional stage of B-ALL with surface μ-chains. The stage of differentiation correlates both clinically as well as genetically [1].

Immunophenotype of T-LBL/T-ALL

The lymphoblasts typically express TdT, CD7, and CD3. In a similar approach as in B-LBL/ALL, T-ALL/LBL has been subcategorized into different stages according to intrathymic differentiation [10]: the first stage (pro-T or TI-I) is characterized by expression of cyCD3 and CD7; the pre-T (T-II) shows a phenotype with expression of cyCD3, CD7, and CD5 or CD2; the cortical stage (T-III) exhibits expression of cyCD3 and CD1a, with variable expression of sCD3, and the final medullary or mature stage (T-IV) shows typically a cyCD3+ and sCD3+/CD1a- immunophenotype [11]. Additionally, CD8 and CD4 are frequently co-expressed, especially in the cortical stage. CD10 may be positive, but is not specific for T-precursor neoplasms, because it is also typically expressed in mature T-cell lymphomas with a follicular T-helper cell immunophenotype [12]. CD79a may be weakly expressed in cases of T-LBL [13], as well as myeloid antigens such as CD13 and CD33 [14]. A rather rare ALL, termed early T-cell precursor lymphoblastic leukemia, is separated from other T-ALL/LBL cases; it usually shows a characteristic immunophenotype with co-expression of CD7 together with one or more stem cell or myeloid markers including CD34, CD117, HLA-DR, CD13, CD33, CD11b, and CD65. The tumor cells may express cyCD3, CD2, and/or CD4, whereas CD8 and CD1a are constantly negative, as is CD5 in most cases [15].

Differential Diagnosis

B-LBL has to be distinguished from other diffusely infiltrating B-cell lymphomas (Table 7.1). Blasts of B-LBL have to be distinguished from normal immature B-cell precursors (so-called hematogones), which may be abundant in reactive and inflammatory conditions. Hematogones [16] are CD10+, and typically show a spectrum of B-cell differentiation [17], with heterogeneous expression of immature and mature B-cell antigens in the same specimen [18]. Moreover, hematogones are usually randomly dispersed, and do not form clusters in bone marrow biopsies as would be typical for ALL.

T-LBL has to be distinguished from other more mature T-cell lymphoproliferations, especially in cases with mature or medullary differentiation with absence of CD34 and CD1a, when also TdT may be not expressed anymore [19]. Another diagnostic pitfall is the distinction between thymoma and T-lymphoblastic lymphoma in needle biopsies of mediastinal tumors. A strongly recommended diagnostic criterion favoring thymoma over T-LBL is the demonstration of numerous cytokeratin-positive epithelial cells by immunohistochemistry [20].

Furthermore, T-LBL has to be distinguished from indolent T-lymphoblastic proliferations (iT-LBP), which have been described recently in association with Castleman disease, angioimmunoblastic T-cell lymphomas and follicular dendritic cell tumors associated with Castleman disease [21], and also in a case with disseminated multinodal involvement [22]. These peculiar proliferations of TdT+ T-cells mimic T-LBL, but are benign, non-clonal proliferations without requirement of further therapy [23]. They are characterized by confluent groups of TdT+ T cells with a cortical T-cell immunophenotype, with preservation of the surrounding tissue architecture, and a low-proliferation fraction.

Genetic Profile

Almost all B-LBL/ALL show a clonal rearrangement of the immunoglobulin (Ig) chain genes. Additionally, clonal rearrangements of the T-cell receptor (TCR) genes may be detected in many cases of B-ALL/LBL, thus being not beneficial in lineage determination [24]. T-LBL and T-ALL, on

Table 7.1 Clinicopathologic characteristics of B-cell lymphomas with blastoid morphology

	DLBCL	BL	B-LBL
Morphology	Large, mainly CB, IB	Medium sized	Monomorphic, small lymphoid-like
Chromatin	Clumped, irregular	Coarse	Fine
Nucleoli	Prominent, centrally (IB) or membrane-bound (CB)	Variable, randomly distributed	No
Cytoplasm	Moderate/broad	Small, deeply basophilic, typically numerous vacuoles	Very narrow
Architecture	Diffuse infiltrates	Diffuse, prominent starry-sky pattern	Diffuse
Immunophenotype	GCB/non-GCB; mature B-cell, BCL-2+/−, MUM-1+/−, CD10+/−, BCL-6+/−	MatureBcell,CD10+,BCL-6+,BCL-2-,MUM-1-	Precursor B-cell phenotype
Cytogenetics	Complex; see Ref. [76]	C-MYC-Translocation	Numerous; see Ref. [26, 28]

Abbreviations: *DLBCL* diffuse large B-cell lymphoma, *BL* Burkitt lymphoma, *B-LBL* B-lymphoblastic lymphoma, *CB* centroblasts, *IB* immunoblasts, *GCB* germinal center B cell-like, *non-GCB* nongerminal center B cell-like

the other hand, show besides almost constant rearrangement of the TCR chain genes concomitant rearrangements of the IgH genes in about 20% of cases [25].

Cytogenetic and molecular abnormalities are detected in almost all cases of B-ALL/LBL, frequently defining specific entities acknowledged in the WHO classification [1, 26]; these include mainly balanced translocations or numeric aberrations, and they are associated with unique phenotypic and prognostic features [1]. Chromosomal abnormalities reported in T-LBL and T-ALL frequently comprise rearrangements of the regulatory regions of TCR genes and oncogenes such as *TLX-1*, *TLX3*, *TAL1*, and *LMO2* [27, 28].

For detailed description of genetic alterations in LBL/ALL, see the detailed chapters.

Burkitt Lymphoma

The three epidemiologic variants of Burkitt lymphomas (BL) constitute the endemic form mostly encountered in central Africa, the sporadic form occurring all over the world, and the immunodeficiency-associated variant linked with various forms of immunosuppression, mainly HIV infection.

Sporadic BL represents the most common subtype in Western Europe and the USA. In these regions it accounts for approximately 30–50% of all lymphomas of childhood [29], whereas it is a rare tumor in the overall population.

All BL variants are highly aggressive tumors, which differ slightly in their geographical distribution, clinical presentation, EBV association, biology, genetics [30], as well as morphologic and immunophenotypic features [31–33].

All three forms most often occur at extranodal sites, with different preferential sites for the respective variants [34]; in endemic BL, the jaws are most often involved (in >50% of cases) [35], whereas the sporadic variant recurrently presents with abdominal masses, thereby involving the terminal ileum and the ileo-cecal region [31]. In immunodeficiency-associated BL, the bone marrow is often primarily involved [36], thereby presenting as an overt acute Burkitt leukemia (in previous classification schemes termed FAB L3 leukemia), and it also may show lymph-node involvement, whereas a nodal involvement is rare in both other variants. All three variants tend to involve the CNS as well as breasts, kidneys, gonads, and other solid organs [29].

Morphology and Immunohistology (Fig. 7.2)

Typical BL is composed of monomorphic medium sized, cohesively lying blastoid cells with usually a "salt-and-pepper" chromatin distribution, which may be also finely dispersed. They exhibit multiple, not exceptionally prominent nucleoli, which are mainly located in the paracentral area of the nucleus. The cytoplasms are medium sized and deeply basophilic, sometimes with squared-off borders in jigsaw-like patterns. BL is one of the most-rapidly dividing human tumors, with a doubling time of approximately 24–48 hours [37]. Mitotic figures are exceedingly often encountered, as well as high rates of apoptotic figures. A characteristic finding is the so-called starry-sky appearance of the otherwise deeply basophilic infiltrates, caused by numerous pale-appearing phagocytic histiocytes scattered among the blastoid cells which are incorporating apoptotic bodies from the rapidly dividing tumor cells. It should be emphasized, however, that the starry-sky picture is not specific for BL, but may be seen in other cases of rapidly growing hematopoietic tumor infiltrates [38]. On touch preparations or on bone marrow-smears the deeply basophilic cytoplasms exhibit many oil-red-positive lipid-rich vacuoles.

In several cases a slight pleomorphism of nuclei may be noted; the cells may show more prominent nucleoli, and especially in immunodeficiency-associated BL even a subtle plasmacytoid differentiation may occur, which does not contradict the diagnosis of BL [39].

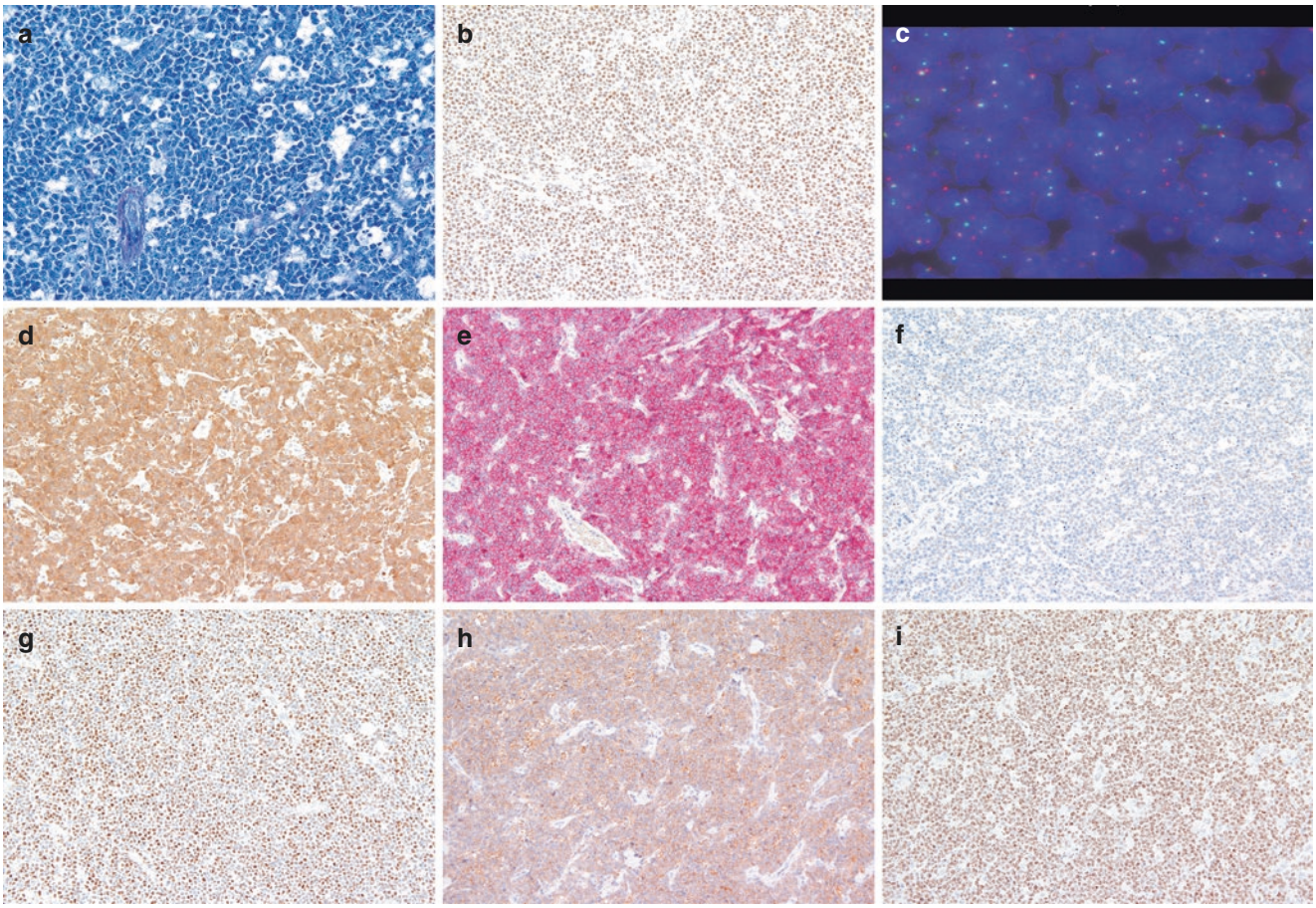

Fig. 7.2 Burkitt lymphoma. Burkitt lymphoma is a highly aggressive lymphoma with cohesive monomorphous medium-sized tumor cells with finely dispersed chromatin and inconspicuous nucleoli, deeply basophilic cytoplasms, and many starry-sky macrophages in the background (**a**). The molecular hallmark of BL is the translocation of *MYC* at band 8q24 (**c**): fluorescence in situ hybridization (FISH), *MYC* break-apart probe, one allele shows co-localization of probes (yellow signal), the other allele a separation of probes (separate red and green signal) indicative of translocation which leads to a strong expression of MYC protein (**b**). Tumor cells typically express TCL1 (**d**), CD38 (**e**), BCL6 (**g**), and CD10 (**h**). BCL2 is usually negative or weakly positive (**f**). The proliferation rate is nearly 100% (**i**)

However, the characteristic morphologic features will only be detected in specimens with optimal fixation [40]; especially in small samples with only few tumor cells the starry-sky pattern may be absent, and the tumor cells, if only poorly fixated, may show more variability in size and shape. In these cases, immunophenotypic and molecular features, as well as the clinical presentation may be of significant importance for the correct diagnosis [41–44].

The tumor cells typically express pan-B-cell markers such as CD19, CD20, CD79a, PAX-5, and CD22 together with surface IgM (sIgM+) [38]. Additionally, antigens typically expressed by germinal-center derived B cells, such as CD10, BCL-6, and GCET-1, are detected in almost all specimens. The tumor cells show strong nuclear expression of the c-myc protein in the vast majority of cases [45]. The proliferation fraction (Ki-67) is exceptionally high with almost 100% of the tumor cells positively stained. Typically, BL cells express CD38, TCL-1, and lack CD44 expression [42]. The tumor cells may be positive for CD43 and CD77 [30], whereas they are negative with CD23, CD5, CD138, TdT, CD34, and Cyclin-D1.

Usually both MUM-1 and BCL-2 are negative or only very weakly and inhomogeneously expressed [46].

Genetic Profile

BL show a characteristic translocation of *MYC* at 8q24 locus to the heavy chain of the immunoglobulin gene (IgH) on chromosome 14 (14q32), t(8;14)(q24;q32) or, less commonly, to the light-chain gene loci, kappa on chromosome 2, t(2;8), or the lambda locus at 22q11, t(8;22), respectively [36]. Interestingly, the location of the breakpoints is different in sporadic and ID-associated cases, where the breakpoints are near or within *MYC*, compared to endemic cases, where the breakpoints are scattered in a wide range upstream of *MYC* [47]. Moreover, several other genetic aberrations may be noted in BL, such as gains of chromosomes, typically +1q, +7, +12, or losses of e.g. 17p [48].

Approximately 10% of all otherwise typical BL lack *MYC* translocation [39]. In these cases, strict clinicopathologic and

immunophenotypic criteria have to be fulfilled to separate other aggressive lymphomas, which may mimic classical BL.

The WHO classification 2016 has enlisted a new provisional category termed "Burkitt-like lymphoma with 11q aberration" [1]. These lymphomas resemble classical BL both morphologically as well as in their immunoprofiles, but lack *MYC* translocations, and usually do not show 1q abnormalities frequently detected in BL [49]. Morphologically, they may show more cellular pleomorphism, and present with primary nodal involvement [50]. Using flow cytometry, these lymphomas were characterized by lower CD38 expression and more enhanced expression of CD45 than classical *MYC*+ BL [51] and showed expression of CD56/CD16 in 60% of cases [51].

Differential Diagnosis

Burkitt lymphoma has to be distinguished from other diffusely growing blastoid lymphomas, especially diffuse large B-cell lymphoma [52]. The main morphologic and immunophenotypic features of the respective lymphoma subtypes are listed in Table 7.1.

Diffuse Large B-Cell Lymphoma (DLBCL)

DLBCL constitute approximately 20% of childhood NHL [2, 53]. In recent years, much effort has been made to classify the large group of DLBCL into various morphologic, genetic, and molecular subtypes and entities, which present with variable clinicopathologic characteristics as well as prognosis and outcome. In children, the lymphomas usually present at a single site and commonly with nodal involvement. The most common extranodal sites involved are the abdomen and the mediastinum. DLBCL is often associated with various conditions of either primary or secondary immunodeficiency [54, 55]. Transformation of a previous indolent B-cell lymphoma, such as CLL or FL, which is often the case in adults, almost never occurs in the pediatric population. Distinct lymphoma subtypes with peculiar clinicopathologic and molecular characteristics listed as separate entities among the large B-cell lymphomas, such as T-cell/histiocyte-rich large B-cell lymphomas (T/H-RLBCL), primary DLBCL of the CNS, as well as primary mediastinal B-cell lymphoma (PMBCL), have also been described in children.

Morphology and Immunohistology (Fig. 7.3)

The tissue involved usually shows entire destruction of the architecture, with mostly dense and diffuse infiltrates of sheets of blastoid appearing medium to large tumor cells. Basically, three morphologic variants of DLBCL can be discerned strictly on morphologic grounds [56]. The most common morphologic subtype, centroblastic lymphoma (CB), is composed of a polymorphic mixture of centroblasts (CB) with vesicular nuclei, finely or irregularly distributed nuclear chromatin, and 2–4, sometimes prominent nucleoli, which are typically membrane-bound. The cytoplasms of archetypal CB are medium sized, and slightly basophilic, and may cytologically show various vacuoles on touch preparations. The sheets of CB are intermixed with various numbers of immunoblasts (IB), which are slightly larger than CB, show finely distributed chromatin, and have one single, centrally located, prominent nucleolus. The cytoplasms usually are more ample and basophilic and may demonstrate plasmacytoid differentiation. Centroblastic lymphoma may show multilobated nuclei, frequently in tumors arising in extranodal sites such as bone lymphomas. The immunoblastic variant is characterized by an almost pure (>90%) population of IB. Very large, strikingly polymorphic tumor cells characterize the third morphologic variant, anaplastic diffuse large B-cell lymphoma, with tumor cells showing slight resemblance to Hodgkin and Reed-Sternberg cells or cells of anaplastic large cell lymphoma (ALCL), arranged in an often sinusoidal and cohesive infiltration pattern.

Besides the three morphologic variants, further rare morphologic variants have been reported, such as spindle- or signet-ring-shaped tumor cell populations or lymphomas with a striking fibrillary matrix [57].

Tumor cells of DLBCL show a mature B-cell phenotype with expression of various B-cell markers such as CD19, CD20, CD79a, and PAX-5 together with surface and cytoplasmic immunoglobulin (mostly IgM). Expression of CD10 is variable, but can be detected in roughly 50% of pediatric cases [52]. Based on findings of the gene expression profiles of a large series of DLBCL using DNA microarrays, most of the lymphomas could be assigned to a certain molecular signature [58]. The gene expression profiles of the tumor cells were compared to signatures of their postulated physiologic counterparts, and were identified as either germinal center origin-like (GCB) or post-germinal center origin/activated B-cell-like (ABC) and type 3 subgroups. These three subgroups were demonstrated to provide fundamental survival differences in patients treated by chemotherapy, therefore the distinction of the GCB from the non-GCB subtype is an important predictive factor in DLBCL. Based on the seminal results obtained with molecular techniques, the identification of the cell of origin (COO) in almost each case of DLBCL in the daily practice of routine pathology laboratories became available soon thereafter by immunohistochemistry. With the use of expression patterns of certain antigens such as CD10, BCL-6, IRF4/MUM-1, FOX-P1, GCET-1, and LMO2, an

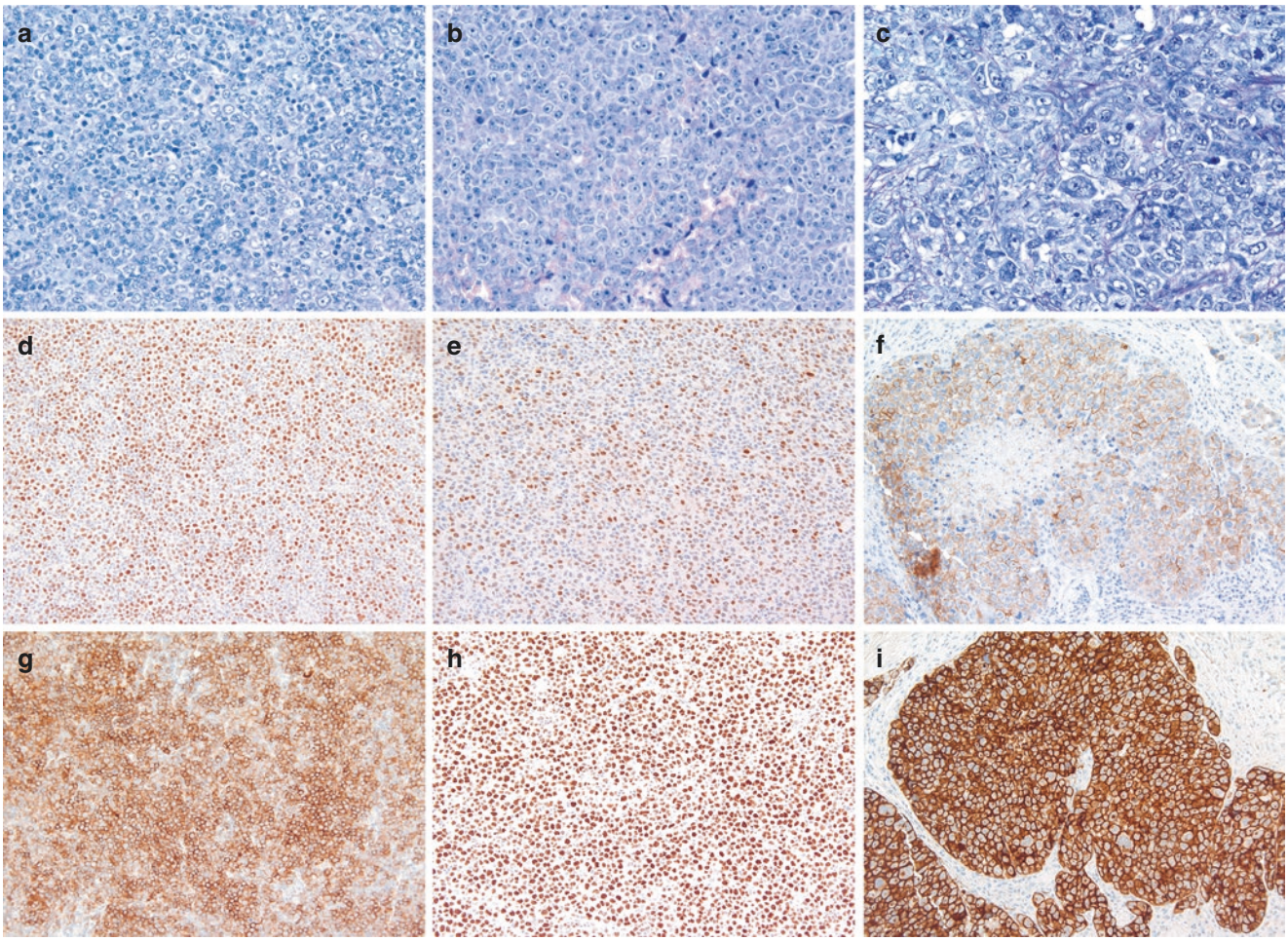

Fig. 7.3 Diffuse large B-cell lymphoma (DLBCL). DLBCL can be divided into morphologic and molecular subgroups, the latter according to cell of origin. The three common morphologic variants of DLBCL are depicted: left column represents a case with centroblastic morphology with vesicular nuclei with some small nuclear membrane bound nucleoli (**a**) with a GCB immunophenotype: positivity for BCL6 (**d**) and CD10 (**g**). The middle column shows immunoblastic morphology (**b**) with monomorphous blasts with single centrally located nucleoli and basophilic cytoplasms. Immunoblastic DLBCLs often exhibit a non-GCB immunophenotype: positivity for MUM-1 (**e**) and FOXP1 (**h**). The anaplastic variant is depicted in the right column: very large bizarre pleomorphic tumor cells (**c**) and positivity for CD30 (**i**); CD19 (**f**) is indicative of a B-cell origin

assignment to the two basic molecular subtypes of GCB and ABC (the latter also termed non-GCB) may be achieved by application of one of the three main algorithm systems. The widely used Hans algorithm [59] is a comfortably simple classifier with assessment of expression of the three antigens CD10, BCL-6, and IRF4/MUM-1 with a cutoff at 30%. Cases positive for CD10 are assigned as GCB-like, whereas cases negative for CD10 have to express BCL-6 without MUM-1 co-expression qualifying for assignment to the GCB immunophenotype. CD10-negative cases with negativity for BCL-6 or considerable MUM-1 expression are allocated to the non-GCB immunophenotype. The Choi classification algorithm [60] uses two additional markers, GCET-1 and FOX-P1, for assignment to either GCB or non-GCB immunophenotypes. The third algorithm frequently applied by a pathologist is the Visco-Young algorithm [61]; in this classification system, the application of only three markers, namely CD10, BCL-6, and FOX-P1, is needed for classification into GCB vs. non-GCB subtypes. The frequency of GCB and non-GCB subtypes varies based mainly on age, geographical distribution, and the method used for classification. Children have considerably more DLBCL with GCB immunophenotypes (80–95%) [62] than adult patients (approximately 60% GCB vs. 40% ABC). DLBCL with a GCB-like immunophenotype have an improved prognostic outcome compared to DLBCL with the non-GCB immunophenotype [59, 63]. Additionally to the antigens expressed to delineate COO, expression of MYC and BCL-2 is assessed with a cutoff of 50% for BCL-2 and 40% for MYC, respectively [64]. DLBCL with co-expression of these two markers ("double expressors") were shown to be associated with inferior survival [65] as well as an increased risk for CNS

relapse. These lymphomas were demonstrated to be more common in the ABC subgroup. A small subgroup of de-novo arising DLBCL may express CD5, associated with high-risk prognostic features, and usually assigned to the ABC subtype [66]. CD30 is expressed in approximately 20% of cases, often associated with anaplastic morphology [67].

Genetic Profile

In almost all cases, clonal rearrangements of Ig heavy- and light-chain genes can be detected. In the GCB subtype the Ig genes usually demonstrate ongoing somatic hypermutation, whereas in the ABC subtypes, already accomplished somatic hypermutation can be demonstrated [68].

Numerous studies have revealed a broad spectrum of somatic mutations within the large group of DLBCL; these mutations are in many cases associated with the specific COO. It has been shown that mutations within *EZH2* and *GNA13* are almost exclusively associated with the GCB subtype, whereas mutations for *MYD88*, *CARD11*, and *CD79b* are restricted to ABC-type lymphomas [69]. The most common translocation in DLBCL is *BCL-6* at 3q27, detected in approximately 30% of DLBCL cases [70]. Rearrangements of *BCL-2* are detected in roughly 40% of GCB lymphomas [71], whereas they are very rare in the ABC subgroup [72]. By contrast, alterations of *BCL-6* are more common in the ABC subgroup [73], as well as copy number alterations at 6q21 and 9p21. *MYC* translocation, without a concurrent translocation of *BCL-2* and/or *BCL-6* ("single hit"), is found in approximately 10% of adult DLBCL, whereas it is more often detected in pediatric cases of DLBCL, suggesting a possible relationship with Burkitt lymphoma [74]. However, unlike in BL, *MYC* translocation in DLBCL is usually associated with a more complex karyotype. *BCL-2* translocation is only very rarely seen in children [74, 75].

Differential Diagnosis

DLBCL, NOS, has to be distinguished from other blastoid B-cell lymphomas such as BL and LBL (Table 7.1).

Based on their unique clinicopathologic features large B-cell lymphomas with only a minority of lesional B cells scattered in a reactive background of numerous histiocytes and T cells, the category of T-cell/histiocyte-rich large B-cell lymphoma (THRLBCL) has been separated from the NOS group as a distinct entity. Similarly, due to the unique localization within the mediastinum, distinct clinical, (immuno) phenotypic, and genetic features, a separate category of primary mediastinal large B-cell lymphomas (PMBL) has been denoted in the WHO classification system. DLBCL with evidence of EBV association should also be assigned into a specific entity, as should DLBCL primarily arising in the CNS.

Primary Mediastinal (Thymic) Large B-Cell Lymphoma and Mediastinal Gray-Zone Lymphoma

Primary mediastinal (thymic) large B-cell lymphoma (PMBL) is a blastoid large B-cell lymphoma with a characteristic clinicopathologic presentation. It is thought to arise from medullary thymic B cells and is located in the anterior mediastinum of mainly young female adults (median age 35a, female-male ratio approximately 2:1). These tumors represent only 2–3% of NHL, and are even more infrequently denoted in children [77]. Gene expression profiling has shown an overlap between PMBL and cases of classical Hodgkin's lymphoma of the nodular sclerosing subtype (NS) [78]; hence, the WHO classification acknowledges a separate category of B-cell lymphomas, unclassifiable, with features intermediate between diffuse large B-cell lymphoma and classical Hodgkin's lymphoma (so-called mediastinal gray-zone lymphomas, MGZL). These lymphomas also reveal a predominant localization in the mediastinum, and show pathologic features that are intermediate between PMBL and cHL of mainly NS subtype [79]. In contrast to PMBL, MGZLs are more often diagnosed in young male patients, and appear to have an inferior outcome compared to PMBL [80].

Clinically, both lymphomas most often present as a large mediastinal, often bulky tumor mass with infiltration of adjacent organs such as lung, pericardium, or walls of the nearby great vessels. Progression of disease often leads to involvement of extranodal tissue, such as CNS, kidneys, or the GI tract; bone marrow involvement as well as lymph node affection of PMBL is very rare [81].

Cases of PMBL identified by gene expression profiling with a presentation outside of the mediastinum have been reported recently [78, 82, 83]; however, these extramediastinal lymphomas may be missed by routine histopathologic workup, since gene expression profiling is usually not performed.

Morphology and Immunohistology (Fig. 7.4)

PMBLs are characterized by diffuse infiltrates of mostly large-sized tumor cells with usually round or slightly lobulated nuclei and broad clear cytoplasms [84]. Occasionally cells with features of RS cells may be intermingled. Typically, tumor cell aggregates are accompanied by dense alveolar fibrosis, and in many cases necrosis is present.

The gray-zone lymphomas often show areas resembling cHL of NS subtype, with a broad spectrum of cytologic

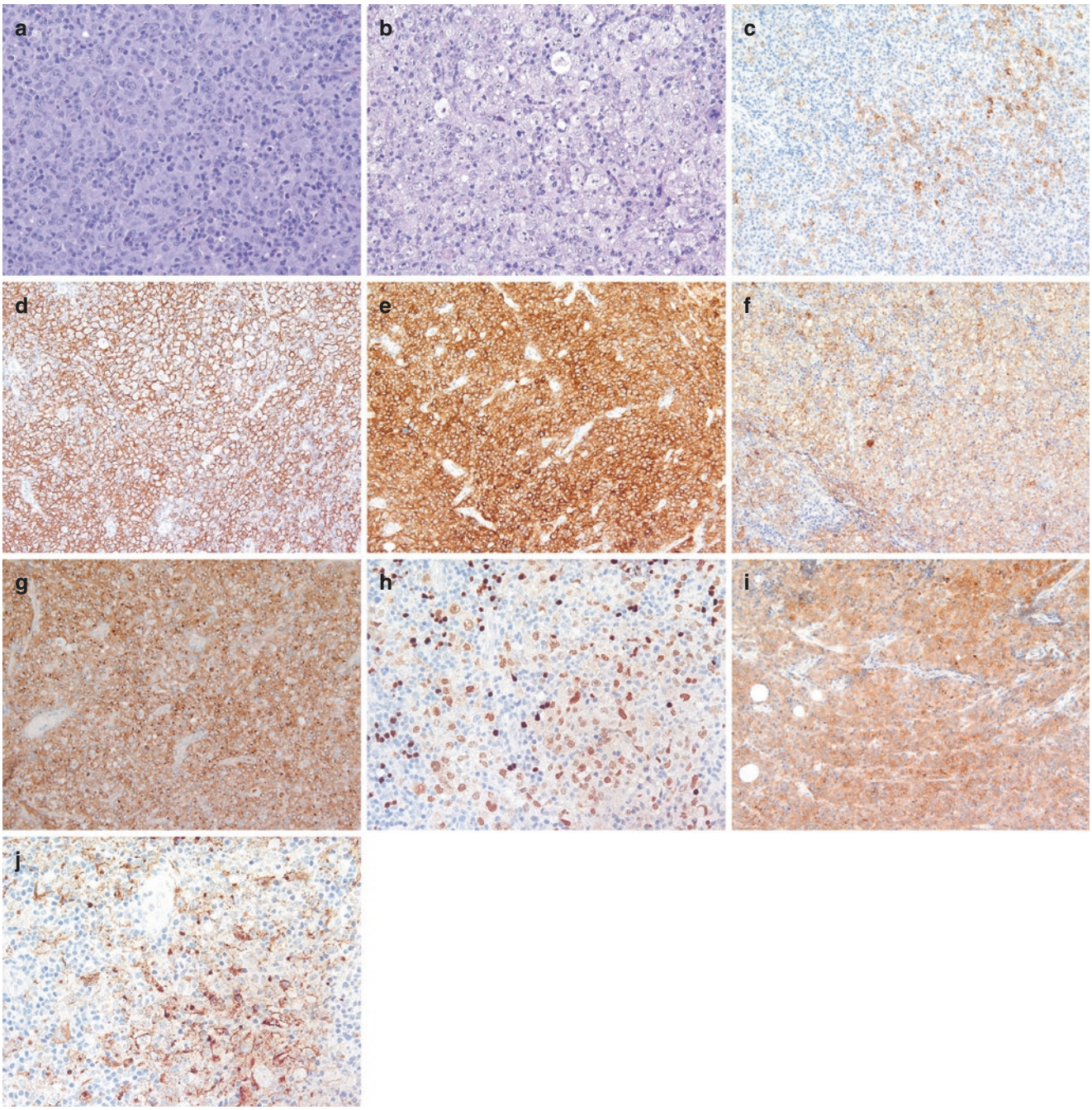

Fig. 7.4 Primary mediastinal B-cell lymphoma (PMBL) and mediastinal gray-zone lymphoma (MGZL). A typical example of PMBL is depicted in the left column: PMBL consists of large-sized tumor cells with slightly lobulated nuclei (**a**). Tumor cells are in many cases positive for CD23 (**c**), usually positive for PD-L1 (**e**), and often for CD30 (**g**). MAL (**i**) is typically positive. MGZL exhibit a broad morphologic and immunohistologic spectrum. The right column shows a cohesive diffuse infiltration of Hodgkin- and Reed-Sternberg-like blasts (**b**) with very strong positivity for CD20 (**d**) and PD-L1 (**f**), weak expression of PAX-5 (**h**) and CD15 (**j**)

appearances of the lesional cells; in some areas, the infiltrates may demonstrate a great resemblance to HL with sheets of mostly mononuclear atypical cells with occasional multinucleated RS cells intermingled in the diffuse fibrotic background of PMBL; some areas may show sheets of blastoid lymphoid cells with abundant pale cytoplasms resembling PMBL, located in a sometimes vaguely nodular fibrotic background with lymphocytes, histiocytes, and eosinophilic granulocytes, typically found in the reactive background of cHL [79, 85].

Immunophenotypically, PMBL express B-lineage-associated antigens such as CD19, CD20, CD79a, and

Table 7.2 Comparison of antigen expression in primary mediastinal B-cell lymphoma, mediastinal gray-zone lymphoma, and classical Hodgkin's disease

Antigen	PMBL	MGZL	cHL
CD30	+/−	+	+
CD20	+	+	−/+
CD79a	+	+	−/+
CD45	+	+	−
CD23	+	+/−	−
PAX-5	+	+	+ (weak)
Oct-2	+	+	−
BOB-1	+	+	−
PD-L1, 2	+	+	+
CD15	−	+/−	+
MAL	+	+	
ALK-1	−	−	−
MUM-1	+	+	+
EBV	−	−	+/−

Abbreviations: *PMBL* primary mediastinal B-cell lymphoma, *MGZL* mediastinal gray-zone lymphoma, *cHL* classical Hodgkin's disease

PAX-5. The transcription factors BOB-1 and OCT2, typically not detected in tumor cells of cHL, are strongly positive [86]. Most cases (>80%) show a weak to moderate, heterogeneous expression of CD30. CD10 is usually absent, whereas BCL-2 and BCL-6 may be variably expressed; MUM-1 is usually positive. Up to 3/4 of PMBL express CD23 [87], in contrast to other DLBCL; they were shown to express MAL antigens [88, 89] and usually reveal a strong expression of PD-L1 and PD-L2 [90].

In MGZL, the neoplastic cells usually are positive for CD45, which is in contrast to cHL cells. CD20 and CD79a are strongly expressed in most cases, as is CD30. There may be heterogeneous co-expression of CD15 together with pronounced expression of the B-cell transcription factors BOB1, Oct-2, and PU.1 [91], but there are also cases lacking strong expression of B-cell markers with pronounced expression of CD30 and CD15, thus resembling cHL immunophenotypically. A comparison of the immunophenotypic peculiarities of PMBL, MGZL, and cHL is given in Table 7.2.

Genetic Profile

Immunoglobulin genes are clonally rearranged in most instances of PMBL and MGZL. Gene expression profiling studies have revealed a unique signature of PMBL different from other large B-cell lymphomas but similar with cHL [78, 82]. In 75% of cases, alterations such as translocations, gains, and amplifications of *PDL1* at the 9p24.1 chromosomal location can be detected [82, 90], most likely leading to the overexpression of PDL1 detected by immunohistology. In roughly 50% of cases chromosomal gains at 2p16.1 are found, thereby associated with amplification of *REL* and *BCL11A* [92].

Similarly to PMBL, MGZL show genetic aberrations of the 9p24.1 locus [93] with frequent gains and amplifications of *PDL2*, as well as gains of *REL* at 2p16.1. Moreover, gains of *MYC* have been reported in up to 20% [93]. Methylation analysis demonstrated a close epigenetic relationship of PMBL, cHL, and MGZL and a unique epigenetic signature for MGZL [94], revealing a different methylation of CpG islands in contrast to PMBL and cHL, thus supporting its separation as a distinct entity of the WHO classification.

Pediatric-Type Follicular Lymphoma

Follicular lymphoma (FL) in the pediatric population differs from the "usual" FL of adulthood clinically, histologically, immunophenotypically, and also genetically. The pediatric type of FL (PTFL) was listed as a variant of FL already in the 2008 WHO classification. It primarily occurs in children and young adults, but has been reported in adults infrequently [95, 96]. Clinically, this variant often involves cervical lymph nodes or other sites of the head and neck [97], and usually presents as a limited disease with often only one singular involved lymph node (Stage I) [95]. Extranodal disease or bone marrow involvement has not been reported so far [95–98]. The disease shows a striking male predominance, with a M:F ratio of >10:1. Most affected individuals are between 1 and 18 years, with only singular reported cases >40a.

Morphology and Immunohistology (Fig. 7.5)

This FL variant is characterized by high-grade histology. The lymph node architecture is usually completely or subtotally effaced by an infiltration of large expansive follicular structures which show irregularly shaped germinal centers, often in a serpiginous infiltration pattern, lacking polarization [95]. Sometimes the infiltrates are surrounded by a rim of normal residual lymphoid tissue giving the impression of a "node-in-node" proliferation [99]. In contrast to the neoplastic follicles of classical FL, the follicles of PTFL show a starry-sky pattern with numerous tingible body macrophages. Mantle zones are usually very narrow or may even lack completely. The infiltrates are composed of a monotonously appearing population of medium-sized blastoid cells with round nuclei and rather inconspicuous nucleoli. The cytoplasms are usually rather small and only slightly basophilic, best appreciated in the Giemsa stain. Occasionally the cells resemble centroblasts. Based on the rather monotonous appearance of the cells, which fulfill the criteria of FL grade

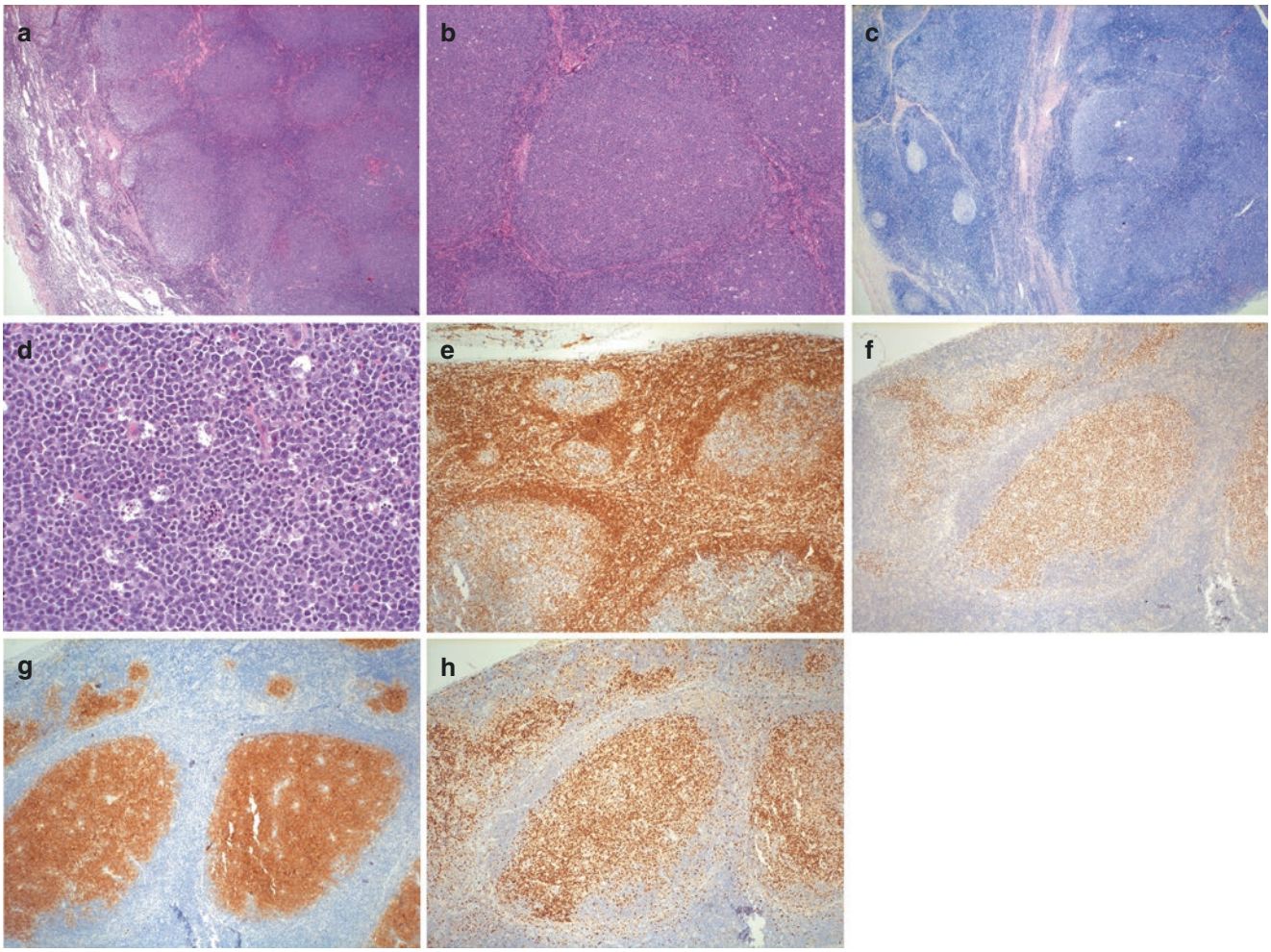

Fig. 7.5 Pediatric-type follicular lymphoma. Strictly follicular infiltration of the lymph node with a "node-in node" appearance denoted in (**a**) and (**c**); (**b**) shows a neoplastic follicle without polarization, lacking a well-defined mantle zone; the neoplastic follicles are composed of medium-sized blastoid cells with numerous starry-sky macrophages intermingled (**d**); most tumor cells are immunohistologically bcl-2 negative (**e**), bcl-6+ (**f**), and CD10+ (**g**) and exhibit a high proliferation fraction with Ki-67 (**h**)

3, grading is not done in these cases [100]. Mitotic figures are quite numerous. By definition, diffuse areas with morphologic features of DLBCL must be absent [96].

The lesional cells show a mature B-cell immunophenotype with expression of CD19, CD20, CD79a, PAX-5, and CD22. In many cases the expression of the B-cell markers is restricted to the purely follicular infiltrates, whereas interfollicular B cells may be lacking. Equally to the counterpart of classical FL, the cells in PTFL show strong expression of BCL-6 and CD10 [101]. By contrast yet, they usually lack expression of BCL-2, but weak expression by a minority of cells is occasionally seen [98]. The attenuated mantle-zone cells may be highlighted wit IgD and CD23. The proliferation fraction (Ki-67) usually reveals a high proliferation rate of >30%, again lacking a polarization which is a typical finding in non-neoplastic hyperplastic follicles. IRF-4/MUM-1 is always negative. MUM-1 positivity should raise the possibility of LBCL with *IRF-4* rearrangement. CD43 expression, which is rarely seen in classical FL, has been reported in PTFL in up to 25% of cases [100].

Genetic Profile

PTFL show clonal Ig rearrangements by PCR-based clonality detection techniques. In contrast to classical FL, they do not reveal translocations for *BCL-2* and *BCL-6*, and do not reveal *IRF-4* aberrations. The most common genetic aberrations are deletions of 1p36 affecting *TNFRSF14* [102, 103], similar to the classic FL. Recently, mutations of the *MAP2K1* were reported in up to 50% of PTFL cases [104]. *TNFRSF14* and *MAP2K1* mutations occur independently in most cases, suggesting that both mutations may play an important role in PTFL lymphomagenesis [105].

Differential Diagnosis

PTFL has to be differentiated from classical follicular lymphoma. In most cases morphology and immunophenotypic analysis, combined with the characteristic clinical presentation allows accurate distinction (Table 7.3). Classical follicular lymphoma is very rare under the age of 18, hence a diagnosis of usual FL should be made extremely warily in this age group, and, if so, only with the utilization of all diagnostic techniques, including demonstration of *BCL-2* rearrangements by FISH. The distinction between PTFL and reactive causes, mainly florid follicular hyperplasia, may sometimes provide diagnostic problems. The key diagnostic feature in distinguishing PTFL from reactive processes is architectural effacement of the enlarged lymph nodes. It may be noteworthy that cases of florid follicular hyperplasia, mainly arising in the head and neck region of young boys, may present with populations of clonal CD10+ B cells by FACS immunophenotyping [106]. PTFL have to be distinguished from large B-cell lymphomas with *IRF-4* rearrangements and from pediatric-type nodal marginal-zone lymphomas (detailed description see below) (Table 7.4).

Table 7.3 Clinicopathologic characteristics of classical and pediatric-type follicular lymphoma

	Classical FL	PTFL
Median age	6th decade	<40a
M:F	1:1,7	10:1
Stage at presentation	40–70% stage IV	I–II
Predilection sites	Widespread disease	Head and neck, solitary LN
Bone marrow involvement	40–70%	–
Diffuse areas	Common	–
Composition of follicles	Centrocytes, centroblasts	Blastoid intermediate-sized cells
Grading	Grade 1, 2, 3	– (grade 3 morphology)
Polarization of follicles	–	–
Starry-sky pattern	–	+
Interfollicular proliferation	+	–
CD10+/BCL-6+	+	+
BCL-2+	+ (90% grade 1/2, <50% grade 3)	usually may show weak staining in a minority of cells
Ki-67	<20% (grade 1/2), >20% grade 3	>30%
t(14;18) (q32;q21)	+ (85–90%)	–

Abbreviations: *FL* follicular lymphoma, *PTFL* pediatric-type follicular lymphoma, *LN* lymph node

Table 7.4 Clinicopathologic characteristics of pediatric indolent B-cell lymphomas

	PTFL	LBCL, IRF4 +	PMZL
Age range	<40a (median 11a) [100]	4-79a (median 12a) [107]	2-27a (median 16a) [109]
Male:Female	10:1	1:1	20:1
Localization	LN head and neck	Waldeyer ring, LN head and neck	LN head and neck
Morphologic pattern	Purely follicular, grade 3	Diffuse, occasionally follicular	Large follicles, diffuse areas, often PTGC
CD10+	+	+	–
BCL-2+	–/+ (weak)	+	+
BCL-6+	+	+	–
IRF4/MUM-1	–	+	–
CD43+	–	–	+
Clonality	+	+	+

Abbreviations: *PTFL* pediatric-type follicular lymphoma, *LBCL* IRF-4+, large B-cell lymphoma, IRF-4+, *PMZL* pediatric marginal-zone lymphoma, *PTGC* progressive transformation of germinal centers, *LN*, lymph node

Large B-Cell Lymphoma with *IRF4* Rearrangement

Large B-cell lymphoma (LBCL) with *IRF4* rearrangement is a rare form of B-cell lymphoma characterized by strong immunohistologic expression of IRF4/MUM-1, in most cases accompanied by rearrangement of the *IRF4* gene [107]. It occurs predominantly in children and young adults and shows a predominant predilection site of the head and neck regions and involvement of the Waldeyer ring [95]. In contrast to PTFL, the sex distribution is equal.

Morphology and Immunohistology (Fig. 7.6)

The lymphomas present either with follicular, follicular and diffuse, or purely diffuse infiltration patterns. The cells are medium- to large-sized blastoid-appearing cells with open chromatin and small nucleoli. Notably and in contrast to PTFL, in follicular structures starry-sky macrophages are very infrequently seen, and mitotic figures are not quite numerous. The neoplastic follicular structures repeatedly are arranged in a back-to back pattern, with only small or no mantle zones, lacking serpinginous follicles frequently encountered in PTFL [107]. A portion of cases reported present with an entirely diffuse growth pattern.

The follicular and diffuse infiltrates are composed of B cells with a mature immunophenotype, with expression of CD20, CD19, CD79a, PAX-5 with frequent co-expression of bcl-6 and bcl-2 [108]. MUM-1/IRF-4 is typically and consis-

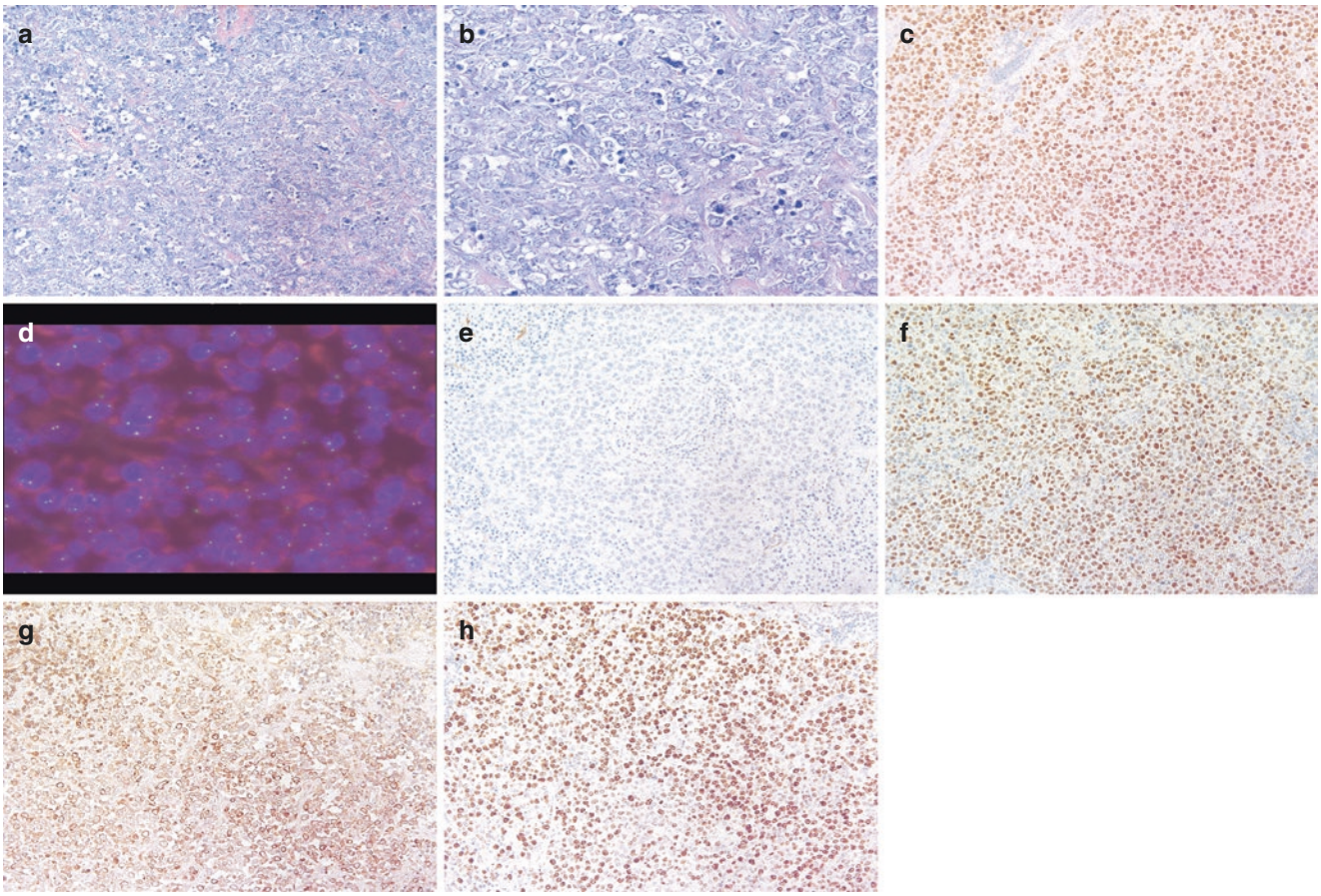

Fig. 7.6 Large B-cell lymphoma with *IRF4* rearrangement (LBCL, IRF4+). LBCL consists of medium to large cells (**a**) and (**b**) with characteristic positivity for IRF/MUM-1 protein (**c**) and rearrangement of the *IRF4* gene. (**d**) Fluorescence in situ hybridization with an *IRF4* (6p25) break-apart probe: one allele shows co-localization of both probes (yellow signal), whereas the other allele shows separation of signals (separate red and green signals) indicative of translocation. CD10 (**e**) is negative in roughly ¼ of cases. BCL6 (**f**) and BCL2 (**g**) are frequently expressed. The lymphomas usually show a high proliferation rate (**h**)

tently strongly expressed. Weak to moderate CD10 expression has been reported in roughly 2/3 of cases [99, 107]. These lymphomas usually show a high proliferation fraction.

Genetic Profile

The lymphomas consistently show clonal rearrangements of the Ig genes and in almost all cases a rearrangement of the *IRF4* gene with the *IgH* locus, whereas the light-chain gene locus is only occasionally involved. Interestingly, some cases exhibit alterations of the *BCL-6* gene locus [99, 107], whereas *MYC* and *BCL-2* rearrangements are absent.

Pediatric Marginal-Zone Lymphoma

Pediatric marginal-zone lymphoma (PMZL) shows distinct clinical and histologic characteristics, therefore representing a specific subtype among nodal marginal-zone lymphomas in the WHO classification 2016. It is a very rare type of an indolent lymphoma most often occurring in one single lymph node of the head and neck region, almost solely in boys and young males (male-female ratio 20:1). It has an excellent prognosis with a very low recurrence rate.

Morphology and Immunohistology (Fig. 7.7)

The histologic presentation is quite similar to conventional marginal-zone lymphomas presenting in the adult population; however, often large follicles with resemblance to progressively transformed germinal centers (PTGC) are denoted, in context with marked expansions of the marginal zone. The mantle-zone cells frequently infiltrate the residual fragmented germinal centers. Sometimes, the mantle zones are only difficult to discern without immunohistologic stains.

The cells show a mature B-cell phenotype with expression of CD20, CD79a, CD19, and PAX-5, with frequent co-expression of CD43 [109]. Light-chain restriction of the

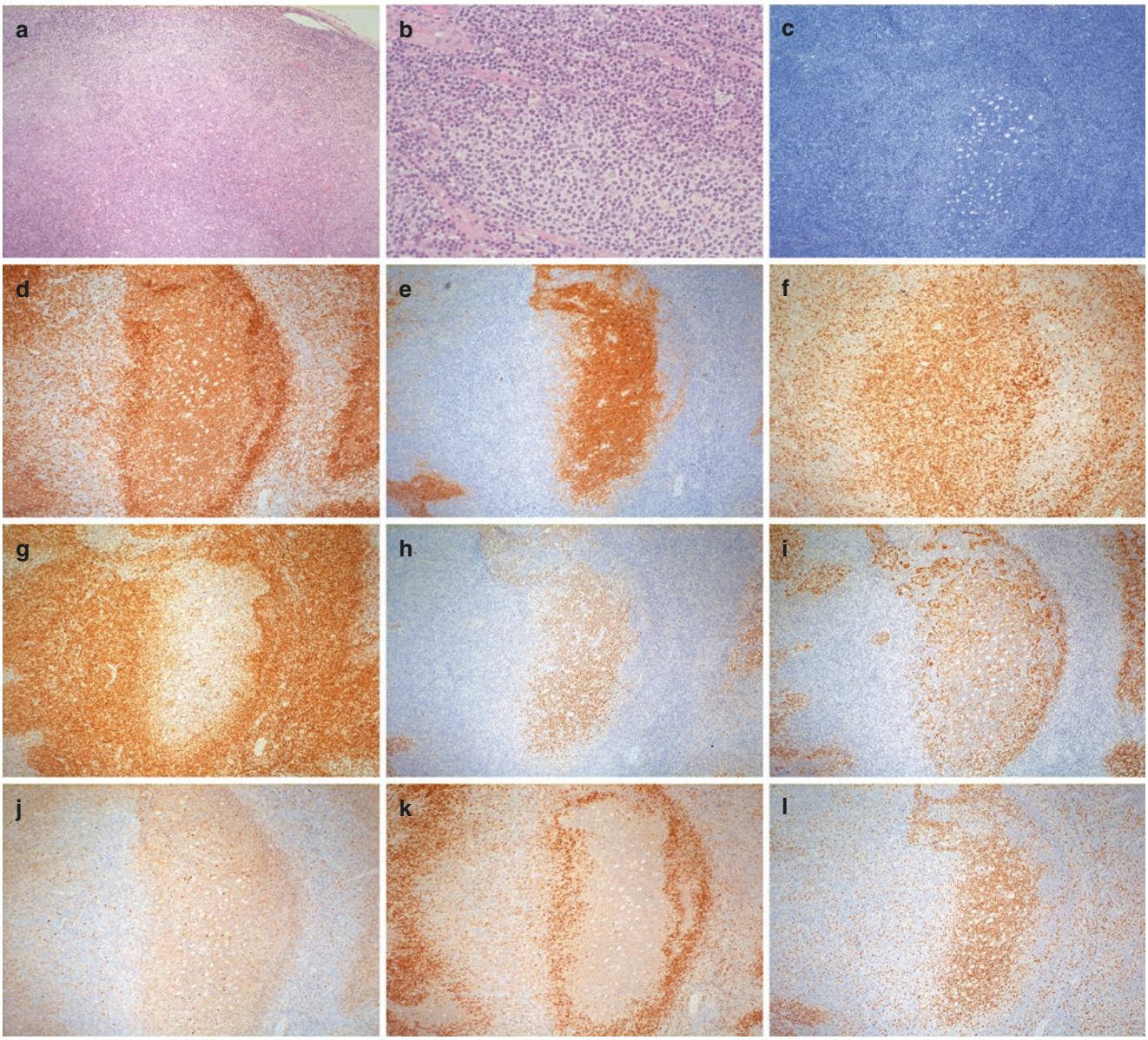

Fig. 7.7 Pediatric marginal-zone lymphoma. Large, ill-defined follicles with markedly expanded marginal zones (**a**) and (**b**); the reactive enlarged germinal centers contain numerous starry-sky macrophages and mitotic figures, but show narrow mantle zones (**c**). CD79a depicts the reactive germinal center, the darker stained remnants of the mantle-zone and the marginal-zone cells (**d**); germinal center cells are positive with CD10 (**e**) bcl-6 (**h**) and IgM (**j**) and negative with bcl-2 (**g**); most marginal-zone cells are positive with CD43 (**f**); CD21 highlights the marked expansion and disruption of the FDRC meshwork within the reactive germinal center (**i**); the remains of the mantle zone are denoted with IgD (**k**); Ki-67 shows highly proliferating cells in the germinal center and several proliferating marginal-zone cells (**l**)

marginal-zone cells can be detected with antibodies against kappa and lambda. IgD staining highlights residual mantle-zone cells arranged in irregular and expanded patterns, whereas CD10 and BCL-6 show positivity of the residual germinal center cells. The proliferation fraction is rather low.

Genetic Profile

Clonal rearrangements of the Ig heavy- and light-chain regions can be detected by PCR in almost all cases. Trisomy 18 and trisomy 3 have been reported in a proportion of cases [110].

Differential Diagnosis

The differential diagnosis with marginal-zone hyperplasia may be difficult [111], since these conditions may present with monotypic immunoglobulin expression, and they may also express CD43 [112]. Interestingly, an association with *Haemophilus influenzae* infection has been reported [111]. Molecular studies in these hyperplasias, however, fail to show monoclonality; therefore, in all cases with attenuation of the marginal zone, molecular studies are strongly recommended. PMZL has to be distinguished from PTFL and LCL with IRF4 rearrangements (Table 7.4).

Anaplastic Large Cell Lymphoma, ALK+

Anaplastic large cell lymphoma, ALK+ (ALCL, ALK+), is a T-cell lymphoma characterized by large pleomorphic tumor cells with typically strong expression of CD30 and rearrangement of the *ALK* gene on chromosome 2p23 [113] with various translocation partners, the most common binding-partner thereby being Nucleophosmin (*NPM*) on chromosome 5q35. Those nodal-based tumors, which share morphologic and immunomorphologic similarities with the ALK-positive cases, but lack this peculiar genetic aberration, are summarized in a separate entity in the WHO classification 2016, the ALK-negative ALCL. Similarly excluded from this category of ALK+ ALCL are primary cutaneous CD30+ ALCL cases as well as B-cell lymphomas with anaplastic morphology.

ALK+ ALCL occurs predominantly in children and young adults, thereby representing up to 20% of all pediatric lymphomas [2, 38]. It has been shown that up to 90% of ALCL in children were ALK+, whereas only roughly 30% of adult cases were associated with *ALK* rearrangements [114]. ALCL has a slight male preponderance with a male to female ratio of 1.5:1. ALCL frequently involves both lymph nodes and extranodal sites such as skin, bone, or soft tissues [115, 116]. Most patients present with widespread disease at the time of diagnosis and frequently demonstrate only subtle bone marrow involvement, sometimes only detected by immunohistology [115, 117]. B-symptoms, such as intermittent fever, are quite typical [118].

Morphology and Immunohistology (Fig. 7.8)

Morphologically, the spectrum of ALCL encompasses a great variety of different morphologic patterns [119], sometimes coexisting in one and the same tissue affected [120]. All morphologic variants, however, should harbor a characteristic large "hallmark cell" at least to a minor extent. This eponymous anaplastic cell is characterized by an eccentrically located, typically horseshoe- or kidney-shaped nucleus with a characteristic paranuclear eosinophilic region. Smaller variants of these cells may be discerned in some cases, as well as cells with nuclear (pseudo-)inclusions, the latter also being referred as "doughnut" cells. In case of lymph node affection, ALCL usually shows a paracortical and often sinusoidal infiltration pattern of cohesively growing, highly polymorphic medium- and large-sized tumor cells, thereby often simulating carcinomatous infiltration. The most common morphologic type of ALCL is the common type, which accounts for approximately 60% of all cases [120, 121]. This subtype is composed of mainly large, pleomorphic, sometimes multilobated tumor cells. The frequent hallmark cells may resemble Reed-Sternberg cells, but usually have less prominent nucleoli. The nuclear chromatin is irregularly distributed or finely dispersed. The cytoplasms are abundant in most cases, and show a characteristic dove-grey color in the Giemsa-stain. The lymphohistiocytic morphologic variant accounts for approximately 10% of cases and is characterized by typical, but usually slightly smaller tumor cells intermixed with numerous reactive histiocytes [120, 122], sometimes masquerading the true neoplastic nature of the process. The neoplastic cells tend to cluster around vessels and can be highlighted by immunohistology with antibodies against CD30 and ALK-1. One further pattern (5–10%) is the small cell variant, composed mainly of a population of rather monomorphic, small cells with slightly irregular nuclei [120, 123]. Also in this variant, a pronounced accumulation of small cells together with hallmark cells can be discerned in the vicinity of blood vessels [120]. The cells in this variant may also appear as so-called fried egg cells with centrally located nuclei and abundant pale cytoplasms. This is the variant mostly misdiagnosed as peripheral T-cell lymphoma (PTCL). An additional rare morphologic variant, the Hodgkin-like pattern, exists, mimicking the nodular sclerosis subtype of classical Hodgkin's lymphoma [124]. Other not so well-characterized rare variants include the sarcomatoid, signet-ring, neutrophil-rich and giant cell types [1]. In up to 20% of cases, more than one morphologic pattern is seen in a single affected site ("composite pattern") [120]; in cases of relapse, morphologic patterns may change [125].

By immunohistology, tumor cells express CD30 in a membrane-bound fashion, often with a dot-like enhancement of the Golgi zone. Most tumors show an aberrant T-cell immunophenotype with frequent antigen loss. Using a large panel of T-cell antibodies such as CD2, CD3, CD4, CD5, CD7, and CD8 will accomplish assignment to the T-cell lineage in most cases [126]. The most frequent T-cell-associated antigens deleted in ALCL are CD3, CD5, and CD7 [6, 127], whereas CD4 and less specific T-cell markers such as CD45RO and CD43 are expressed more frequently. The remaining cases without demonstration of any T-cell markers are labeled "null cell phenotype" but show evidence of T-cell differentiation on the genetic level. T-cell and null cell types of ALCL do not show clinicopathologic or genetic differences and are therefore regarded as a single entity [120]. Most cases of systemic ALCLs express EMA and CD25, and they also recurrently express cytotoxic markers such as TIA-1, Granzyme-B, or Perforin [128]. CD15 is usually absent, but may be weakly expressed in a subset of tumor cells [120]. Macrophage-associated antibodies, such as PGM-1 or CD163 give negative staining results, and EBV is constantly negative in ALCL [129]. By definition, ALK+ ALCL express ALK proteins, thereby exhibiting both nuclear/nucleolar and a cytoplasmic staining pattern in cases with underlying NPM-ALK translocation [120, 121]. An interesting feature of the small cell variant is the almost exclusive nuclear staining pattern of ALK [120, 126], whereas the larger cells clustering around vessels normally show ALK staining in both

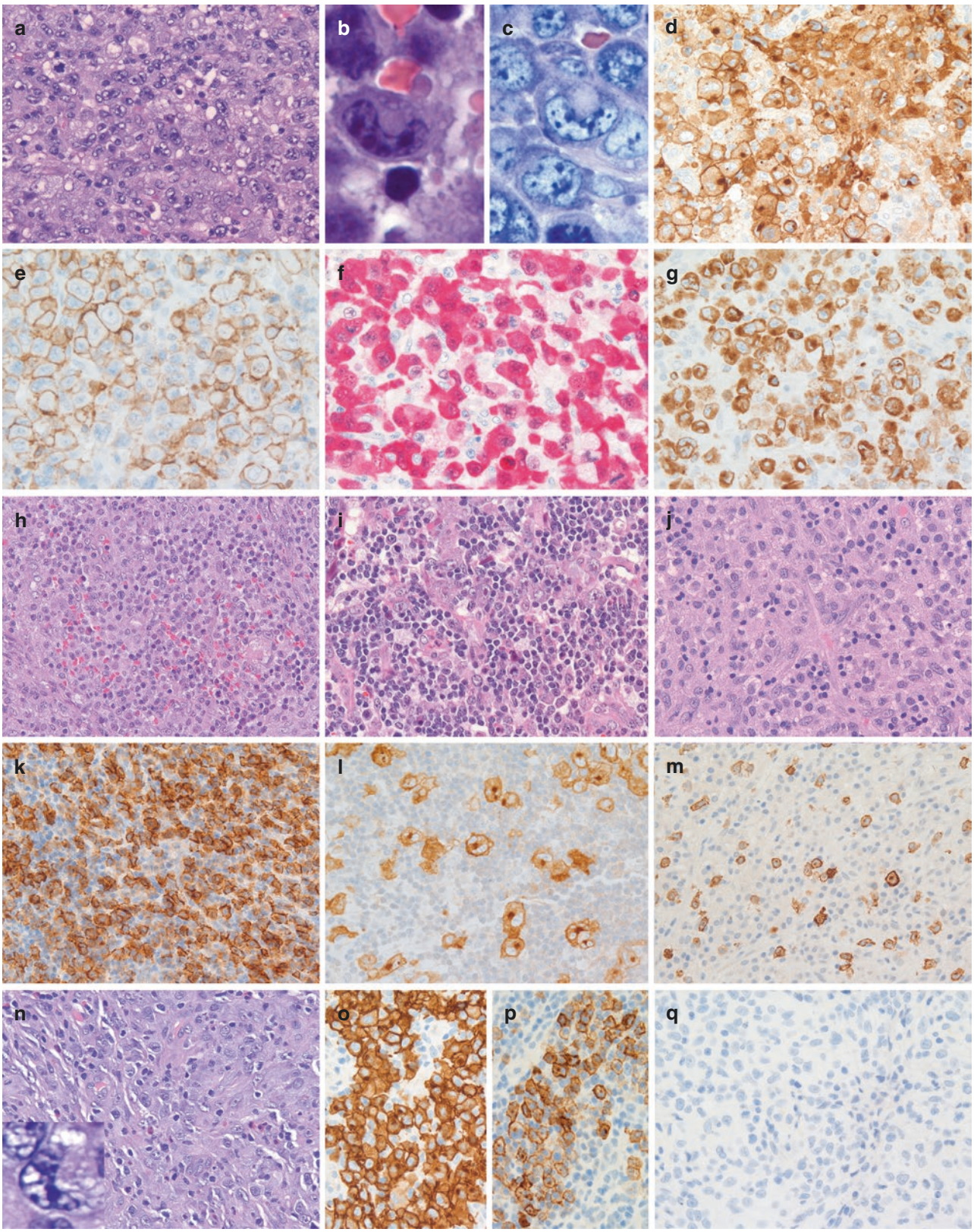

Fig. 7.8 Anaplastic large cell lymphoma (ALCL). Anaplastic large cell lymphoma (ALCL): ALK-positive ALCL, common type (**a**–**g**) is a tumor composed of blasts occasionally exhibiting "horseshoe" or kidney-shaped nuclei (hallmark cells; **b** and **c**). CD30 (**d**) and CD43 (**e**) are strongly positive, and ALK1 is typically expressed in both nuclei and in cytoplasms (**f**) in case of NPM-ALK translocation. Cytotoxic markers are frequently expressed (Perforin, **g**). Morphologic subtypes are the small cell variant (**h**), (**k**: CD30), Hodgkin-like ALCL with HRS-like cells (**i**), (**l**: CD30). Lymphohistiocytic variant of ALCL is shown in (**j**), (**m**: CD30). ALK-negative ALCL (**n**–**q**) is morphologically indistinguishable from ALK-positive ALCL. Hallmark cells are present (insert n); tumor cells show strong expression of CD30 (**o**) and EMA is frequently positive (**p**); ALK1 is per definition negative (**q**)

subcellular localizations. 20–25% of cases, however, exhibit an exclusive cytoplasmic ALK staining pattern [120, 121], and these cases were shown to harbor variant translocations involving the *ALK* gene on 2p23, but not the *NPM* gene. These partners include genes such as *TPM3*, *ATIC*, *TFG*, *CLTC*, *MSN*, or *TPM4* [130–133]. Fusion of these partners with *ALK* results in upregulation of ALK expression and activation of its kinase function. The superior prognosis of *ALK*-rearranged cases to those without *ALK* alterations is independent of the various *ALK* fusion partners [134].

Differential Diagnosis

ALCL, ALK+, has to be differentiated from ALK-negative systemic cases, which are listed under a separate entity in the WHO classification [1], as well as from primary cutaneous ALCL (C-ALCL), which is included as a specific type within the spectrum of primary cutaneous CD30-positive T-cell lymphoproliferative disorders [1]. Primary C-ALCL typically occurs in older patients, and many cases undergo spontaneous regression without treatment in contrast to the systemic cases. ALK staining is rarely found in primary C-ALCL [135], whereas C-ALCL is sometimes associated with a *DUSP22-IRF4* rearrangement on chromosome 6p25.3 [136], also found in several cases of systemic ALK-negative ALCL [137] as well as in rare cases of lymphomatoid papulosis [138].

It is important to distinguish ALCL from classical Hodgkin's lymphoma (cHL), not only due to fundamentally different treatment strategies. Although ALCL and cHL share some common features such as strong expression of CD30 and atypical anaplastic cells, they are biologically and genetically, as well as clinicopathologically different diseases.

ALK+ ALCL also has to be distinguished from a rare B-cell lymphoma expressing the ALK protein, which has striking immunoblastic/plasmablastic features [1]. These ALK+ LBCL show a typical granular cytoplasmic ALK staining, frequently associated with a *CLTC-ALK* fusion, also sometimes found in ALK+ ALCL; rare cases may even contain the "classical" *NPM-ALK* translocation [139]. However, these lymphomas are CD30-negative and express plasma cell markers such as CD38 and VS38c together with MUM-1 and EMA, but lack the B-cell markers CD20, CD79a, and PAX-5 [140].

ALK+ALCL has to be distinguished from various non-hematopoietic tumors, which may show weak to moderate ALK-positivity. Aberrant expression of ALK has been observed in a variety of pediatric cancers, including glioma, rhabdomyosarcoma, inflammatory myofibroblastic tumor, and Ewing sarcoma [141].

Anaplastic Large Cell Lymphoma, ALK-Negative

ALK-negative ALCL has recently been listed as a separate entity in the WHO classification of hematopoietic and lymphoid tumors [1]. It is morphologically indistinguishable from ALK+ ALCL and lacks, by definition, any association with *ALK* alterations. It usually occurs in adults, and is only very rarely seen in the pediatric population [126, 134, 142]. It affects both nodal and extranodal tissue, and patients frequently present with widespread disease.

Morphology and Immunohistology (Fig. 7.8)

ALK-negative ALCLs show identical morphologic features as their ALK+ counterparts with the exception that morphologic variant patterns should not be prominent. Hence, in almost all cases large classical type anaplastic tumor cells, including hallmark cells arranged in a cohesive pattern are detected [137].

The immunophenotype is – with the exception of ALK expression – identical to ALK+ ALCL, with uniformly strong expression of CD30, and about half of the cases co-express EMA and one or more T-cell markers, most frequently CD2, CD4, and CD3, more often than CD5 [137]. CD43 is usually positive, and significant expression of cytotoxic markers such as TIA-1, Granzyme-B, and Perforin is frequently seen. Interestingly, cases with rearrangements of *DUSP22-IRF4*, which occurs in roughly 30% of the ALK-negative ALCLs, frequently lack expression of cytotoxic markers [137]. Cases with *TP63* rearrangements (detected in about 8% of cases) show overexpression of p63 proteins; however, the protein expression is not always associated with actual rearrangement of *TP63* on the genetic level, since it has also been found in non-rearranged cases [137].

Differential Diagnosis

The main differential diagnosis of ALK-negative ALCL is peripheral T-cell lymphoma (PTCL). PTCL can express CD30, but the staining is usually weaker and more heterogeneous. It has to be noted that some PTCL may even show large pleomorphic tumor cells arranged in a cohesive infiltration pattern, thus strict morphologic and immunophenotypic criteria have to be applied in questionable cases. Cases with evidence of either *DUSP22* or *TP63* rearrangements are considered as ALCL [137, 142].

ALK-negative systemic ALCL has to be distinguished from C-ALCL, which may have a similar morphology and

immunophenotype, and may even harbor a *DUSP22* translocation [143]. Thorough clinical work-up is therefore mandatory in these cases.

Peripheral T-Cell Lymphoma

Peripheral T-cell lymphoma (PTCL) is a heterogeneous group of lymphomatous diseases and only rarely encountered in the pediatric population [144]. The WHO classification 2016 lists several subtypes and entities. The most common subtype in adults is PTCL, NOS, and this subtype is also the most common entity seen in children [145], followed by extranodal NK/T-cell lymphoma, hepatosplenic T-cell lymphoma, and subcutaneous panniculitis-like T-cell lymphoma, while T-cell lymphomas with a follicular T-helper (FTH) cell immunophenotype, including angioimmunoblastic T-cell lymphoma (AITL), seem to be exceedingly rare in childhood [144, 146].

Because of this scarcity of PTCL in the pediatric population, only brief descriptions of the major pathologic findings are provided in the following section.

Peripheral T-Cell Lymphoma, NOS

PTCL, NOS, are lymphomas without characteristics of specifically designated subtypes of mature TCL in the WHO classification [1]. These lymphomas are mainly nodal-based diseases, with infrequent involvement of the peripheral blood and bone marrow, as well as the skin and the GI tract.

Morphology and Immunohistology (Fig. 7.9)

The affected lymph nodes usually show destruction of the normal architecture by diffuse, mainly paracortical infiltrates of neoplastic T cells with a broad range of cytologically diverse neoplastic T cells [147]. The cells may be small, medium, or large sized and slightly or even highly pleomorphic, with irregularly shaped, sometimes hyperchromatic nuclei, showing mildly basophilic or occasionally clear cytoplasms. There may be a substantial inflammatory background with eosinophilic granulocytes, histiocytes and plasma cells, and sometimes Reed-Sternberg cells may be intermingled, and also EBV+ large B cells may be seen. Immunophenotypically, PTCL, NOS are composed of CD3+/TCRbeta+ and mostly CD4+ T cells with frequent losses of antigens such as CD5 and CD7. Some cases may show CD30 expression, which is inhomogeneous and not as strong as in ALCL. By definition, these lymphomas lack pronounced expression of more than two or three FTH antigens, such as CD10, BCL-6, PD1, CXCL13, ICOS, and CXCR5 [1].

Extranodal T-/NK-Cell Lymphoproliferative Disorders (EBV-T/NK-LPD)

EBV-T/NK-LPDs are associated with chronic active EBV infection and are more prevalent in East Asia and Central and South America. There are two major categories listed under this term, which exhibit overlapping clinical and pathologic features. These conditions are systemic EBV-T/NK-lymphoma of childhood and chronic active EBV infection (CAEBV), the latter presenting with a large spectrum of clinical syndromes, lasting from localized indolent forms such as hydroa vacciniforme-like LPD to a systemic illness characterized by clonal proliferation of EBV-infected T or NK cells.

The systemic lymphomatous form (EBV+ TCL of childhood) is a clonal proliferation of cytotoxic T cells, which occurs shortly after acute EBV infection, and is usually accompanied by hemophagocytic syndrome [148, 149].

Morphology and Immunohistology

A pronounced proliferation of histiocytes, frequently exhibiting hemophagocytosis together with a proliferation of cytotoxic CD8+ T cells is found in the spleen, the liver, and the bone marrow [149]. In case of lymph node affection, in the beginning a relative preservation of the architecture is noted, followed by depletion of B-cell areas, with an often paracortical and interfollicular proliferation of only slightly atypical T cells with expression of CD2, CD3, CD8, EBER-1, TIA-1, and GB-7, but negativity for LMP-1 and CD56 [149].

Hepatosplenic T-Cell Lymphoma

Hepatosplenic T-cell lymphoma is an aggressive systemic extranodal lymphoma characterized by proliferation of T cells with a gamma/delta T-cell receptor equipment, which infiltrate preferentially splenic and liver tissue, without involvement of peripheral lymph nodes. It is usually accompanied by systemic symptoms and shows infiltration of the bone marrow in almost all cases [1]. A substantial number of HSTCL is noted in the context of immunosuppression [150].

Morphology and Immunohistology

The tumor cells infiltrate the cords and the sinusoids of the spleen; within the liver a striking sinusoidal infiltration pattern is typical, similar to the infiltration pattern in the bone marrow, which may be subtle and overlooked without immunohistologic staining. The neoplastic lymphocytes are small to medium sized with slightly irregular nuclei and pale cytoplasms. Immunologically, the cells are characterized by expression of CD3, TCR gamma-delta, and TIA-1, and the neoplastic cells frequently are "double-negative" (CD4-, CD8-). They usually show loss of CD5 antigen expression [1, 150].

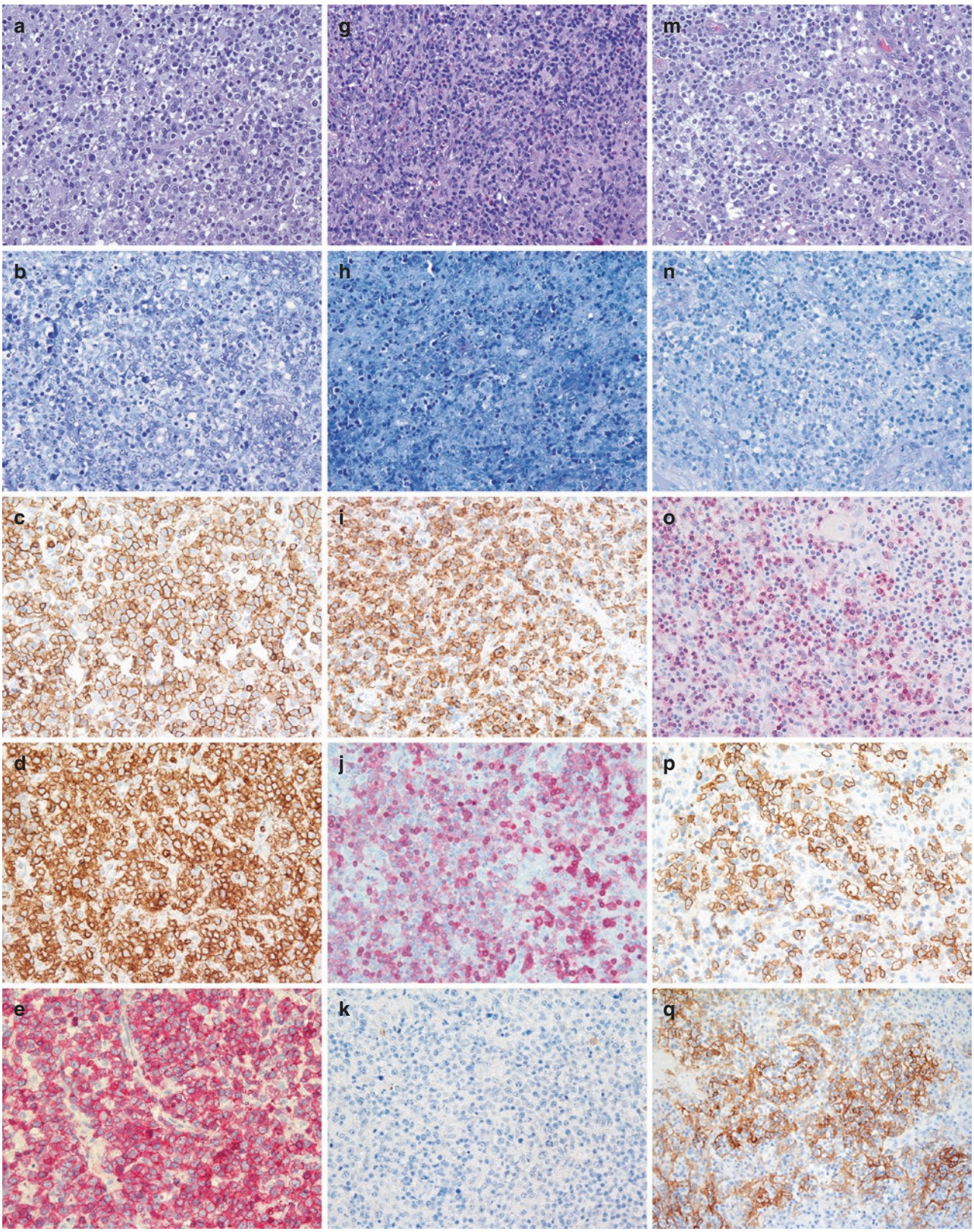

Fig. 7.9 Peripheral T-cell lymphoma (PTCL). Peripheral T-cell lymphoma, NOS, may show diverse morphologies (**a** and **g**), with a medium to large cell variant (**a**–**f**) or a small cell variant (**g**–**l**). T-cell markers such as CD2 (**d**) and CD3 (**j**) help define T-cell origin. Loss of markers normally present on non-neoplastic T cells (e.g. CD7, f) may be noticed. CD30 is negative in this case of small cell PTCL (**k**). ALK1 is always negative (**l**). Angioimmunoblastic T-cell lymphoma (**m**–**r**) is a distinct nodal subtype of PTCL, composed of characteristic clear cells (**m**) with expression of T-cell markers such as T-cell receptor beta-chain (βF1, o) arranged around vessels with an activated endothelium. Neoplastic cells express a follicular T-helper cell phenotype with positivity for PD1 (**p**). Networks of CD21+ follicular dendritic cells are characteristically expanded (**q**) and EBV is detectable in bystander B cells (EBV-ISH, **r**)

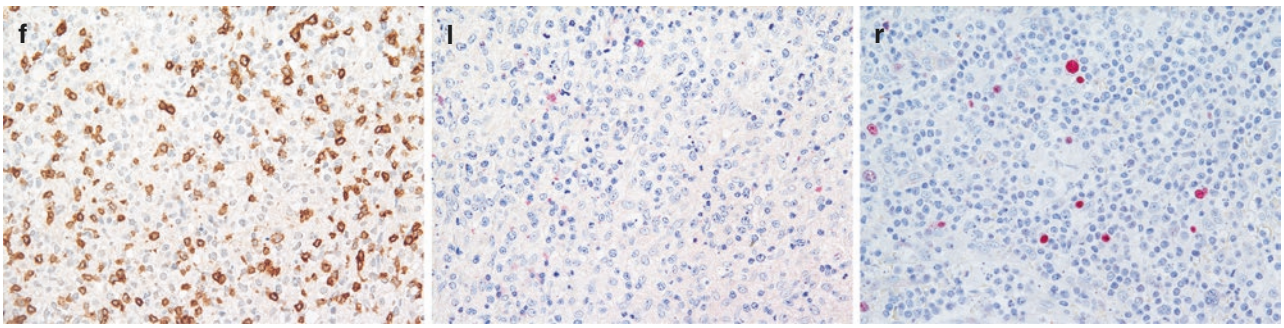

Fig. 7.9 (continued)

Subcutaneous Panniculitis-Like T-Cell Lymphoma

Subcutaneous panniculitis-like T-cell lymphoma (SPTCL) is a cytotoxic T-cell lymphoma rarely reported in the pediatric population [151, 152]. Patients usually present with widespread subcutaneous nodules, mainly on the extremities and trunk. It is occasionally associated with a hemophagocytic syndrome [152] and may show overlapping features with lupus panniculitis [153].

Morphology and Immunohistology

The epidermal and dermal compartments are usually inconspicuous, whereas the subcutaneous tissue shows mainly lobular infiltration of atypical T cells with numerous histiocytes with vacuolated cytoplasms. Rimming of adipocytes by atypical hyperchromatic lymphocytes is a constant and typical feature, as are fat necrosis, and karyorrhectic debris. Other inflammatory cells, such as plasmacytoid dendritic cells and plasma cells, which are a common feature in lupus panniculitis, are typically absent [154]. Immunophenotypically, the T cells express a mature CD3+/CD8+ T-cell immunophenotype with expression of the TCR alpha-beta and cytotoxic markers such as TIA-1, GB7, and Perforin. They are negative with CD56 [153]. Besides differential diagnostic overlap with lupus panniculitis, the distinction from cutaneous gamma/delta T-cell lymphoma may be difficult, but clinically essential; this peculiar lymphoma subtype usually shows a pronounced dermal and epidermal component together with panniculitis-like features, and shows a gamma/delta TCR equipment, which can be confirmed by immunohistology reliably on paraffin-embedded tissue sections [155].

Nodal Lymphomas of Follicular T-helper (FTH) Cell Origin, Including AITL

PTCL with a FTH immunophenotype were separated from the PTCL, NOS, category in the recent revised WHO classification 2016 [1]. They are characterized by overlapping clinicopathologic and genetic features, and the most often diagnosed subtype in both children and adults thereby being angioimmunoblastic T-cell lymphoma (AITL), described in the following section.

Morphology and Immunohistology (Fig. 7.9)

These systemic lymphomas are composed of polymorphic mature T cells with a characteristic expression of at least two to three markers characteristic for follicular helper T cells, such as CD10, BCL-6, PD1, CXCL13, ICOS, and CXCR5. Lymph nodes affected show an effaced architecture with a characteristic sparing of the cortical sinuses, and often preserved capsules with infiltration of the perinodal fatty tissue [1]. In the paracortical expanded areas, proliferations of branching vessels with high endothelial cells (HEV) and vast proliferations of follicular dendritic cell meshworks are a characteristic feature. The lesional T cells are usually small to medium sized and only slightly atypical, and typically present with pale or clear cytoplasms, frequently in the vicinity of arborizing HEV. The T cells are arranged within a usually abundant inflammatory background, containing numerous histiocytes, plasma cells and transformed B-blasts, sometimes with Reed-Sternberg-like features, which show evidence of EBV infection in most cases. Three patterns of infiltration are recognized, with pattern 1 showing only subtle changes: in this pattern, neoplastic T-cell surround regular-appearing germinal centers; in pattern 2, regressive remnants of B-cell follicles are seen surrounded by numerous T cells, which are more easily detected by morphology than in pattern 1; pattern 3 shows a totally effaced lymph node architecture with only few regressed B-cell follicles in the cortical areas [156]. Immunophenotypically, a mature CD4+ T-cell phenotype can be discerned, with characteristic expression of several FTH markers listed above, and occasional losses of T-cell antigens. The B cells and plasma cells are usually polyclonal with regard to Ig light-chain expression. The FDRC meshworks are positive with CD21 and CD23. Genetically, these lymphomas usually show clonally rearranged TCR chain genes, and a substantial number (25–30%) also show clonally rearranged Ig-chain genes [157], especially in cases with the

presence of a substantial EBV+ population. Using cytogenetics, clonal aberrations (such as trisomies 3, 5, and 21 as well as loss of X or 6q) are found in a vast majority of cases [158]. Typical gene mutations found in AITL are mutations in *IDH2*, *TET2*, *DNMT3A*, and *RHOA* [159].

Post-transplant Lymphoproliferative Disorders

Post-transplant lymphoproliferative disorders (PTLDs) arise in recipients of solid organ or hematopoietic stem cell transplantation (HSCT) in the setting of immunosuppression. PTLDs encompass a spectrum of lymphoid and plasmacytoid proliferations, ranging from early, usually polyclonal lesions to fully developed monoclonal lymphomas. Most of the proliferations are associated with EBV, although a portion (20%) of lymphomas, especially in adults, lack EBV association and are indistinguishable from lymphomas arising in the immunocompetent host. The EBV cases tend to occur later after transplantation (TX) than EBV+ cases. Almost all PTLDs are of B-cell origin with only very few T-cell lymphoproliferations and few cases of classical Hodgkin's lymphomas. T-cell-derived PTLDs are mostly EBV-negative.

Among pediatric recipients, the incidence of PTLD is higher than in adults [160] and is strongly associated with post-transplant primary EBV infection [161].

The highest rates of PTLD have been reported in patients receiving intestinal and lung as well as heart/lung allografts with an incidence up to 16% after 5 years post TX [162], whereas renal allograft recipients are at lower risk for PTLD (up to 2.4% after 5 years). PTLD after HSCT usually derives from donor B cells and occurs mainly within the first 3–6 months after TX [163]. The incidence is with <2% usually lower than in patients receiving solid organ TX, but may increase with risk factors such as T-cell-depleted grafts, degree of donor-recipient mismatch, or extent of immunosuppression [164]. PTLD may involve lymph nodes, but also extranodal sites, and frequently affects the site of the allograft. Affection of the allograft is ordinarily associated with EBV, occurs early after TX and is most commonly observed in lung and intestinal transplant recipients [165, 166].

Morphology and Immunohistology (Fig. 7.10)

PTLDs comprise a large, somehow blurred spectrum of various lymphoproliferative disorders. The spectrum ranges from the early, nondestructive PTLDs through polymorphic PTLDs to the clinically destructive and most aggressive forms of monomorphic PTLDs (Table 7.5).

The nondestructive forms are defined as lymphoid proliferations with preservation of the underlying architecture of the organ involved, forming mass lesions in most instances. Within this category, basically three disorders are subsummarized: Plasmacytic hyperplasia (PH), florid follicular hyperplasia (FFH), and infectious mononucleosis-like (IM) PTLDs. These PTLDs correspond to the formerly so-called early lesions, a term which should no longer be used due to potential confusion with lesions arising early after TX [167].

The nondestructive lesions more often occur in children and young adults without prior EBV infection. Usually they affect lymph nodes or lymphoid tissue of the Waldeyer ring such as tonsils and adenoids [168].

PH is the most common type of the nondestructive lesions and shows numerous mature plasma cells in the context with lymphocytes and occasional immunoblasts located in the expanded interfollicular regions. The uninvolved, preserved areas show either inconspicuous lymphoid follicles or reactive expanded follicles, especially in affected tonsils or adenoids [167]. FFH is characterized by large germinal centers with signs of polarization, composed of numerous centroblasts, starry-sky macrophages, and only few centrocytes [169]. Mitotic figures are abundant, and mantle zones are regularly unaffected. These cases lack a significant plasmacellular proliferation.

IM-like lesions show the typical morphology of IM, with marked paracortical expansion with a usually diffuse proliferation of a heterogeneous population of immunoblasts, scattered in a background of numerous polymorphic lymphocytes. Reactive follicles are infrequently detected [167].

The B cells and plasma cells in the nondestructive PTLDs exhibit polytypic expression of kappa and lambda light chains. Both B cells and T cells display a fully developed immunoprofile without antigen loss.

As a prerequisite for the categorization into this form of PTLD, EBV positivity has to be demonstrated in a substantial quantity of lesional cells. The demonstration of EBV in the tissue is best accomplished with the EBER in situ hybridization. Typically, IM-like PTLD shows positivity for LMP-1 in the proliferating immunoblasts with frequent coexpression of CD30 [170].

Genetic Profile of Nondestructive PTLDs

Usually the lesions in the nondestructive forms of PTLD show a polyclonal pattern of both IgH and IgL chain genes. However, cases showing small oligoclonal and even monoclonal rearrangements with unknown significances have been reported in the past.

The polymorphic PTLDs (P-PTLD) are characterized by a polymorphous proliferation of a spectrum of lymphoid cells of variable cell size and differentiation [171]. Usually, small

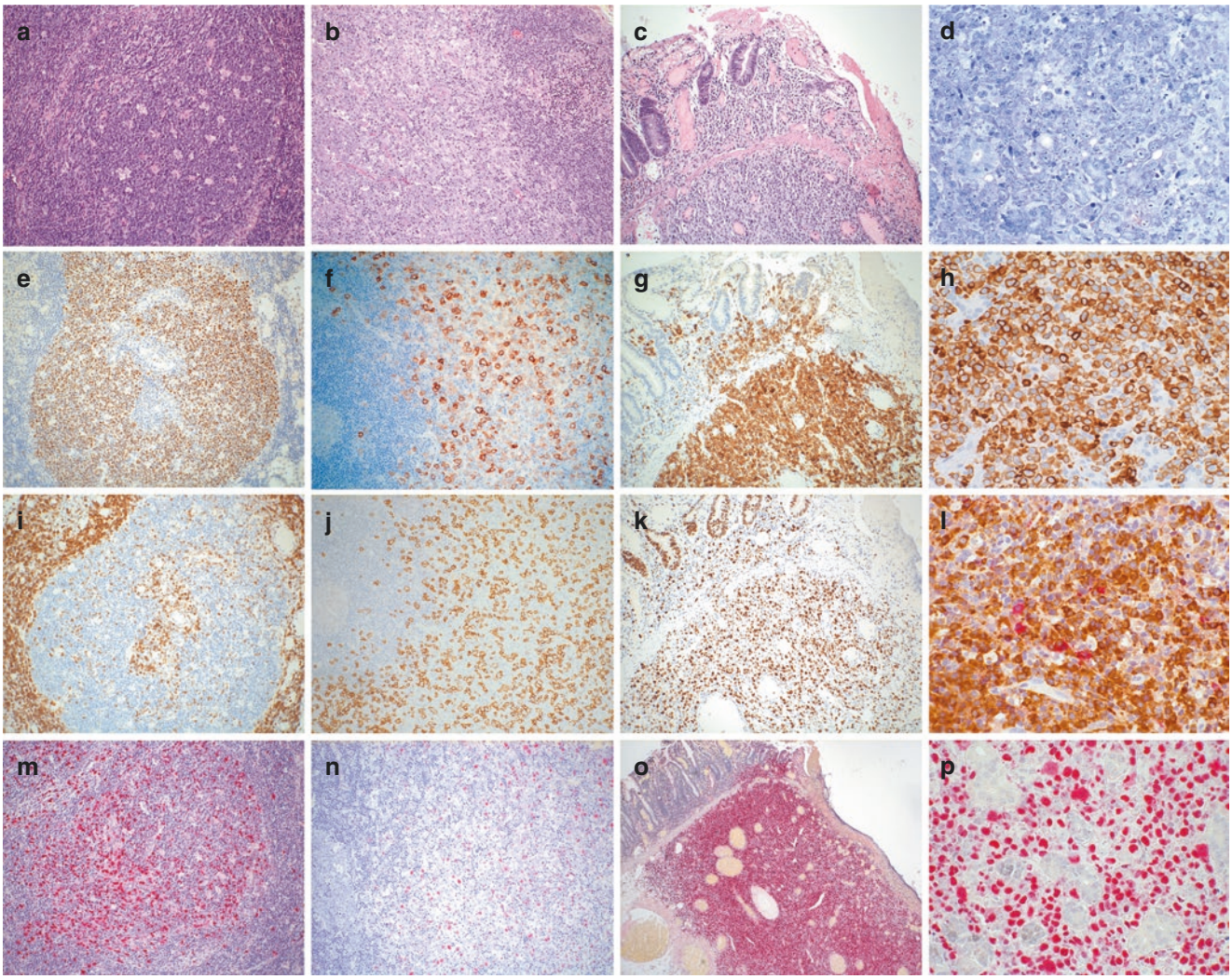

Fig. 7.10 Post-transplant lymphoproliferative disorders (PTLDs). Both left rows (**a**, **e**, **i**, **m** and **b**, **f**, **j**, **n**, respectively) show nondestructive representatives of PTLD with florid follicular hyperplasia (FFH) in the very left row; germinal center cells are bcl-6+ (**e**), bcl-2-negative (**i**), and numerous cells within the GC are EBV-positive (EBER-ISH) (**m**). The next row (**b**, **j**, **f**, **n**) denotes infectious-mononucleosis-like features, with marked interfollicular expansion, containing a diffuse polymorphous proliferation of immunoblasts and plasmacytoid cells, many with co-expression of CD30 (**f**); numerous CD8+ T cells are intermingled (**j**); the proliferating B cells are EBV-infected (**n**). The right panels (**c**, **g**, **k**, **o** and **d**, **h**, **l**, **p**, respectively) show examples of destructive lesions, with a polymorphic PTLD presenting with an ulcerative mass lesion in the small intestine in (**c**); the polymorphic tumor cells infiltrate the bowel wall; cells are positive for CD79a (**g**), and show a high proliferation fraction (Ki-67) (**k**), and are EBV+ (**o**); the far right panel shows a monomorphous DLBCL-type PTLD with numerous IB and plasmablasts (**d**), with expression of CD79a (**h**), and monotypic expression of the Ig-light-chain κ (**l**) (Double staining with κ in brown, and λ in magenta); EBV-positivity demonstrated in (**p**)

and intermediate lymphoid cells are intermixed with immunoblasts and plasmacytoid cells as well as mature plasma cells. In contrast to the first category of the nondestructive lesions, the P-PTLDs form mass lesions and show alterations or destruction of the underlying tissue architecture. A large proportion of reactive T cells is frequently detected. Some lesions are found to be associated with areas of geographical necrosis. Typically, large pleomorphic cells are intermingled, showing marked similarities to classic Hodgkin and Reed-Sternberg cells. The distinction of cases with areas showing more monotonous appearing infiltrates to cases of the monomorphic PTLD (M-PTLD) category may be challenging, as there are no clear-cut, well-defined criteria so far. As a basic rule, lesions that would unequivocally be diagnosed as overt lymphoma in a non-transplant recipient should best be termed M-PTLD in the setting associated post TX. Some cases with numerous RS-like cells may be difficult to discern from true cHL PTLD. These lesions have been termed "Hodgkin-like PTLD" in the past [172].

Immunohistology demonstrates numerous reactive CD3+ T cells intermixed with a variable number of the actual lesional CD20+ B cells. Ig light-chain restriction of plasma-

Table 7.5 PTLD classification WHO 2016 [1] (slightly modified)

PTLD	Type	Tissue Effacement	Histology	Immunohistology	IgH/TCR clonality
Nondestructive		*No*			*Polyclonal*
	Plasmacytic hyperplasia		Pronounced interfollicular plasma cell proliferation	Polytypic κ and λ, EBV+	
	Florid follicular hyperplasia		Numerous large hyperplastic GC	Polytypic B cells, plasma cells, EBV+	
	Infectious mononucleosis-like		Diffuse interfollicular polymorphous proliferation, numerous IB, plasmablasts	Polytypic B cells, plasma cells, CD30+ IB, EBV+	
Destructive		*Yes*			*Monoclonal*
	Polymorphic		Spectrum of lymphoid differentiation	Monotypic κ and λ, EBV+	
	Monomorphic		Mostly DLBCL or plasma cell neoplasia	Clonal B cells or T cells	
	Classical Hodgkin's lymphoma	Yes	Same as cHL	EBV+/−	

Abbreviations: *GC* germinal center, *IB* immunoblast, *DLBCL* diffuse large B-cell lymphoma, *cHL* classical Hodgkin's lymphoma

cytoid cells and plasma cells may be focally detected. The large bizarre Hodgkin-and RS-like cells usually demonstrate pronounced expression of CD30, show frequent co-expression of CD79a and CD20, and usually lack CD15 expression. The EBV association is established with EBER-ISH. In contrast to "true" cHL PTLD, the "Hodgkin-like" cases exhibit EBV positivity additionally in a significant portion of small- and intermediate-sized lymphoid cells [173].

Genetic profile of P-PTLD: Most cases of P-PTLD show clonally rearranged Ig heavy and light-chain genes, whereas T-cell receptor genes are usually not clonally rearranged.

The M-PTLDs are usually of B-cell origin, although some T/NK lymphomas have been reported in the setting of SOT. The monomorphic lymphomas are the most common types of PTLD, constituting 60–80% in most reported large series [174]. These lymphomas share basically an identical morphology and immunophenotypic profile as lymphomas arising in immunocompetent hosts. Most lymphomas fulfill the morphologic and immunohistologic criteria for a diagnosis of DLBCL or to a lesser frequency Burkitt lymphoma or a plasma cell proliferation.

Of note, the lesions corresponding to DLBCL frequently show a rather polymorphous infiltrate of large transformed tumor cells with occasional RS-like cells, and necrosis is sometimes a prominent finding. The plasmacytic proliferations are composed of sheets of plasma cells with few intermingled lymphoid cells.

An indolent peculiar-specific subtype, namely EBV+ extranodal marginal-zone B-cell lymphoma of mucosa-associated lymphoid tissue (MALT-lymphoma), has been added into the category of M-PTLD [175]; these lesions present with cutaneous or subcutaneous localizations and must be distinguished from other extranodal, frequently gastric or pulmonary EBV-negative MALT lymphomas and other EBV-negative small B-cell lymphoma proliferations, which are not included in the PTLD categories. These EBV+ MALT lymphomas have an easily discernable plasmacytic differentiation with intermixture of lymphoid, marginal-zone like cells with abundant pale cytoplasms.

Immunohistologically, the B-cell lymphoproliferations show a mature B-cell immunophenotype with expression of PAX-5, CD19, CD20, and CD79a, with frequently demonstrable monotypic Ig light-chain expression. CD30 is often expressed by a large number of tumor cells, especially in cases with EBV association. The EBV+ cases exhibit more often a non-GCB immunophenotype (CD10-/BCL-6-/MUM-1+), whereas the EBV-negative DLBCLs are more often of the GCB immunophenotype with expression of CD10 and BCL-6, without co-expression of MUM-1. BL occurring after TX is usually EBV+ and shows an identical immunoprofile as the classical forms.

The plasmacytoma-like proliferations may be EBV-positive or EBV-negative, are CD38+/CD138+, usually lack CD20 and CD19, and show monotypic expression of both Ig heavy and light chains. EBV association has to be demonstrated in case of MALT-type PTLD. Besides expression of B-cell markers, these lymphomas were reported with frequent expression of IgA [175].

Genetically, the M-PTLDs show clonal rearrangements of IgH and IgL chain genes in almost all cases. Of note, a substantial number of clearly B-cell PTLDs were reported to harbor monoclonal T-cell receptor gene rearrangements, especially in cases with numerous reactive CD8+ T cells in the background [1].

The monomorphic T/NK PTLDs are rare, constituting only 5–15% of all PTLDs [176]. They fulfill the criteria similarly to lymphomas arising in non-immunocompromised patients both morphologically and immunophenotypically. In most instances PTCL, NOS, or hepatosplenic T-cell lymphomas are seen [177]. Other forms include T-large granular lymphocytic leukemia, SPTCL, extranodal NK/T-cell lymphoma, nasal type, ALCL (either systemic or cutaneous), and even mycosis

fungoides/Sézary's syndrome [178]. Immunohistologically, various T-cell and/or NK-cell markers are expressed, such as CD2, CD3, CD5, CD7, and CD4 or CD8. Cases of HSTCL typically show loss of T-cell markers, such as CD5, and are T-cell receptor gamma delta-positive, with frequent co-expression of CD56 and CD4/CD8 double-negativity [179]. About 30% of the T/NK PTLD are EBV-positive [180].

Genetically, T -PTLDs Show Clonal Rearrangements of the T-Cell Receptor Chain Genes.

References

1. Swerdlow S, Campo E, Harris NL, Jaffe E, Pileri S, Stein H, et al. WHO classification of tumours of haematopoietic and lymphoid tissues. Lyon: IARC Press; 2017.
2. Sandlund JT, Downing JR, Crist WM. Non-Hodgkin's lymphoma in childhood. N Engl J Med. 1996;334(19):1238–48.
3. Thomas DA, Kantarjian HM. Lymphoblastic lymphoma. Hematol Oncol Clin North Am. 2001;15(1):51–95, vi.
4. Lin P, Jones D, Dorfman DM, Medeiros LJ. Precursor B-cell lymphoblastic lymphoma: a predominantly extranodal tumor with low propensity for leukemic involvement. Am J Surg Pathol. 2000;24(11):1480–90.
5. Tricot G, Broeckaert-Van Orshoven A, Van Hoof A, Verwilghen RL. Sudan black B positivity in acute lymphoblastic leukaemia. Br J Haematol. 1982;51(4):615–21.
6. Perkins SL, Segal GH, Kjeldsberg CR. Classification of non-Hodgkin's lymphomas in children. Semin Diagn Pathol. 1995;12(4):303–13.
7. Oschlies I, Burkhardt B, Chassagne-Clement C, d'Amore ES, Hansson U, Hebeda K, et al. Diagnosis and immunophenotype of 188 pediatric lymphoblastic lymphomas treated within a randomized prospective trial: experiences and preliminary recommendations from the European childhood lymphoma pathology panel. Am J Surg Pathol. 2011;35(6):836–44.
8. Arber DA, Snyder DS, Fine M, Dagis A, Niland J, Slovak ML. Myeloperoxidase immunoreactivity in adult acute lymphoblastic leukemia. Am J Clin Pathol. 2001;116(1):25–33.
9. Greaves MF, Janossy G, Peto J, Kay H. Immunologically defined subclasses of acute lymphoblastic leukaemia in children: their relationship to presentation features and prognosis. Br J Haematol. 1981;48(2):179–97.
10. Bene MC, Castoldi G, Knapp W, Ludwig WD, Matutes E, Orfao A, et al. Proposals for the immunological classification of acute leukemias. European Group for the Immunological Characterization of Leukemias (EGIL). Leukemia. 1995;9(10):1783–6.
11. Chiaretti S, Zini G, Bassan R. Diagnosis and subclassification of acute lymphoblastic leukemia. Mediterr J Hematol Infect Dis. 2014;6(1):e2014073.
12. Attygalle A, Al-Jehani R, Diss TC, Munson P, Liu H, Du MQ, et al. Neoplastic T cells in angioimmunoblastic T-cell lymphoma express CD10. Blood. 2002;99(2):627–33.
13. Pilozzi E, Pulford K, Jones M, Muller-Hermelink HK, Falini B, Ralfkiaer E, et al. Co-expression of CD79a (JCB117) and CD3 by lymphoblastic lymphoma. J Pathol. 1998;186(2):140–3.
14. Uckun FM, Sather HN, Gaynon PS, Arthur DC, Trigg ME, Tubergen DG, et al. Clinical features and treatment outcome of children with myeloid antigen positive acute lymphoblastic leukemia: a report from the Children's Cancer Group. Blood. 1997;90(1):28–35.
15. Coustan-Smith E, Mullighan CG, Onciu M, Behm FG, Raimondi SC, Pei D, et al. Early T-cell precursor leukaemia: a subtype of very high-risk acute lymphoblastic leukaemia. Lancet Oncol. 2009;10(2):147–56.
16. Chantepie SP, Cornet E, Salaun V, Reman O. Hematogones: an overview. Leuk Res. 2013;37(11):1404–11.
17. McKenna RW, Washington LT, Aquino DB, Picker LJ, Kroft SH. Immunophenotypic analysis of hematogones (B-lymphocyte precursors) in 662 consecutive bone marrow specimens by 4-color flow cytometry. Blood. 2001;98(8):2498–507.
18. Al-Shieban S, Byrne E, Trivedi P, Morilla R, Matutes E, Naresh KN. Immunohistochemical distinction of haematogones from B lymphoblastic leukaemia/lymphoma or B-cell acute lymphoblastic leukaemia (B-ALL) on bone marrow trephine biopsies: a study on 62 patients. Br J Haematol. 2011;154(4):466–70.
19. Yasmeen S, Rajkumar A, Grossman H, Szallasi A. Terminal deoxynucleotidyl transferase (TdT)-negative lymphoblastic leukemia in pediatric patients: incidence and clinical significance. Pediatr Dev Pathol. 2017;20:463–8.
20. Adam P, Hakroush S, Hofmann I, Reidenbach S, Marx A, Strobel P. Thymoma with loss of keratin expression (and giant cells): a potential diagnostic pitfall. Virchows Arch. 2014;465(3):313–20.
21. Ohgami RS, Zhao S, Ohgami JK, Leavitt MO, Zehnder JL, West RB, et al. TdT+ T-lymphoblastic populations are increased in Castleman disease, in Castleman disease in association with follicular dendritic cell tumors, and in angioimmunoblastic T-cell lymphoma. Am J Surg Pathol. 2012;36(11):1619–28.
22. Ohgami RS, Sendamarai AK, Atwater SK, Liedtke M, Fleming MD, Natkunam Y, et al. Indolent T-lymphoblastic proliferation with disseminated multinodal involvement and partial CD33 expression. Am J Surg Pathol. 2014;38(9):1298–304.
23. Ohgami RS, Arber DA, Zehnder JL, Natkunam Y, Warnke RA. Indolent T-lymphoblastic proliferation (iT-LBP): a review of clinical and pathologic features and distinction from malignant T-lymphoblastic lymphoma. Adv Anat Pathol. 2013;20(3):137–40.
24. van der Velden VH, Bruggemann M, Hoogeveen PG, de Bie M, Hart PG, Raff T, et al. TCRB gene rearrangements in childhood and adult precursor-B-ALL: frequency, applicability as MRD-PCR target, and stability between diagnosis and relapse. Leukemia. 2004;18(12):1971–80.
25. Szczepanski T, Pongers-Willemse MJ, Langerak AW, Harts WA, Wijkhuijs AJ, van Wering ER, et al. Ig heavy chain gene rearrangements in T-cell acute lymphoblastic leukemia exhibit predominant DH6-19 and DH7-27 gene usage, can result in complete V-D-J rearrangements, and are rare in T-cell receptor alpha beta lineage. Blood. 1999;93(12):4079–85.
26. Mullighan CG. Molecular genetics of B-precursor acute lymphoblastic leukemia. J Clin Invest. 2012;122(10):3407–15.
27. Miles RR, Shah RK, Frazer JK. Molecular genetics of childhood, adolescent and young adult non-Hodgkin lymphoma. Br J Haematol. 2016;173(4):582–96.
28. Graux C, Cools J, Michaux L, Vandenberghe P, Hagemeijer A. Cytogenetics and molecular genetics of T-cell acute lymphoblastic leukemia: from thymocyte to lymphoblast. Leukemia. 2006;20(9):1496–510.
29. Magrath I. Epidemiology: clues to the pathogenesis of Burkitt lymphoma. Br J Haematol. 2012;156(6):744–56.
30. Barth TF, Muller S, Pawlita M, Siebert R, Rother JU, Mechtersheimer G, et al. Homogeneous immunophenotype and paucity of secondary genomic aberrations are distinctive features of endemic but not of sporadic Burkitt's lymphoma and diffuse large B-cell lymphoma with MYC rearrangement. J Pathol. 2004;203(4):940–5.

31. Magrath I. The pathogenesis of Burkitt's lymphoma. Adv Cancer Res. 1990;55:133–270.
32. Hochberg J, Waxman IM, Kelly KM, Morris E, Cairo MS. Adolescent non-Hodgkin lymphoma and Hodgkin lymphoma: state of the science. Br J Haematol. 2009;144(1):24–40.
33. Chuang SS, Huang WT, Hsieh PP, Jung YC, Ye H, Du MQ, et al. Sporadic paediatric and adult Burkitt lymphomas share similar phenotypic and genotypic features. Histopathology. 2008;52(4):427–35.
34. Cairo MS, Sposto R, Perkins SL, Meadows AT, Hoover-Regan ML, Anderson JR, et al. Burkitt's and Burkitt-like lymphoma in children and adolescents: a review of the Children's Cancer Group experience. Br J Haematol. 2003;120(4):660–70.
35. Burkitt D. A sarcoma involving the jaws in African children. Br J Surg. 1958;46(197):218–23.
36. Hecht JL, Aster JC. Molecular biology of Burkitt's lymphoma. J Clin Oncol. 2000;18(21):3707–21.
37. Woo KB, Funkhouser WK, Sullivan C, Alabaster O. Analysis of the proliferation kinetics of Burkitt's lymphoma cells. Cell Tissue Kinet. 1980;13(6):591–604.
38. Perkins SL. Work-up and diagnosis of pediatric non-Hodgkin's lymphomas. Pediatr Dev Pathol. 2000;3(4):374–90.
39. Hummel M, Bentink S, Berger H, Klapper W, Wessendorf S, Barth TF, et al. A biologic definition of Burkitt's lymphoma from transcriptional and genomic profiling. N Engl J Med. 2006;354(23):2419–30.
40. Lones MA, Auperin A, Raphael M, McCarthy K, Perkins SL, MacLennan KA, et al. Mature B-cell lymphoma/leukemia in children and adolescents: intergroup pathologist consensus with the revised European-American Lymphoma Classification. Ann Oncol. 2000;11(1):47–51.
41. Dave SS, Fu K, Wright GW, Lam LT, Kluin P, Boerma EJ, et al. Molecular diagnosis of Burkitt's lymphoma. N Engl J Med. 2006;354(23):2431–42.
42. Rodig SJ, Vergilio JA, Shahsafaei A, Dorfman DM. Characteristic expression patterns of TCL1, CD38, and CD44 identify aggressive lymphomas harboring a MYC translocation. Am J Surg Pathol. 2008;32(1):113–22.
43. Cogliatti SB, Novak U, Henz S, Schmid U, Moller P, Barth TF, et al. Diagnosis of Burkitt lymphoma in due time: a practical approach. Br J Haematol. 2006;134(3):294–301.
44. Naresh KN, Ibrahim HA, Lazzi S, Rince P, Onorati M, Ambrosio MR, et al. Diagnosis of Burkitt lymphoma using an algorithmic approach--applicable in both resource-poor and resource-rich countries. Br J Haematol. 2011;154(6):770–6.
45. Tapia G, Lopez R, Munoz-Marmol AM, Mate JL, Sanz C, Marginet R, et al. Immunohistochemical detection of MYC protein correlates with MYC gene status in aggressive B cell lymphomas. Histopathology. 2011;59(4):672–8.
46. Masque-Soler N, Szczepanowski M, Kohler CW, Aukema SM, Nagel I, Richter J, et al. Clinical and pathological features of Burkitt lymphoma showing expression of BCL2--an analysis including gene expression in formalin-fixed paraffin-embedded tissue. Br J Haematol. 2015;171(4):501–8.
47. Pelicci PG, Knowles DM 2nd, Magrath I, Dalla-Favera R. Chromosomal breakpoints and structural alterations of the c-myc locus differ in endemic and sporadic forms of Burkitt lymphoma. Proc Natl Acad Sci U S A. 1986;83(9):2984–8.
48. Onciu M, Schlette E, Zhou Y, Raimondi SC, Giles FJ, Kantarjian HM, et al. Secondary chromosomal abnormalities predict outcome in pediatric and adult high-stage Burkitt lymphoma. Cancer. 2006;107(5):1084–92.
49. Salaverria I, Martin-Guerrero I, Wagener R, Kreuz M, Kohler CW, Richter J, et al. A recurrent 11q aberration pattern characterizes a subset of MYC-negative high-grade B-cell lymphomas resembling Burkitt lymphoma. Blood. 2014;123(8):1187–98.
50. Rimsza L, Pittaluga S, Dirnhofer S, Copie-Bergman C, de Leval L, Facchetti F, et al. The clinicopathologic spectrum of mature aggressive B cell lymphomas. Virchows Arch. 2017;471(4):453–66.
51. Rymkiewicz G, Grygalewicz B, Chechlinska M, Blachnio K, Bystydzienski Z, Romejko-Jarosinska J, et al. A comprehensive flow-cytometry-based immunophenotypic characterization of Burkitt-like lymphoma with 11q aberration. Mod Pathol. 2018;31(5):732–43.
52. Frost M, Newell J, Lones MA, Tripp SR, Cairo MS, Perkins SL. Comparative immunohistochemical analysis of pediatric Burkitt lymphoma and diffuse large B-cell lymphoma. Am J Clin Pathol. 2004;121(3):384–92.
53. Cairo MS, Sposto R, Hoover-Regan M, Meadows AT, Anderson JR, Siegel SE, et al. Childhood and adolescent large-cell lymphoma (LCL): a review of the Children's Cancer Group experience. Am J Hematol. 2003;72(1):53–63.
54. Preciado MV, Fallo A, Chabay P, Calcagno L, De Matteo E. Epstein Barr virus-associated lymphoma in HIV-infected children. Pathol Res Pract. 2002;198(5):327–32.
55. Furutani E, Shimamura A. Germline genetic predisposition to hematologic malignancy. J Clin Oncol. 2017;35(9):1018–28.
56. Engelhard M, Brittinger G, Huhn D, Gerhartz HH, Meusers P, Siegert W, et al. Subclassification of diffuse large B-cell lymphomas according to the Kiel classification: distinction of centroblastic and immunoblastic lymphomas is a significant prognostic risk factor. Blood. 1997;89(7):2291–7.
57. Dargent JL, Meiers I, Lespagnard L, Ma Y, Dehou MF, Verhest A. Diffuse large B-cell lymphoma with fibrillary matrix. Diagn Cytopathol. 2002;27(4):223–6.
58. Rosenwald A, Wright G, Chan WC, Connors JM, Campo E, Fisher RI, et al. The use of molecular profiling to predict survival after chemotherapy for diffuse large-B-cell lymphoma. N Engl J Med. 2002;346(25):1937–47.
59. Hans CP, Weisenburger DD, Greiner TC, Gascoyne RD, Delabie J, Ott G, et al. Confirmation of the molecular classification of diffuse large B-cell lymphoma by immunohistochemistry using a tissue microarray. Blood. 2004;103(1):275–82.
60. Choi WW, Weisenburger DD, Greiner TC, Piris MA, Banham AH, Delabie J, et al. A new immunostain algorithm classifies diffuse large B-cell lymphoma into molecular subtypes with high accuracy. Clin Cancer Res. 2009;15(17):5494–502.
61. Visco C, Li Y, Xu-Monette ZY, Miranda RN, Green TM, Li Y, et al. Comprehensive gene expression profiling and immunohistochemical studies support application of immunophenotypic algorithm for molecular subtype classification in diffuse large B-cell lymphoma: a report from the International DLBCL Rituximab-CHOP Consortium Program Study. Leukemia. 2012;26(9):2103–13.
62. Miles RR, Raphael M, McCarthy K, Wotherspoon A, Lones MA, Terrier-Lacombe MJ, et al. Pediatric diffuse large B-cell lymphoma demonstrates a high proliferation index, frequent c-Myc protein expression, and a high incidence of germinal center subtype: report of the French-American-British (FAB) international study group. Pediatr Blood Cancer. 2008;51(3):369–74.
63. Sehn LH, Gascoyne RD. Diffuse large B-cell lymphoma: optimizing outcome in the context of clinical and biologic heterogeneity. Blood. 2015;125(1):22–32.
64. Green TM, Young KH, Visco C, Xu-Monette ZY, Orazi A, Go RS, et al. Immunohistochemical double-hit score is a strong predictor of outcome in patients with diffuse large B-cell lymphoma treated with rituximab plus cyclophosphamide, doxorubicin, vincristine, and prednisone. J Clin Oncol. 2012;30(28):3460–7.
65. Sarkozy C, Traverse-Glehen A, Coiffier B. Double-hit and double-protein-expression lymphomas: aggressive and refractory lymphomas. Lancet Oncol. 2015;16(15):e555–e67.
66. Xu-Monette ZY, Tu M, Jabbar KJ, Cao X, Tzankov A, Visco C, et al. Clinical and biological significance of de novo CD5+

diffuse large B-cell lymphoma in Western countries. Oncotarget. 2015;6(8):5615–33.
67. Hu S, Xu-Monette ZY, Balasubramanyam A, Manyam GC, Visco C, Tzankov A, et al. CD30 expression defines a novel subgroup of diffuse large B-cell lymphoma with favorable prognosis and distinct gene expression signature: a report from the International DLBCL Rituximab-CHOP Consortium Program Study. Blood. 2013;121(14):2715–24.
68. Sebastian E, Alcoceba M, Balanzategui A, Marin L, Montes-Moreno S, Flores T, et al. Molecular characterization of immunoglobulin gene rearrangements in diffuse large B-cell lymphoma: antigen-driven origin and IGHV4-34 as a particular subgroup of the non-GCB subtype. Am J Pathol. 2012;181(5):1879–88.
69. Dobashi A. Molecular pathogenesis of diffuse large B-cell lymphoma. J Clin Exp Hematop. 2016;56(2):71–8.
70. Pasqualucci L, Bereshchenko O, Niu H, Klein U, Basso K, Guglielmino R, et al. Molecular pathogenesis of non-Hodgkin's lymphoma: the role of Bcl-6. Leuk Lymphoma. 2003;44(Suppl 3):S5–12.
71. Visco C, Tzankov A, Xu-Monette ZY, Miranda RN, Tai YC, Li Y, et al. Patients with diffuse large B-cell lymphoma of germinal center origin with BCL2 translocations have poor outcome, irrespective of MYC status: a report from an International DLBCL rituximab-CHOP Consortium Program Study. Haematologica. 2013;98(2):255–63.
72. Huang JZ, Sanger WG, Greiner TC, Staudt LM, Weisenburger DD, Pickering DL, et al. The t(14;18) defines a unique subset of diffuse large B-cell lymphoma with a germinal center B-cell gene expression profile. Blood. 2002;99(7):2285–90.
73. Iqbal J, Greiner TC, Patel K, Dave BJ, Smith L, Ji J, et al. Distinctive patterns of BCL6 molecular alterations and their functional consequences in different subgroups of diffuse large B-cell lymphoma. Leukemia. 2007;21(11):2332–43.
74. Deffenbacher KE, Iqbal J, Sanger W, Shen Y, Lachel C, Liu Z, et al. Molecular distinctions between pediatric and adult mature B-cell non-Hodgkin lymphomas identified through genomic profiling. Blood. 2012;119(16):3757–66.
75. Oschlies I, Klapper W, Zimmermann M, Krams M, Wacker HH, Burkhardt B, et al. Diffuse large B-cell lymphoma in pediatric patients belongs predominantly to the germinal-center type B-cell lymphomas: a clinicopathologic analysis of cases included in the German BFM (Berlin-Frankfurt-Munster) Multicenter Trial. Blood. 2006;107(10):4047–52.
76. Schmitz R, Wright GW, Huang DW, Johnson CA, Phelan JD, Wang JQ, et al. Genetics and pathogenesis of diffuse large B-cell lymphoma. N Engl J Med. 2018;378(15):1396–407.
77. Oschlies I, Burkhardt B, Salaverria I, Rosenwald A, d'Amore ES, Szczepanowski M, et al. Clinical, pathological and genetic features of primary mediastinal large B-cell lymphomas and mediastinal gray zone lymphomas in children. Haematologica. 2011;96(2):262–8.
78. Rosenwald A, Wright G, Leroy K, Yu X, Gaulard P, Gascoyne RD, et al. Molecular diagnosis of primary mediastinal B cell lymphoma identifies a clinically favorable subgroup of diffuse large B cell lymphoma related to Hodgkin lymphoma. J Exp Med. 2003;198(6):851–62.
79. Traverse-Glehen A, Pittaluga S, Gaulard P, Sorbara L, Alonso MA, Raffeld M, et al. Mediastinal gray zone lymphoma: the missing link between classic Hodgkin's lymphoma and mediastinal large B-cell lymphoma. Am J Surg Pathol. 2005;29(11):1411–21.
80. Dunleavy K, Wilson WH. Primary mediastinal B-cell lymphoma and mediastinal gray zone lymphoma: do they require a unique therapeutic approach? Blood. 2015;125(1):33–9.
81. Dunleavy K, Steidl C. Emerging biological insights and novel treatment strategies in primary mediastinal large B-cell lymphoma. Semin Hematol. 2015;52(2):119–25.
82. Savage KJ, Monti S, Kutok JL, Cattoretti G, Neuberg D, De Leval L, et al. The molecular signature of mediastinal large B-cell lymphoma differs from that of other diffuse large B-cell lymphomas and shares features with classical Hodgkin lymphoma. Blood. 2003;102(12):3871–9.
83. Yuan J, Wright G, Rosenwald A, Steidl C, Gascoyne RD, Connors JM, et al. Identification of primary mediastinal large B-cell lymphoma at nonmediastinal sites by gene expression profiling. Am J Surg Pathol. 2015;39(10):1322–30.
84. Moller P, Moldenhauer G, Momburg F, Lammler B, Eberlein-Gonska M, Kiesel S, et al. Mediastinal lymphoma of clear cell type is a tumor corresponding to terminal steps of B cell differentiation. Blood. 1987;69(4):1087–95.
85. Garcia JF, Mollejo M, Fraga M, Forteza J, Muniesa JA, Perez-Guillermo M, et al. Large B-cell lymphoma with Hodgkin's features. Histopathology. 2005;47(1):101–10.
86. Pileri SA, Gaidano G, Zinzani PL, Falini B, Gaulard P, Zucca E, et al. Primary mediastinal B-cell lymphoma: high frequency of BCL-6 mutations and consistent expression of the transcription factors OCT-2, BOB.1, and PU.1 in the absence of immunoglobulins. Am J Pathol. 2003;162(1):243–53.
87. Calaminici M, Piper K, Lee AM, Norton AJ. CD23 expression in mediastinal large B-cell lymphomas. Histopathology. 2004;45(6):619–24.
88. Copie-Bergman C, Gaulard P, Maouche-Chretien L, Briere J, Haioun C, Alonso MA, et al. The MAL gene is expressed in primary mediastinal large B-cell lymphoma. Blood. 1999;94(10):3567–75.
89. Gentry M, Bodo J, Durkin L, Hsi ED. Performance of a commercially available MAL antibody in the diagnosis of primary mediastinal large B-cell lymphoma. Am J Surg Pathol. 2017;41(2):189–94.
90. Johnson PW, Davies AJ. Primary mediastinal B-cell lymphoma. Hematology Am Soc Hematol Educ Program. 2008;2008:349–58.
91. Grant C, Dunleavy K, Eberle FC, Pittaluga S, Wilson WH, Jaffe ES. Primary mediastinal large B-cell lymphoma, classic Hodgkin lymphoma presenting in the mediastinum, and mediastinal gray zone lymphoma: what is the oncologist to do? Curr Hematol Malig Rep. 2011;6(3):157–63.
92. Weniger MA, Gesk S, Ehrlich S, Martin-Subero JI, Dyer MJ, Siebert R, et al. Gains of REL in primary mediastinal B-cell lymphoma coincide with nuclear accumulation of REL protein. Genes Chromosomes Cancer. 2007;46(4):406–15.
93. Eberle FC, Salaverria I, Steidl C, Summers TA Jr, Pittaluga S, Neriah SB, et al. Gray zone lymphoma: chromosomal aberrations with immunophenotypic and clinical correlations. Mod Pathol. 2011;24(12):1586–97.
94. Eberle FC, Rodriguez-Canales J, Wei L, Hanson JC, Killian JK, Sun HW, et al. Methylation profiling of mediastinal gray zone lymphoma reveals a distinctive signature with elements shared by classical Hodgkin's lymphoma and primary mediastinal large B-cell lymphoma. Haematologica. 2011;96(4):558–66.
95. Liu Q, Salaverria I, Pittaluga S, Jegalian AG, Xi L, Siebert R, et al. Follicular lymphomas in children and young adults: a comparison of the pediatric variant with usual follicular lymphoma. Am J Surg Pathol. 2013;37(3):333–43.
96. Louissaint A Jr, Ackerman AM, Dias-Santagata D, Ferry JA, Hochberg EP, Huang MS, et al. Pediatric-type nodal follicular lymphoma: an indolent clonal proliferation in children and adults with high proliferation index and no BCL2 rearrangement. Blood. 2012;120(12):2395–404.
97. Agrawal R, Wang J. Pediatric follicular lymphoma: a rare clinicopathologic entity. Arch Pathol Lab Med. 2009;133(1):142–6.
98. Oschlies I, Salaverria I, Mahn F, Meinhardt A, Zimmermann M, Woessmann W, et al. Pediatric follicular lymphoma--a clinicopathological study of a population-based series of patients treated within the Non-Hodgkin's Lymphoma--Berlin-Frankfurt-

Munster (NHL-BFM) multicenter trials. Haematologica. 2010;95(2):253–9.
99. Quintanilla-Martinez L, Sander B, Chan JK, Xerri L, Ott G, Campo E, et al. Indolent lymphomas in the pediatric population: follicular lymphoma, IRF4/MUM1+ lymphoma, nodal marginal zone lymphoma and chronic lymphocytic leukemia. Virchows Arch. 2016;468(2):141–57.
100. Lorsbach RB, Shay-Seymore D, Moore J, Banks PM, Hasserjian RP, Sandlund JT, et al. Clinicopathologic analysis of follicular lymphoma occurring in children. Blood. 2002;99(6):1959–64.
101. Karnik T, Ozawa MG, Lefterova M, Luna-Fineman S, Alvarez E, Link M, et al. The utility of IgM, CD21, HGAL and LMO2 in the diagnosis of pediatric follicular lymphoma. Hum Pathol. 2015;46(4):629–33.
102. Martin-Guerrero I, Salaverria I, Burkhardt B, Szczepanowski M, Baudis M, Bens S, et al. Recurrent loss of heterozygosity in 1p36 associated with TNFRSF14 mutations in IRF4 translocation negative pediatric follicular lymphomas. Haematologica. 2013;98(8):1237–41.
103. Schmidt J, Gong S, Marafioti T, Mankel B, Gonzalez-Farre B, Balague O, et al. Genome-wide analysis of pediatric-type follicular lymphoma reveals low genetic complexity and recurrent alterations of TNFRSF14 gene. Blood. 2016;128(8):1101–11.
104. Louissaint A Jr, Schafernak KT, Geyer JT, Kovach AE, Ghandi M, Gratzinger D, et al. Pediatric-type nodal follicular lymphoma: a biologically distinct lymphoma with frequent MAPK pathway mutations. Blood. 2016;128(8):1093–100.
105. Schmidt J, Ramis-Zaldivar JE, Nadeu F, Gonzalez-Farre B, Navarro A, Egan C, et al. Mutations of MAP2K1 are frequent in pediatric-type follicular lymphoma and result in ERK pathway activation. Blood. 2017;130(3):323–7.
106. Kussick SJ, Kalnoski M, Braziel RM, Wood BL. Prominent clonal B-cell populations identified by flow cytometry in histologically reactive lymphoid proliferations. Am J Clin Pathol. 2004;121(4):464–72.
107. Salaverria I, Philipp C, Oschlies I, Kohler CW, Kreuz M, Szczepanowski M, et al. Translocations activating IRF4 identify a subtype of germinal center-derived B-cell lymphoma affecting predominantly children and young adults. Blood. 2011;118(1):139–47.
108. de Leval L, Bonnet C, Copie-Bergman C, Seidel L, Baia M, Briere J, et al. Diffuse large B-cell lymphoma of Waldeyer's ring has distinct clinicopathologic features: a GELA study. Ann Oncol. 2012;23(12):3143–51.
109. Taddesse-Heath L, Pittaluga S, Sorbara L, Bussey M, Raffeld M, Jaffe ES. Marginal zone B-cell lymphoma in children and young adults. Am J Surg Pathol. 2003;27(4):522–31.
110. Rizzo KA, Streubel B, Pittaluga S, Chott A, Xi L, Raffeld M, et al. Marginal zone lymphomas in children and the young adult population; characterization of genetic aberrations by FISH and RT-PCR. Mod Pathol. 2010;23(6):866–73.
111. Kluin PM, Langerak AW, Beverdam-Vincent J, Geurts-Giele WR, Visser L, Rutgers B, et al. Paediatric nodal marginal zone B-cell lymphadenopathy of the neck: a Haemophilus influenzae-driven immune disorder? J Pathol. 2015;236(3):302–14.
112. Attygalle AD, Liu H, Shirali S, Diss TC, Loddenkemper C, Stein H, et al. Atypical marginal zone hyperplasia of mucosa-associated lymphoid tissue: a reactive condition of childhood showing immunoglobulin lambda light-chain restriction. Blood. 2004;104(10):3343–8.
113. Morris SW, Kirstein MN, Valentine MB, Dittmer KG, Shapiro DN, Saltman DL, et al. Fusion of a kinase gene, ALK, to a nucleolar protein gene, NPM, in non-Hodgkin's lymphoma. Science. 1994;263(5151):1281–4.
114. Pulford K, Lamant L, Morris SW, Butler LH, Wood KM, Stroud D, et al. Detection of anaplastic lymphoma kinase (ALK) and nucleolar protein nucleophosmin (NPM)-ALK proteins in normal and neoplastic cells with the monoclonal antibody ALK1. Blood. 1997;89(4):1394–404.
115. Brugieres L, Deley MC, Pacquement H, Meguerian-Bedoyan Z, Terrier-Lacombe MJ, Robert A, et al. CD30(+) anaplastic large-cell lymphoma in children: analysis of 82 patients enrolled in two consecutive studies of the French Society of Pediatric Oncology. Blood. 1998;92(10):3591–8.
116. Alessandri AJ, Pritchard SL, Schultz KR, Massing BG. A population-based study of pediatric anaplastic large cell lymphoma. Cancer. 2002;94(6):1830–5.
117. Hapgood G, Savage KJ. The biology and management of systemic anaplastic large cell lymphoma. Blood. 2015;126(1):17–25.
118. Falini B, Pileri S, Zinzani PL, Carbone A, Zagonel V, Wolf-Peeters C, et al. ALK+ lymphoma: clinico-pathological findings and outcome. Blood. 1999;93(8):2697–706.
119. Kadin ME. Anaplastic large cell lymphoma and its morphological variants. Cancer Surv. 1997;30:77–86.
120. Benharroch D, Meguerian-Bedoyan Z, Lamant L, Amin C, Brugieres L, Terrier-Lacombe MJ, et al. ALK-positive lymphoma: a single disease with a broad spectrum of morphology. Blood. 1998;91(6):2076–84.
121. Falini B, Bigerna B, Fizzotti M, Pulford K, Pileri SA, Delsol G, et al. ALK expression defines a distinct group of T/null lymphomas ("ALK lymphomas") with a wide morphological spectrum. Am J Pathol. 1998;153(3):875–86.
122. Pileri SA, Pulford K, Mori S, Mason DY, Sabattini E, Roncador G, et al. Frequent expression of the NPM-ALK chimeric fusion protein in anaplastic large-cell lymphoma, lympho-histiocytic type. Am J Pathol. 1997;150(4):1207–11.
123. Kinney MC, Collins RD, Greer JP, Whitlock JA, Sioutos N, Kadin ME. A small-cell-predominant variant of primary Ki-1 (CD30)+ T-cell lymphoma. Am J Surg Pathol. 1993;17(9):859–68.
124. Vassallo J, Lamant L, Brugieres L, Gaillard F, Campo E, Brousset P, et al. ALK-positive anaplastic large cell lymphoma mimicking nodular sclerosis Hodgkin's lymphoma: report of 10 cases. Am J Surg Pathol. 2006;30(2):223–9.
125. Hodges KB, Collins RD, Greer JP, Kadin ME, Kinney MC. Transformation of the small cell variant Ki-1+ lymphoma to anaplastic large cell lymphoma: pathologic and clinical features. Am J Surg Pathol. 1999;23(1):49–58.
126. Stein H, Foss HD, Durkop H, Marafioti T, Delsol G, Pulford K, et al. CD30(+) anaplastic large cell lymphoma: a review of its histopathologic, genetic, and clinical features. Blood. 2000;96(12):3681–95.
127. Penny RJ, Blaustein JC, Longtine JA, Pinkus GS. Ki-1-positive large cell lymphomas, a heterogenous group of neoplasms. Morphologic, immunophenotypic, genotypic, and clinical features of 24 cases. Cancer. 1991;68(2):362–73.
128. Foss HD, Anagnostopoulos I, Araujo I, Assaf C, Demel G, Kummer JA, et al. Anaplastic large-cell lymphomas of T-cell and null-cell phenotype express cytotoxic molecules. Blood. 1996;88(10):4005–11.
129. Brousset P, Rochaix P, Chittal S, Rubie H, Robert A, Delsol G. High incidence of Epstein-Barr virus detection in Hodgkin's disease and absence of detection in anaplastic large-cell lymphoma in children. Histopathology. 1993;23(2):189–91.
130. Duyster J, Bai RY, Morris SW. Translocations involving anaplastic lymphoma kinase (ALK). Oncogene. 2001;20(40):5623–37.
131. Morris SW, Xue L, Ma Z, Kinney MC. Alk+ CD30+ lymphomas: a distinct molecular genetic subtype of non-Hodgkin's lymphoma. Br J Haematol. 2001;113(2):275–95.
132. Pulford K, Lamant L, Espinos E, Jiang Q, Xue L, Turturro F, et al. The emerging normal and disease-related roles of anaplastic lymphoma kinase. Cell Mol Life Sci. 2004;61(23):2939–53.

133. Pulford K, Morris SW, Turturro F. Anaplastic lymphoma kinase proteins in growth control and cancer. J Cell Physiol. 2004;199(3):330–58.
134. Falini B. Anaplastic large cell lymphoma: pathological, molecular and clinical features. Br J Haematol. 2001;114(4):741–60.
135. Oschlies I, Lisfeld J, Lamant L, Nakazawa A, d'Amore ES, Hansson U, et al. ALK-positive anaplastic large cell lymphoma limited to the skin: clinical, histopathological and molecular analysis of 6 pediatric cases. A report from the ALCL99 study. Haematologica. 2013;98(1):50–6.
136. Onaindia A, Montes-Moreno S, Rodriguez-Pinilla SM, Batlle A, Gonzalez de Villambrosia S, Rodriguez AM, et al. Primary cutaneous anaplastic large cell lymphomas with 6p25.3 rearrangement exhibit particular histological features. Histopathology. 2015;66(6):846–55.
137. Parrilla Castellar ER, Jaffe ES, Said JW, Swerdlow SH, Ketterling RP, Knudson RA, et al. ALK-negative anaplastic large cell lymphoma is a genetically heterogeneous disease with widely disparate clinical outcomes. Blood. 2014;124(9):1473–80.
138. Karai LJ, Kadin ME, Hsi ED, Sluzevich JC, Ketterling RP, Knudson RA, et al. Chromosomal rearrangements of 6p25.3 define a new subtype of lymphomatoid papulosis. Am J Surg Pathol. 2013;37(8):1173–81.
139. Onciu M, Behm FG, Downing JR, Shurtleff SA, Raimondi SC, Ma Z, et al. ALK-positive plasmablastic B-cell lymphoma with expression of the NPM-ALK fusion transcript: report of 2 cases. Blood. 2003;102(7):2642–4.
140. Delsol G, Lamant L, Mariame B, Pulford K, Dastugue N, Brousset P, et al. A new subtype of large B-cell lymphoma expressing the ALK kinase and lacking the 2; 5 translocation. Blood. 1997;89(5):1483–90.
141. Chiarle R, Voena C, Ambrogio C, Piva R, Inghirami G. The anaplastic lymphoma kinase in the pathogenesis of cancer. Nat Rev Cancer. 2008;8(1):11–23.
142. Ferreri AJ, Govi S, Pileri SA, Savage KJ. Anaplastic large cell lymphoma, ALK-negative. Crit Rev Oncol Hematol. 2013;85(2):206–15.
143. Pham-Ledard A, Prochazkova-Carlotti M, Laharanne E, Vergier B, Jouary T, Beylot-Barry M, et al. IRF4 gene rearrangements define a subgroup of CD30-positive cutaneous T-cell lymphoma: a study of 54 cases. J Invest Dermatol. 2010;130(3):816–25.
144. Mellgren K, Attarbaschi A, Abla O, Alexander S, Bomken S, Bubanska E, et al. Non-anaplastic peripheral T cell lymphoma in children and adolescents-an international review of 143 cases. Ann Hematol. 2016;95(8):1295–305.
145. Kontny U, Oschlies I, Woessmann W, Burkhardt B, Lisfeld J, Salzburg J, et al. Non-anaplastic peripheral T-cell lymphoma in children and adolescents--a retrospective analysis of the NHL-BFM study group. Br J Haematol. 2015;168(6):835–44.
146. Windsor R, Stiller C, Webb D. Peripheral T-cell lymphoma in childhood: population-based experience in the United Kingdom over 20 years. Pediatr Blood Cancer. 2008;50(4):784–7.
147. Jaffe ES. Pathobiology of peripheral T-cell lymphomas. Hematology Am Soc Hematol Educ Program. 2006;2006:317–22.
148. Dojcinov SD, Fend F, Quintanilla-Martinez L. EBV-positive lymphoproliferations of B- T- and NK-cell derivation in non-immunocompromised hosts. Pathogens. 2018;7(1):28.
149. Quintanilla-Martinez L, Kumar S, Fend F, Reyes E, Teruya-Feldstein J, Kingma DW, et al. Fulminant EBV(+) T-cell lymphoproliferative disorder following acute/chronic EBV infection: a distinct clinicopathologic syndrome. Blood. 2000;96(2):443–51.
150. Belhadj K, Reyes F, Farcet JP, Tilly H, Bastard C, Angonin R, et al. Hepatosplenic gammadelta T-cell lymphoma is a rare clinicopathologic entity with poor outcome: report on a series of 21 patients. Blood. 2003;102(13):4261–9.
151. Huppmann AR, Xi L, Raffeld M, Pittaluga S, Jaffe ES. Subcutaneous panniculitis-like T-cell lymphoma in the pediatric age group: a lymphoma of low malignant potential. Pediatr Blood Cancer. 2013;60(7):1165–70.
152. Oschlies I, Simonitsch-Klupp I, Maldyk J, Konovalov D, Abramov D, Myakova N, et al. Subcutaneous panniculitis-like T-cell lymphoma in children: a detailed clinicopathological description of 11 multifocal cases with a high frequency of haemophagocytic syndrome. Br J Dermatol. 2015;172(3):793–7.
153. Willemze R, Jansen PM, Cerroni L, Berti E, Santucci M, Assaf C, et al. Subcutaneous panniculitis-like T-cell lymphoma: definition, classification, and prognostic factors: an EORTC Cutaneous Lymphoma Group Study of 83 cases. Blood. 2008;111(2):838–45.
154. Pincus LB, LeBoit PE, McCalmont TH, Ricci R, Buzio C, Fox LP, et al. Subcutaneous panniculitis-like T-cell lymphoma with overlapping clinicopathologic features of lupus erythematosus: coexistence of 2 entities? Am J Dermatopathol. 2009;31(6):520–6.
155. Rodriguez-Pinilla SM, Ortiz-Romero PL, Monsalvez V, Tomas IE, Almagro M, Sevilla A, et al. TCR-gamma expression in primary cutaneous T-cell lymphomas. Am J Surg Pathol. 2013;37(3):375–84.
156. Rodriguez-Justo M, Attygalle AD, Munson P, Roncador G, Marafioti T, Piris MA. Angioimmunoblastic T-cell lymphoma with hyperplastic germinal centres: a neoplasia with origin in the outer zone of the germinal centre? Clinicopathological and immunohistochemical study of 10 cases with follicular T-cell markers. Mod Pathol. 2009;22(6):753–61.
157. Attygalle AD, Chuang SS, Diss TC, Du MQ, Isaacson PG, Dogan A. Distinguishing angioimmunoblastic T-cell lymphoma from peripheral T-cell lymphoma, unspecified, using morphology, immunophenotype and molecular genetics. Histopathology. 2007;50(4):498–508.
158. Schlegelberger B, Zhang Y, Weber-Matthiesen K, Grote W. Detection of aberrant clones in nearly all cases of angioimmunoblastic lymphadenopathy with dysproteinemia-type T-cell lymphoma by combined interphase and metaphase cytogenetics. Blood. 1994;84(8):2640–8.
159. Cortes JR, Palomero T. The curious origins of angioimmunoblastic T-cell lymphoma. Curr Opin Hematol. 2016;23(4):434–43.
160. Chinnock R, Webber SA, Dipchand AI, Brown RN, George JF, Pediatric Heart Transplant Study. A 16-year multi-institutional study of the role of age and EBV status on PTLD incidence among pediatric heart transplant recipients. Am J Transplant. 2012;12(11):3061–8.
161. Green M, Michaels MG. Epstein-Barr virus infection and post-transplant lymphoproliferative disorder. Am J Transplant. 2013;13(Suppl 3):41–54; quiz.
162. Webber SA, Naftel DC, Fricker FJ, Olesnevich P, Blume ED, Addonizio L, et al. Lymphoproliferative disorders after paediatric heart transplantation: a multi-institutional study. Lancet. 2006;367(9506):233–9.
163. Rouce RH, Louis CU, Heslop HE. Epstein-Barr virus lymphoproliferative disease after hematopoietic stem cell transplant. Curr Opin Hematol. 2014;21(6):476–81.
164. Uhlin M, Wikell H, Sundin M, Blennow O, Maeurer M, Ringden O, et al. Risk factors for Epstein-Barr virus-related post-transplant lymphoproliferative disease after allogeneic hematopoietic stem cell transplantation. Haematologica. 2014;99(2):346–52.
165. Ramos E, Hernandez F, Andres A, Martinez-Ojinaga E, Molina M, Sarria J, et al. Post-transplant lymphoproliferative disorders and other malignancies after pediatric intestinal transplantation: incidence, clinical features and outcome. Pediatr Transplant. 2013;17(5):472–8.
166. Bakker NA, van Imhoff GW, Verschuuren EA, van Son WJ, van der Heide JJ H, Veeger NJ, et al. Early onset post-transplant

lymphoproliferative disease is associated with allograft localization. Clin Transpl. 2005;19(3):327–34.
167. Nelson BP, Wolniak KL, Evens A, Chenn A, Maddalozzo J, Proytcheva M. Early posttransplant lymphoproliferative disease: clinicopathologic features and correlation with mTOR signaling pathway activation. Am J Clin Pathol. 2012;138(4):568–78.
168. Evens AM, Roy R, Sterrenberg D, Moll MZ, Chadburn A, Gordon LI. Post-transplantation lymphoproliferative disorders: diagnosis, prognosis, and current approaches to therapy. Curr Oncol Rep. 2010;12(6):383–94.
169. Vakiani E, Nandula SV, Subramaniyam S, Keller CE, Alobeid B, Murty VV, et al. Cytogenetic analysis of B-cell posttransplant lymphoproliferations validates the World Health Organization classification and suggests inclusion of florid follicular hyperplasia as a precursor lesion. Hum Pathol. 2007;38(2):315–25.
170. Lones MA, Mishalani S, Shintaku IP, Weiss LM, Nichols WS, Said JW. Changes in tonsils and adenoids in children with posttransplant lymphoproliferative disorder: report of three cases with early involvement of Waldeyer's ring. Hum Pathol. 1995;26(5):525–30.
171. Knowles DM, Cesarman E, Chadburn A, Frizzera G, Chen J, Rose EA, et al. Correlative morphologic and molecular genetic analysis demonstrates three distinct categories of posttransplantation lymphoproliferative disorders. Blood. 1995;85(2):552–65.
172. Pitman SD, Huang Q, Zuppan CW, Rowsell EH, Cao JD, Berdeja JG, et al. Hodgkin lymphoma-like posttransplant lymphoproliferative disorder (HL-like PTLD) simulates monomorphic B-cell PTLD both clinically and pathologically. Am J Surg Pathol. 2006;30(4):470–6.
173. Chetty R, Biddolph S, Gatter K. An immunohistochemical analysis of Reed-Sternberg-like cells in posttransplantation lymphoproliferative disorders: the possible pathogenetic relationship to Reed-Sternberg cells in Hodgkin's disease and Reed-Sternberg-like cells in non-Hodgkin's lymphomas and reactive conditions. Hum Pathol. 1997;28(4):493–8.
174. Al-Mansour Z, Nelson BP, Evens AM. Post-transplant lymphoproliferative disease (PTLD): risk factors, diagnosis, and current treatment strategies. Curr Hematol Malig Rep. 2013;8(3):173–83.
175. Gibson SE, Swerdlow SH, Craig FE, Surti U, Cook JR, Nalesnik MA, et al. EBV-positive extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue in the posttransplant setting: a distinct type of posttransplant lymphoproliferative disorder? Am J Surg Pathol. 2011;35(6):807–15.
176. Wistinghausen B, Gross TG, Bollard C. Post-transplant lymphoproliferative disease in pediatric solid organ transplant recipients. Pediatr Hematol Oncol. 2013;30(6):520–31.
177. Herreman A, Dierickx D, Morscio J, Camps J, Bittoun E, Verhoef G, et al. Clinicopathological characteristics of posttransplant lymphoproliferative disorders of T-cell origin: single-center series of nine cases and meta-analysis of 147 reported cases. Leuk Lymphoma. 2013;54(10):2190–9.
178. Margolskee E, Jobanputra V, Jain P, Chen J, Ganapathi K, Nahum O, et al. Genetic landscape of T- and NK-cell post-transplant lymphoproliferative disorders. Oncotarget. 2016;7(25):37636–48.
179. Swerdlow SH, Jaffe ES, Brousset P, Chan JK, de Leval L, Gaulard P, et al. Cytotoxic T-cell and NK-cell lymphomas: current questions and controversies. Am J Surg Pathol. 2014;38(10):e60–71.
180. Sivaraman P, Lye WC. Epstein-Barr virus-associated T-cell lymphoma in solid organ transplant recipients. Biomed Pharmacother. 2001;55(7):366–8.

Genetic Predisposition to Non-Hodgkin Lymphoma

8

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Introduction

The two main categories of lymphomas are Hodgkin's disease (HD) and non-Hodgkin lymphomas (NHL). Together they not only comprise the most common malignancies in western countries, but, next to leukemias and brain tumors, also the third largest group of neoplasms in children up to 14 years as well as the largest one in teenagers up to 24 years of age [1–3]. In 0–14-year-old children, NHL is slightly more common than Hodgkin lymphoma, whereas the converse is true for teenagers and young, 15–24-year-old adults. Based on their specific biological, (immuno)phenotypic, and genetic features, the recently updated World Health Organization (WHO) classification guidelines distinguish already a large number of different NHL sub-entities [4], although the literature available for this review is still based on a more crude classification that merely comprises B- or T-cell lymphoblastic lymphoma, follicular (FL), diffuse large B-cell (DLBCL), Burkitt (BL), and anaplastic large cell lymphoma (ALCL), a system that hitherto has also formed the essential basis the prognostic classification and, consequently, the allocation to particular forms of treatment.

In children, lymphomas evolve in a tension field, in which a maturing immune system needs to arrange and familiarize itself with its own body's intrinsic components and, at the same time, also to get accustomed to a multitude of environmental exposures, not least various infectious agents [5]. A flawless genetic make-up of all contributing constituents is thus of crucial importance to guarantee the appropriate assembly of the encoded components and their efficient interaction in functional pathways and the required participation in the proper development of the immune system. Equally, dysfunctional or weakened germline components, be it in the form of major single-gene defects or perhaps likewise vital, but less well-recognized genetic modifiers, can easily interfere with the normal physiological development in this particularly vulnerable stage and tilt the balance, among others, also toward neoplastic transformation. Part of these more or less clearly definable genuine heritable preconditions are also normally inert variants in constituents of a well-adapted immune system, which only become relevant under particular circumstances, for instance, the fortuitous exposure to particular environmental hazards. Such either overstimulating or disruptive conditions are chronic infections, primarily those with Epstein-Barr (EBV), human immunodeficiency (HIV) as well as human papillomaviruses (HPV), chronic inflammations, autoimmune diseases, treatments with certain drugs, and organ transplantations (Fig. 8.1) [5–14].

Considering the above, the identification and characterization of predisposing factors has thus rightfully become the focus of special interest especially also in lymphoma research [15]. The recognition and definition of such disease-associated genetic variants is increasingly required for the management and care of patients not least because it often guides the appropriate choice and adaptation of therapy [16–19]. Even when treated successfully, these patients require further surveillance, because they can develop second or secondary neoplasms. The distinction between de novo or inherited disease-relevant germline mutations is a vital prerequisite for assessing the potential consequences for the patient herself as well as her respective family members and, therefore, also for enabling appropriate counseling [13, 20–22]. Last, but not least, the in-depth individual analysis together with the more general screening for such genetic determiners not only satisfies our scientific curiosity. It also broadens our overall knowledge and understanding of the normal physiological function and pathologic consequences of the

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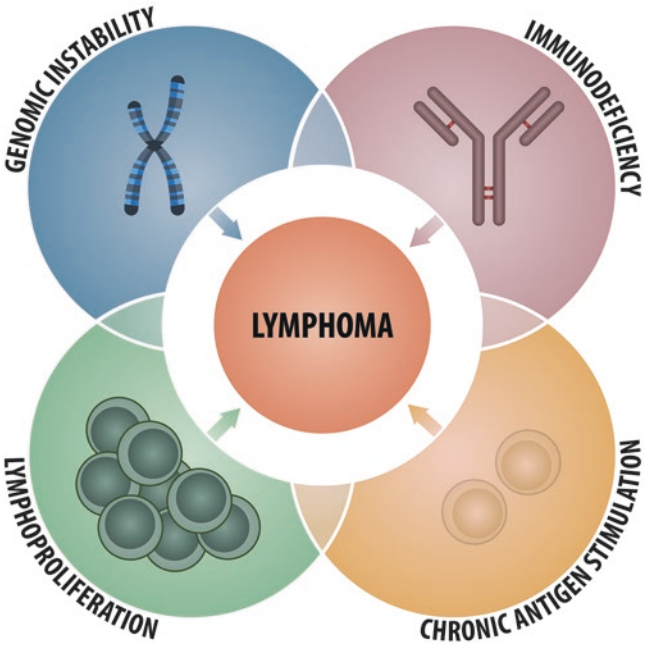

Fig. 8.1 Relevant factors that contribute and participate to lymphoma development in children [5]

respective immune system components and their role in disease mechanisms, which in turn again inevitably enables us to continuously improve and personalize the treatment of the respective patients.

Ascertainment of Genetic Predisposing Factors

There are different tactics one can use to search for and ascertain distinct or more general genetic predisposition factors (Table 8.1) [23]. The special choice of the appropriate mode is primarily a matter of the individual demands and opportunities as well as overall intentions. It can focus on either patient/family-relevant, gene-related, disease-associated or population-based aspects. Whether and when such a predisposing condition is thus suspected and when it becomes apparent depends mainly on the respective screening and verification procedures, which in turn rely on the particular severity and overall consequences of the respective gene defects. In case these generate also obvious physical malformations or other clinical symptoms, such as disturbances of the hematological and/or immune system, they are often known already before lymphoma onset. Conversely, such conditions might only be suspected only once lymphoma is diagnosed. In such scenarios, the careful assessment of medical records and the patient's family history together with his/her physical examination and key laboratory findings will not only help to secure the cause of a preexistent genetic susceptibility but often also provide

Table 8.1 Strategies to ascertain genetic factors that predispose to lymphoma

Based on distinctive or conspicuous clinical features
Ataxia telangiectasia
Nijmegen breakage syndrome
Constitutional mismatch repair syndrome
Primary immunodeficiency syndromes
Other rare DNA repair syndromes
Based on familial predisposition
Twin studies
Familial aggregation
Case-control studies
Cohort studies
Registry-based studies
Based on genetic risk factors
Linkage studies
Genetic association
Candidate genes
Genome-wide association studies (GWAS)
Based on disease
Hodgkin's disease
Non-Hodgkin lymphoma
Diffuse large B-cell lymphoma (DLBCL)
Burkitt lymphoma (BL)
Anaplastic large cell lymphoma (ALCL)

Adapted according to Cerhan and Slager [23]

already those relevant hints, which can ease the identification of the responsible defective gene or at least the category or pathway to which it belongs to [15]. The most relevant indicators comprise dysmorphic features, short stature, various types of cytopenias and immunodeficiencies, specific histopathological lymphoma forms, and/or unproportional treatment toxicities [1, 2, 24–29].

A first global impression about the type and frequency of the various disorders in children and adolescents with NHL can be obtained from information that can be extracted from three large lymphoma trial groups, the "European Intergroup for Childhood NHL (EICNHL)," the "International Berlin-Frankfurt-Münster (i-BFM) Study Group," and the "NHL-Committee of the Italian Association of Pediatric Hematology Oncology (AIEOP)" [1, 2, 30].

Depending on the likelihood that a respective genetic defect is indeed present and directly or indirectly responsible for lymphoma development, the particular conditions can be subdivided into those in which such a connection is undoubtedly established, in which it has not yet been explicitly proven and in which it is either most likely unjustified and/or only an incidental concurrence of two otherwise unrelated events [1, 2]. According to these studies, one can expect that at least 60% of lymphoma cases in children and adolescents occur on the basis of bona fide predisposing genetic germline defects that are even commonly associated with already clinically recognizable syndromes. Compared to that, the group of heterogeneous and hitherto less clear-cut primary immunodefi-

ciency conditions is with up to 15% much smaller, whereas other non-risk syndromes or not unambiguously classifiable conditions make up another 20% and 10%, respectively [2].

Monogenic Disorders

The two main closely intertwined categories of monogenic disorders that not only predispose to lymphoma development but, with a certain propensity also to various other types of malignancies, are the DNA repair deficiency syndromes and inborn errors of immunities that include severe primary (SCID) as well as combined immunodeficiency (CVID) syndromes. Apart from these two groups, the respective lymphoma treatment studies contain also a number of otherwise well-defined genetic syndromes and nongenetic conditions, such as those with merely one or more organ malformations, which seem to be hardly relevant in this context. Given an estimated overall lifetime risk for developing lymphoma of approximately 2%, the frequency of the various disorders and the rarity of their coincidental occurrence, one can expect that this may be an unfortunate pure chance event. Until at least conceptually understandable or proven, any such assumed causal link must therefore remain completely speculative.

Among the noteworthy findings that became apparent in these and other more disease- or condition-specific oriented publications is the unequal distribution of histological subtypes in the different groups. Approximately 85% of patients with ataxia telangiectasia (AT) develop mature B-cell NHLs [5, 30–32], of which diffuse large cell forms (DLBCL) are roughly three times more common than Burkitt lymphoma (BL). Approximately 25% of patients with Nijmegen breakage syndrome (NBS) develop peripheral T-cell lymphoma (PTCL) [17, 28, 30, 33, 34], and approximately 80% of patients with constitutional mismatch repair deficiency (CMMRD) develop T-cell lymphoblastic lymphoma (T-LBL) [26, 35–37]. In contrast, approximately 60% of B-cell lymphoproliferations that take place in patients with primary or secondary immunodeficiencies are oligoclonal and polymorphic [5, 30]. Of note is also the overall inferior prognosis and increased risk of treatment-related toxicity and death in such patients compared to those with sporadic forms of lymphoid malignancies [2].

Since we will only superficially portray the most common and prominent representative examples in each of these categories, we refer the interested reader to the many excellent and extensive reviews of individual disease forms that can be found in the scientific literature as well as in several internet resources and compendia, such as "Online Inheritance of Man (https://www.omim.org/)," "Orphanet (www.orpha.net/)," and "Gene Reviews (www.ncbi.nlm.nih.gov/books/NBK1116/)."

Ataxia Telangiectasia

This autosomal recessive disorder has an estimated worldwide prevalence of 1:40.000–1:100.000. It is caused by mutations in the *ATM* gene, whose protein product is a prominent coordinating member of cellular signaling pathways that respond to DNA double-strand breaks as well as to oxidative and other genotoxic stress situations [31, 38]. The clinical consequences of a constitutional ATM-deficient DNA damage response are cerebellar degeneration, telangiectasia, immunodeficiency, cancer susceptibility, and radiation sensitivity (X- and gamma-rays), the latter of which has to be especially accounted for in the medical management of affected patients.

About two-thirds of AT patients suffer from immune system abnormalities, such as reduced T and B cells and low levels of one or more immunoglobulin classes. The lifetime risk to develop cancers is approximately 25%. The most common ones in those less than 20 years of age are lymphomas and leukemias, whereas adults also develop solid tumors including breast, liver, gastric, and esophageal carcinomas [31, 32, 39, 40].

Nijmegen Breakage Syndrome (NBS)

NBS is a similarly well-characterized and clinically recognizable autosomal recessive disorder that is caused by mutations in the *NBN* gene [33]. Although such cases can occasionally be encountered in any part of the world, a specific Slavic origin founder mutation (NM_02485.4:c.657_661del5) makes this mutation particularly common among Central and Eastern European populations. This circumstance facilitates its easy genetic verification especially in these geographic regions. The *NBN* gene encodes a subunit of the Mre11–Rad50–NBN (NMR) DNA double-strand break (DSB) repair complex [41]. Affected children are exceptionally sensitive to ionizing radiation or radiomimetics and share a strong predisposition to develop malignancies of predominantly lymphoid origin and, to a lesser extent, also brain tumors, such as medulloblastoma and glioma. Thus, more than 50% (56/105) of patients in the Polish NBS registry had developed a malignant disease, more than 90% (51/56) of which were lymphomas [28, 34]. Moreover, compared to sporadic lymphomas in children and in individuals with primary or secondary immunodeficiency disorders, they are primarily mature DLBCLs and BL or T-cell LBL/acute leukemias [28]. The estimated lymphoma risk is exceptionally high in NBS patients. Whereas it is increased already 70–250-fold in AT patients, it is increased more than 1.000-fold in NBS patients and therefore without doubt the highest among all the chromosome breakage and immunodeficiency syndromes [28].

The prognosis is generally poor because NBS patients experience an extremely high rate of malignancies and significant treatment-related toxicities as well as infectious complications. Nevertheless, long-term survival can be achieved already in a substantial number of affected children when one accounts for their specific vulnerability during treatment and transplantation, a procedure that will also reestablish their immunity again [17, 34].

Constitutional Mismatch Repair Deficiency (CMMRD)

CMMRD can be caused by mutations in four genes, *MLH1*, *MSH2*, *MSH6*, and *PMS2*, that regulate DNA mismatch repair [25, 42]. The autosomal-dominant Lynch syndrome (LS) results from heterozygous monoallelic germline loss-of-function mutations that predispose to the development of colorectal cancer, endometrial carcinoma, and other malignancies in adults. The distinct autosomal recessive childhood version of CMMRD, on the other hand, is caused by bi-allelic compound heterozygous or homozygous mutations that affect primarily the *PMS2* gene (60%) [25, 26, 35, 37, 43–46]. Affected children develop leukemias and lymphomas, brain (especially glioblastoma) and embryonic type, as well as LS-associated tumors [37]. Their overall prognosis is generally poor, not least because multiple such neoplasms often occur syn- or metachronously [36]. In contrast to AT and NBS patients, those with CMMRD experience no excessive treatment toxicity and the clinical effects of their immunodeficiency remain much subtler. Since one can often find particular physical attributes, i.e., café-au-lait spots, skin hypopigmentation, and pilomatricomas, in these patients that may otherwise also be encountered in other predisposing conditions, such as AT, Fanconi anemia, neurofibromatosis type 1, Li-Fraumeni syndrome, or Peutz-Jeghers syndrome, their differential diagnostic work-up requires clinical expertise and genetic scrutiny. T-cell malignancies in patients with pigment anomalies and consanguine parents are thus a virtually unmistakable indicator for an underlying causative CMMRD. Although several consortia put together helpful criteria and guidelines to support the diagnostic evaluation and surveillance of patients with CMMRD, their clinical utility has not yet been fully evaluated [25, 43–45]. One of the relevant recommendations put forward is that genetic testing in minors at risk is only warranted in case parents opt for surveillance or to exclude CMMRD prior to hematopoietic stem cell donation [42].

So far, 56 patients with CMMRD and hematological malignancies in 48 families are known in the literature, approximately one-third of which had lymphomas or leukemias [35, 45]. Their median age at diagnosis was 6 years (range 0.4–30 years). With 41 cases, lymphomas are the most frequent malignancies; 27 of them were of T- and 10 of B-cell origin (including 2 BL, 2 DLCBL, and 1 post-transplant lymphoproliferative disease). Of special note is not only the high proportion of T-cell lymphomas but especially also their unique and hitherto unexplainable mediastinal predilection. Approximately two-thirds of these patients were homozygotes and one-third compound heterozygotes. 58% of the mutations affected the *PMS2*, 25% the *MSH6*, and 17% each the *MLH1* and *MSH2* gene [35, 44].

Immunodeficiency Syndromes

The recent 2017 update of the "Primary Immunodeficiency Committee" of the "International Union of Immunological Societies" lists and categorizes 344 genetic defects that cause 354 distinct disorders of immunity [47, 48]. Of these, more than 20 are known to predispose to lymphoma (Table 8.2). Since an in-depth review of all these lymphoma-predisposing disorders is beyond the scope of our review, we will only briefly touch some relevant points in three representative examples. The overall sketchy general conclusions one can draw from publications dealing with this subject are that PID patients have a 1.42-fold excess to develop cancer, which is largely due to lymphoma in specific PID populations [49, 50]. The overall risk of individuals with PID to develop a malignant disease is 4–25%, which after infections constitutes their second leading cause of death. With nearly 60% (8.4% HD and 49.6% NHL) lymphoma is the predominant cancer subtype and thus a considerable problem in primary as well as acquired immunodeficiency syndromes [5, 51]. The predominant type of lymphoma is of B-cell origin, of which many of the small cell types are EBV-related [51–55].

Perforinopathies

The recently conceived term "perforinopathies" refers to a related group of perforin-deficient hyperinflammatory disorders with an increased cancer susceptibility, which may either result from rare congenital gene-impairing mono- or bi-allelic mutations or, in less severe forms, also be due to more common hypomorphic alleles [56–58]. Bi-allelic perforin gene (*PRF1*) mutations, in particular, are the cause of the familial hemophagocytic lymphohistiocytosis type 2 (FHL2) [59], a disease that shares some of its typical presenting features with ALCL and accounts for approximately 10–15% of all pediatric NHL [7, 60–63]. Approximately a quarter of these lymphoma patients carry monoallelic *PRF1* mutations but, remarkably, virtually none in *SH2D1A* or *UNC13D*, genes that are implicated in two other forms of FHL [60]. Mutations in *SH2D1A* are best known for causing the X-linked lymphoproliferative disease (XLP), which makes affected male carriers particularly vulnerable to

Table 8.2 Immunodeficiency and DNA repair syndromes that predispose to lymphoma development

OMIM	Condition/syndrome	Gene	Inheritance	Clinical features	Function	Damaging or trigger agent	Lymphoma types	References
208,900	Ataxia telangiectasia	*ATM*	AR	Progressive ataxia, telangiectasia, cellular & humoral ID, increased radiation sensitivity, chromosomal instability, infertility	DSB sensor, activates repair cascade	IR, AA, Bleo	4–five-fold increase in T-cell neoplasms	[17, 31, 32, 39, 91–93]
251,260	Nijmegen breakage syndrome	*NBN*	AR	Microcephaly & growth retardation, dysmorphic face, radiation sensitivity, chromosomal instability, reduced fertility	HR & NHEJ DSB repair, part of NMR complex, replication	IR, AA, Bleo	DLBCL, Burkitt NHL	[17, 28, 30, 33, 34]
126,391	Ligase 1 deficiency	*LIG1*	AR	Growth retardation, radiation & sun sensitivity, recurrent respiratory infections	NHEJ DSB repair, V(D)J recombination	IR, Bleo	T-cell lymphoma, DLBCL	[47]
606,593	Ligase 4 deficiency	*LIG4*	AR	Microcephaly & growth retardation, dysmorphic face, cellular & humoral immunodeficiency	NHEJ DSB repair, V(D)J recombination	IR, Bleo. EBV	EBV-associated lymphoma	[94–96]
210,900	Bloom syndrome	*RECQL3*	AR	Microcephaly & severe growth retardation	Helicase, chromatid separation, suppresses inappropriate HR, part of NMR complex	UV	B- and T-cell NHL, Burkitt	[97]
250,250	Cartilage hair hypoplasia (CHH)	*RMRP*	AR	Chondrodysplasia, immunodeficiency, recurrent infections	RNA component of mitochondrial RNA processing endoribonuclease		NHL	[98, 99]
276,300	Constitutional mismatch repair deficiency syndrome (CMMRD)	*PMS2* (60%), *MSH6, MSH2, MLH1*	AR	"Neurofibromatosis-like": café-au-lait spot and skin hypopigmentation, mild defects of immunoglobulin class switch, agenesis of the corpus callosum, pilomatricomas	DNA mismatch repair		(mediastinal) T-cell NHL	[25, 26, 35–37, 42–46]
603,553	Familial hemophagocytic lymphohistiocytosis (FHL2)	*PRF1*	AR	Hemophagocytic lymphohistiocytosis	Encodes a secreted glycoprotein that permeabilizes target cell membranes	EBV (NK/T-cell NHL)	ALCL, NK/T-cell NHL	[7, 11, 56–58, 60–63, 100, 101]
308,240	X-linked lymphoproliferative disease	*SH2D1A*	XL	Increased susceptibility to EBV infections	Involved in the bidirectional stimulation of T and B cells	EBV	DLBCL, Burkitt	[14, 52, 102–105]
186,973	Lymphoproliferative syndrome 1	*ITK*	AR	Lymphoproliferation	Non-receptor tyrosine kinase, role in T-cell growth, signaling and function	EBV	DLBCL, Burkitt	[52, 106]
615,122	Lymphoproliferative syndrome 2	*CD27*	AR	Lymphoproliferation, hemophagocytosis	Member of the tumor necrosis factor receptor superfamily	EBV	B-cell	[52, 107]

(continued)

Table 8.2 (continued)

OMIM	Condition/syndrome	Gene	Inheritance	Clinical features	Function	Damaging or trigger agent	Lymphoma types	References
602,840	CD70 deficiency	*TNFSF7*	AR	Immunodeficiency	Cytokine binds to CD27 and contributes to T-cell activation	EBV	B-cell	[52, 108, 109]
603,962	*RASGRP1* deficiency	*RASGRP1*	AR	Lymphoproliferation, defective T- and NK-cell function	Encodes a diacylglycerol-regulated guanidine exchange factor	EBV	B-cell	[110]
615,401	Immunodeficiency 8	*CORO1A*	AR	Infections, cognitive impairment	Actin-regulating protein expressed in hematopoietic cells	HPV, EBV	DLBCL	[52, 111, 112]
615,897	Immunodeficiency 24	*CTPS1 & CTPS2*	AR	Lymphoproliferation	Required for synthesis of a precursor of nucleic acids metabolism	Herpes viruses	B-cell	[52, 113, 114]
609,981	Immunodeficiency 54	*MCM4*	AR	Severe growth retardation, microcephaly, decreased numbers of natural killer cells, recurrent viral infections	Essential for initiation of eukaryotic genome replication	Herpes viruses (cytomegaly, EBV)	B-cell	[115]
300,853	XMEN	*MAGT1*	XL	Recurrent & chronic infections, cytopenias, low CD4 T-cell counts	Magnesium transporter	EBV	DLBCL, Burkitt Hodgkin	[52, 116]
301,000	Wiskott-Aldrich syndrome	*WAS*	XL	Thrombocytopenia, neutropenia, eczema, recurrent infections, autoimmunity	Key regulator of actin polymerization in hematopoietic cells	EBV	B-cell	[65, 66]
124,092 146,933 123,889	Interleukin10 (receptor) deficiency	*IL10, IL-10R1, IL-10R2*	AR	Severe chronic intestinal bowel inflammation	Encode IL10 and the two IL10 receptor chains	Immunosuppressive therapy (azathioprine)	DLBCL	[18]
614,868	T-cell deficiency	*STK4*	AR	Lymphoproliferation, progressive loss of T cells, recurrent infections, warts, abscesses, autoimmunity, cardiac malformations	Serine/threonine kinase 4, can phosphorylate myelin basic protein, undergoes autophosphorylation	HPV, herpes viruses	Cardiac T-cell lymphoma	[55, 117]
602,037	*RHOH* deficiency	*RHOH*	AR	HPV infection, molluscum contagiosum, lung granuloma	Member of the RAS superfamily of guanosine triphosphate -metabolizing enzymes	Human papillomavirus	Burkitt	[118]
601,859	Autoimmune proliferative (Canale-smith) syndrome	*FAS, FASLG*	AD	Lymphoproliferation	Apoptosis defect	–	B- & T-cell lymphomas	[6, 7]

AA alkylating agents, *AR* autosomal recessive, *Bleo* Bleomycin, *EBV* Epstein-Barr virus, *HPV* human papillomavirus, *IR* ionizing radiation, *NK* natural killer (cell), *XL* X-linked

Epstein-Barr virus (EBV) infections [64]. One of the severe complications of the accompanying and uncontrollable lymphoproliferations are B-cell lymphomas that develop in a quarter of the respective patients [64]. Noteworthy in this context is also the postulated predisposing role of an otherwise common activity-diminishing *PRF1* gene variant (SNP A91V; rs35947132) in the nasal form of NK/T-cell lymphoma in adults, which is the most frequent EBV-related NK/T-cell malignancy [63].

Wiskott-Aldrich Syndrome (WAS)

This rare X-linked genetic disorder is caused by heterogeneous mutations in the *WAS* gene, which is exclusively expressed in hematopoietic cells [65–68]. So far, approximately 300 different mutations are known, which are scattered over the entire gene. The encoded gene product (WASp) belongs to a family of proteins that relay signals from the cell surface to the actin cytoskeleton [69]. The wide spectrum of clinical symptoms and hematopoietic effects one encounters in this disorder can be clearly attributed to the different types and location of the respective mutations and which are therefore also directly responsible for the severity of the disease. The ensuing problems range from only mild forms of isolated micro-thrombocytopenia or neutropenia to severe forms of eczema, recurrent infections, and autoimmune and neoplastic diseases. The prevalence of malignancy in retrospective studies of patients with severe clinical presentations and an average age of onset of 9.5 years has been estimated to be around 20% and to especially affect those with autoimmune manifestations [65]. The most frequent, often EBV-associated forms of neoplasms are extra-nodal NHLs [65]. One of the postulated mechanisms that apparently facilitate lymphoma development and progression in this disorder is that malfunctioning dendritic, T and NK cells are incapable to keep virally infected or otherwise altered preneoplastic B cells under control and to eliminate them properly [65, 67].

Interleukin (IL)10 and IL-10 Receptor

Interleukin-10 (IL-10) and IL-10 receptor (IL-10R) deficiencies are the first recognized monogenic causes of very early onset severe inflammatory bowel disease [18, 70–72]. This immunoregulatory disorder predisposes to the development of unique monoclonal EBV-negative DLBCL subtypes of germinal center origin that are characterized by a constitutive activation of the NF-kB pathway and a defective local T-cell immune response. Taking into account all 35 reported patients with IL-10 deficiency (5 with IL-10, 11 with IL-10R1, and 19 with IL-10R2), the likelihood to develop lymphoma is estimated to be 36% (5 of 14) at the age of 7 years [72]. These observations clearly indicate that a defective IL-10 pathway is causatively involved in lymphoma development, although one also needs to point out that apparently neither gut inflammation itself nor a distinct pattern of inflammation seems to be the essential causative factor. The increased risk might rather be more connected with the immunosuppressive therapy in the form of azathioprine, which four of the five patients reported by Neven et al. had received [72]. In line with this observation is that thiopurine treatment of inflammatory bowel diseases in adult patients also increases the risk for such lymphoproliferative disorders significantly [73].

Genetic Factors Predisposing to "Sporadic" NHL

Despite the large number of hitherto already identified predisposing monogenic causes, it is clear that even in these instances, the development of lymphoma is a multifactorial process with some probabilistic elements that depend on and involve a liable genetic architecture as well as the participation and interaction of a multitude of other intrinsic (as regards the respective cells, organs, and organism) as well as extrinsic environmental triggers [74]. So far, our understanding of all these lymphoma-initiating and lymphoma-promoting processes primarily derive from such rare monogenic subtypes. However, it is to be expected that the continuous systematic analyses of the rich source of "sporadic" cases, i.e., those in which such a definable genetic component is not (yet) known, will without doubt provide us with a plethora of novel findings and relevant insights. The best evidence that the class of sporadic lymphoma may indeed encompass many more distinct genetic sub-entities is the growing numbers of novel mutations that are still identified especially in rare forms of immunodeficiencies. The notion of a polygenic causation and possible inheritance of such sporadic cases derives, among others, mainly from the observation that lymphoma risk can aggregate in families, albeit without evidence of a clear-cut Mendelian segregation trait. One common interpretation of this phenomenon is that each lymphoma arises in a particular individual based on the combined risk-contributing effects of a large number of otherwise irrelevant modifying genetic variants.

Again, there are multiple ways to assess a familial predisposition and to identify germline susceptibility loci. These include twin, case-control, and registry-based studies for the former and linkage and genetic association studies for the later [23, 75]. Based on a comprehensive overview of such studies, Cerhan et al. reported that in the United States, the estimated overall lifetime risk for developing NHL outside of rare hereditary syndromes is 1 in 48 (2.1%) [23]. The relative risk for first-degree relatives is 1.7-fold elevated, whereas their absolute lifetime risk is 3.6%. The absolute risk is even lower for specific lymphoma subtypes. One noteworthy observation was that there is apparently both commonality and heterogeneity for risk factors by NHL subtype [10].

Familial Predisposition

Although family history is commonly used to identify individuals with a possible heritable predisposition, especially within the pediatric cancer population, it is hardly able to predict such a susceptibility in most patients [76, 77], a problem that has many reasons, in particular incomplete information on family history, small family size, de novo mutations, and incomplete penetrance. Moreover, parents and other first- or second-degree relatives are often still young and cancer may not have developed yet. Notwithstanding all these obstacles, multiple lines of data nevertheless suggest that a family history of lymphoma is indeed associated with an increased risk of lymphoma. Familial risk is elevated for multiple lymphoma subtypes and familial risk does not seem to be confounded by nongenetic risk factors, although there are likely unidentified environmental risk factors and clustering of known (and unknown) such risk factors within families that are difficult to exclude. This suggests that at least some lymphoma subtypes share a genetic etiology. Moreover, genetic factors are also likely to be subtype-specific because a family history of a particular subtype is also most strongly associated with a risk for the same lymphoma subtype.

Twin Studies

The largest twin study that aimed to elucidate a genetic susceptibility to HD comprised altogether 187 dizygotic and 179 monozygotic twins [78]. Compared to background rates, this study found a 100-fold higher risk for a monozygotic twin to also be affected by HD but no excess risk for a dizygotic twin. The relatively young average age at diagnosis of the twins concordant for HD and the relatively short average interval between diagnoses in each pair of twins further corroborate the importance of genetic factors in this context. There was also a 23-fold higher risk of NHL for a patient's monozygotic twin but only a 14-fold higher risk for a patient's dizygotic twin, which indicates that in these instances a shared environment is probably more relevant for their increased NHL susceptibility.

Familial Aggregation

Case-control, cohort, and registry-based studies investigate whether and to which extent an inherited genetic risk to a particular disease, in this instance lymphoma, aggregates within families. Such studies are to a certain extent complicated by the impossibility to reliably separate a shared genetic background from the impact of a shared environment as well as the need to also account for family size.

The largest case-control study available to date was performed by the "International Lymphoma Epidemiology Consortium." This meta-analysis comprised 17.471 NHL cases and 23.096 controls from 20 case control studies and found a 1.8-fold increased risk for patients who had a first-degree blood-related family member with NHL. Albeit less pronounced, this risk was also elevated for those who had a first-degree relative with HD or leukemia [10, 23].

Owing to the fact that only few large cohort studies with a sufficient number and detailed information of familial lymphoma cases are available, the risk for specific NHL subtypes is difficult to assess. A Swedish study that covered 3.5 million people over a 35 years period found a 7.2- and 8.8-fold higher risk in children and young adults to develop HD if a parent or sibling also had HD [79], whereas another study reported a six-fold higher risk for siblings [80]. A cohort study that included 120.000 female teachers in California concluded that a history of lymphoma in a first-degree relative was associated with a 1.7-fold higher risk of B-cell NHL [81].

In the Utah Cancer Registry, which linked population-based family registry with cancer registry data, the risk of NHL was increased 1.7-fold in first-degree relatives of a proband with NHL [82]. The most comprehensive data available on familial aggregation by lymphoma subtypes compared the cancer experience in first-degree relatives of lymphoma patients with that of relatives and matched population controls. First-degree relatives of HD patients had a 3.1-fold increase in risk of HD whereas risk of HD was not associated with a family history of NHL [83]. One striking finding in these studies is the NHL subtype-specific clustering of risk as exemplified by the fact that first-degree relatives of individuals with DLBCL had a 9.8-fold increased risk of also being affected by DLBCL.

Genetic Risk Factors

Linkage studies, which use multi-case families or sib pairs to search for shared regions of inherited alleles among affected individuals in an unbiased manner, were so far little rewarding as regards lymphoma research, a failure that might be due to small sample sizes or the lack of single high-penetrant variants in the investigated cohorts.

Genetic association studies, which rely on high-throughput genotyping of sequence variation in germline DNA became the predominant analytical method in genetic epidemiology. The two major types of association studies are candidate gene and genome wide association studies (GWAS).

Candidate gene studies are mainly driven by the a priori biologic knowledge of lymphoma and lymphoma-associated diseases, such as infectious or autoimmune ones, as well as those which derive from other cancers. Genes of particular interest in this context are those which are involved in immune function, cell cycle/proliferation, apoptosis, DNA repair, and carcinogen metabolism pathways. However, for a variety of reasons, most of these studies had only very limited success in identifying susceptibility loci in adult NHL. The most robust risk association was found between a tumor necrosis factor (*TNF*; rs1800629)/lymphotoxin-alpha (*LTA*; rs909253) haplotype and DLBCL [84], a SNP

(rs3789068) in the proapoptotic *BCL2L11* gene and B-cell NHL, as well as a SNP (rs3132453) in *PRRC2A* in the HLA class III region and B-cell NHL [85].

GWAS uses dense microarrays with several hundred thousand SNPs that are distributed over the entire genome. As all loci are considered equally, such an analytic approach is considered as being hypothesis-free or "agnostic" [23]. To date, such GWAS studies have successfully identified 11 regions that are associated with follicular lymphoma and 6 with DLBCL risk in adults (Table 8.3). The respective common SNPs define loci with a minor allele frequency below 5%, have small effect sizes, and are of largely unknown function. Moreover, so far hardly any of these loci have been also verified in replicate studies.

Table 8.3 GWAS-discovered loci predisposing to follicular and diffuse large B-cell lymphoma in adults of diverse ethnic origin [86]

Chromosomal location	SNP	Nearest gene	References
Follicular lymphoma			
3q28	rs6444305	*LPP*	[119]
6p21.32	rs10484561	*MHC class II*	[120]
6p21.32	rs2647012	*HLA-*	[121]
6p21.32	–	*HLA-DRβ1 Glu*	[119]
6p21.32	rs17203612	*HLA-DRA*	[119]
6p21.33	rs3130437	*HLA-C*	[119]
6p21.33	rs6457327	*C6orf15* et al. *(STG)*	[119]
8q24.21	rs13254990	*PVT1*	[119]
11q23.3	rs4938573	*CXCR5*	[119]
11q24.3	rs4937362	*ETS1*	[119]
18q21.33	rs17749561	*BCL2*	[119]
Diffuse large B-cell lymphoma (DLBCL)			
2p23.3	rs79480871	*NCOA1*	[122]
3q27	rs6773854	*BCL6/LPP*	[123]
6p21.33	rs2523607	*HLA-B*	[122]
6p25.3	rs116446171	*EXOC2*	[122]
8q24.21	rs13255292	*PVT1*	[122]
8q24.21	rs4733601	*PVT1*	[122]

Genetic Testing, Screening, and Counseling Issues

Although all these epidemiologic and "agnostic" mass screening methods for assessing, exploring, and defining genetic risk factors for lymphoma development have certainly their merits, they are hardly of any value for the daily management of individual lymphoma patients. Compared to that, the hitherto pursued approach to search for and verify a genetic cause in particular individuals, which relied primarily on the recognition of associated symptoms and, as such, on the a priori knowledge and alertness of the treating physicians, was still much more rewarding [15, 27, 51]. However, the growing awareness of the high frequency and heterogeneity of such underlying conditions, some of which are also often difficult to recognize and delineate, as well as the continuous improvement of cost-efficient sequencing methods and bioinformatic tools, will definitely lead to a change in the diagnostic evaluation tactic [77, 86]. Given the increasing interest in the role of germline cancer susceptibility in general and in the pediatric setting in particular, it is to be expected that the assessment of lymphoma-associated genetic predisposition factors will soon be performed in a more systematic manner. It is somehow surprising that, to our knowledge, suitable screening programs have not yet been considered or implemented in current lymphoma treatment studies. Given what is known so far and given the high number and variety of such vastly unexplored predisposing immunodeficiencies, it is expected that compared to other cancer and leukemia predisposing conditions such an endeavor must be especially worthwhile in the lymphoma setting. Several pilot projects dealing with other malignancies in children provide some ideas how such programs could be installed [77, 86, 87]. As outlined in Fig. 8.2, there

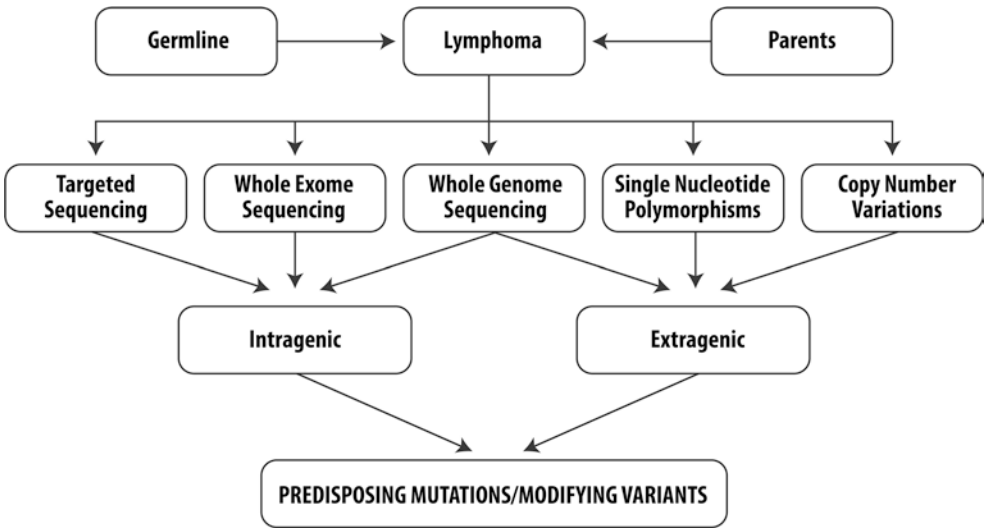

Fig. 8.2 Diagnostic approaches for the genetic assessment of predisposing risk factors in lymphoma patients, whose individual and combined values, advantages, and disadvantages are outlined in the main text

are several stepwise possibilities to do so. The most comprehensive one would be of course to sequence and compare DNA samples from the respective lymphoma together with one from the germline as well as the patient's parents. Depending on the infrastructural possibilities as well as cost/benefit calculations, one could perform such analyses either simultaneously or consecutively. The former is certainly more expensive but has the advantage that one can immediately compare the inheritance patterns of any identified abnormal results and thereby assess their validity and relevance in a rapid manner. The latter is probably cheaper because, in principle, one can concentrate only on the comparative confirmation of a smaller number of potentially relevant preselected markers. However, this approach could turn out to be more work- and also more time-consuming to obtain the essential information. Finally, there is also the question what one looks for and what one wants or needs to achieve in such a setting. For simple, clear-cut and easy to resolve diagnostic question, such as verification of a Nijmegen breakage syndrome or carrier screening for already known mutations, simple PCR analyses are clearly sufficient. For any other diagnostic evaluation, we consider targeted screening as the nowadays necessary minimal and also most cost-efficient standard, whereby the respective screening panel should cover at least all those genes that have already been implicated in lymphoma development [88–90]. More extensive sequencing methods that will eventually also aid the discovery of novel variants of potential relevance and interest, include whole exome sequencing (WES), which sooner or later will in any case most likely replace target sequencing, as well as whole genome sequencing (WGS), which has the advantage that it can also identify mutations in the non-coding extragenic part of the genome [86]. Moreover, a hitherto largely unexplored area in the field of lymphoma predisposition research is the conceivable contribution of the multitude of structural and copy number variations in the genome, especially of those which affect lymphoma-relevant gene regions. Although at present, these variants can be best assessed with DNA arrays, it is foreseeable that also this technique will eventually be replaced by whole genome as well as long-range sequencing procedures. With the appropriate bioinformatic support, these tools are not only able to significantly improve and refine these analyses, but at the same time, they will eventually also allow the simultaneously evaluation of associated epigenetic modifications, such as methylation.

Naturally, these remarkable technological advances and foreseeable developments in the diagnosis and research of lymphoma susceptibility also cause a large number of novel legal, ethical, social, and counseling problems, which can only be successfully resolved in a close interdisciplinary collaboration on a national but, even more so, on an international level. The particular topics that eventually need to be regulated comprise the informed consent and assent for minors undergoing testing, the ensuing implications for healthy siblings and parents of our patients, the timing of referral for genetic testing as well as the provision of a continuous educational and counseling support. All these issues are currently already addressed and discussed by a large number of experts from many countries who work together in two large recently established consortia, namely, the EU-funded COST Action "LEukaemia GENe Discovery by data sharing, mining and collaboration (LEGEND)" and the "IBFM Leukemia & Lymphoma Genetic Predisposition Committee."

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References

1. Arico M, Mussolin L, Carraro E, Buffardi S, Santoro N, D'Angelo P, et al. Non-Hodgkin lymphoma in children with an associated inherited condition: a retrospective analysis of the Associazione Italiana Ematologia Oncologia Pediatrica (AIEOP). Pediatr Blood Cancer. 2015;62(10):1782–9. https://doi.org/10.1002/pbc.25565.
2. Attarbaschi A, Carraro E, Abla O, Barzilai-Birenboim S, Bomken S, Brugieres L, et al. Non-Hodgkin lymphoma and pre-existing conditions: spectrum, clinical characteristics and outcome in 213 children and adolescents. Haematologica. 2016;101(12):1581–91. https://doi.org/10.3324/haematol.2016.147116.
3. Vijai J, Wang Z, Berndt SI, Skibola CF, Slager SL, de Sanjose S, et al. A genome-wide association study of marginal zone lymphoma shows association to the HLA region. Nat Commun. 2015;6(1):5751. https://doi.org/10.1038/ncomms6751.
4. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127(20):2375–90. https://doi.org/10.1182/blood-2016-01-643569.
5. Tran H, Nourse J, Hall S, Green M, Griffiths L, Gandhi MK. Immunodeficiency-associated lymphomas. Blood Rev. 2008;22(5):261–81. https://doi.org/10.1016/j.blre.2008.03.009.
6. Straus SE, Jaffe ES, Puck JM, Dale JK, Elkon KB, Rosen-Wolff A, et al. The development of lymphomas in families with autoimmune lymphoproliferative syndrome with germline Fas mutations and defective lymphocyte apoptosis. Blood. 2001;98(1):194–200.
7. Clementi R, Dagna L, Dianzani U, Dupre L, Dianzani I, Ponzoni M, et al. Inherited perforin and Fas mutations in a patient with autoimmune lymphoproliferative syndrome and lymphoma. N Engl J Med. 2004;351(14):1419–24. https://doi.org/10.1056/NEJMoa041432.
8. Alexander DD, Mink PJ, Adami HO, Chang ET, Cole P, Mandel JS, et al. The non-Hodgkin lymphomas: a review of the epidemiologic literature. Int J Cancer. 2007;120 Suppl 12(S12):1–39. https://doi.org/10.1002/ijc.22719.
9. Cunningham-Rundles C. The many faces of common variable immunodeficiency. Hematology Am Soc Hematol Educ Program. 2012;2012:301–5. https://doi.org/10.1182/asheducation-2012.1.301.
10. Morton LM, Slager SL, Cerhan JR, Wang SS, Vajdic CM, Skibola CF, et al. Etiologic heterogeneity among non-Hodgkin lymphoma subtypes: the InterLymph Non-Hodgkin Lymphoma Subtypes

Project. J Natl Cancer Inst Monogr. 2014;2014(48):130–44. https://doi.org/10.1093/jncimonographs/lgu013.
11. Osinska I, Popko K, Demkow U. Perforin: an important player in immune response. Cent Eur J Immunol. 2014;39(1):109–15. https://doi.org/10.5114/ceji.2014.42135.
12. Skibola CF, Curry JD, Nieters A. Genetic susceptibility to lymphoma. Haematologica. 2007;92(7):960–9.
13. Similuk M, Rao VK, Churpek J, Lenardo M. Predispositions to lymphoma: a practical review for Genetic Counselors. J Genet Couns. 2016;25(6):1157–70. https://doi.org/10.1007/s10897-016-9979-0.
14. Sharapova SO, Fedorova AS, Pashchenko OE, Vahliarskaya SS, Guryanova IE, Migas AA, et al. Novel mutations in SH2D1A gene in X-linked lymphoproliferative syndrome, diagnosed after B-cell non-Hodgkin lymphoma. J Pediatr Hematol Oncol. 2017;39(4):e203–e6. https://doi.org/10.1097/MPH.0000000000000815.
15. Jongmans MC, Loeffen JL, Waanders E, Hoogerbrugge PM, Ligtenberg MJ, Kuiper RP, et al. Recognition of genetic predisposition in pediatric cancer patients: an easy-to-use selection tool. Eur J Med Genet. 2016;59(3):116–25. https://doi.org/10.1016/j.ejmg.2016.01.008.
16. Seidemann K, Tiemann M, Henze G, Sauerbrey A, Muller S, Reiter A. Therapy for non-Hodgkin lymphoma in children with primary immunodeficiency: analysis of 19 patients from the BFM trials. Med Pediatr Oncol. 1999;33(6):536–44.
17. Bienemann K, Burkhardt B, Modlich S, Meyer U, Moricke A, Bienemann K, et al. Promising therapy results for lymphoid malignancies in children with chromosomal breakage syndromes (Ataxia telangiectasia or Nijmegen-breakage syndrome): a retrospective survey. Br J Haematol. 2011;155(4):468–76. https://doi.org/10.1111/j.1365-2141.2011.08863.x.
18. Kotlarz D, Beier R, Murugan D, Diestelhorst J, Jensen O, Boztug K, et al. Loss of interleukin-10 signaling and infantile inflammatory bowel disease: implications for diagnosis and therapy. Gastroenterology. 2012;143(2):347–55. https://doi.org/10.1053/j.gastro.2012.04.045.
19. Chihara D, Nastoupil LJ, Williams JN, Lee P, Koff JL, Flowers CR. New insights into the epidemiology of non-Hodgkin lymphoma and implications for therapy. Expert Rev Anticancer Ther. 2015;15(5):531–44. https://doi.org/10.1586/14737140.2015.1023712.
20. Hampel H, Bennett RL, Buchanan A, Pearlman R, Wiesner GL, Guideline Development Group ACoMGaGPPaGCaNSoGCPGC. A practice guideline from the American College of Medical Genetics and Genomics and the National Society of Genetic Counselors: referral indications for cancer predisposition assessment. Genet Med. 2015;17(1):70–87.
21. Wolfe Schneider K, Jasperson K. Unique genetic counseling considerations in the pediatric oncology setting. Curr Genet Med Rep. 2015;3(2):65–73. https://doi.org/10.1007/s40142-015-0064-z.
22. Walsh MF, Chang VY, Kohlmann WK, Scott HS, Cunniff C, Bourdeaut F, et al. Recommendations for childhood cancer screening and surveillance in DNA repair disorders. Clin Cancer Res. 2017;23(11):e23–31. https://doi.org/10.1158/1078-0432.CCR-17-0465.
23. Cerhan JR, Slager SL. Familial predisposition and genetic risk factors for lymphoma. Blood. 2015;126(20):2265–73. https://doi.org/10.1182/blood-2015-04-537498.
24. Desai AV, Perpich M, Godley LA. Clinical assessment and diagnosis of Germline predisposition to hematopoietic malignancies: the University of Chicago Experience. Front Pediatr. 2017;5:252. https://doi.org/10.3389/fped.2017.00252.
25. Bakry D, Aronson M, Durno C, Rimawi H, Farah R, Alharbi QK, et al. Genetic and clinical determinants of constitutional mismatch repair deficiency syndrome: report from the constitutional mismatch repair deficiency consortium. Eur J Cancer. 2014;50(5):987–96. https://doi.org/10.1016/j.ejca.2013.12.005.
26. Ripperger T, Beger C, Rahner N, Sykora KW, Bockmeyer CL, Lehmann U, et al. Constitutional mismatch repair deficiency and childhood leukemia/lymphoma – report on a novel biallelic MSH6 mutation. Haematologica. 2010;95(5):841–4. https://doi.org/10.3324/haematol.2009.015503.
27. Ripperger T, Bielack SS, Borkhardt A, Brecht IB, Burkhardt B, Calaminus G, et al. Childhood cancer predisposition syndromes-a concise review and recommendations by the Cancer Predisposition Working Group of the Society for Pediatric Oncology and Hematology. Am J Med Genet A. 2017;173(4):1017–37. https://doi.org/10.1002/ajmg.a.38142.
28. Gladkowska-Dura M, Dzierzanowska-Fangrat K, Dura WT, van Krieken JH, Chrzanowska KH, van Dongen JJ, et al. Unique morphological spectrum of lymphomas in Nijmegen breakage syndrome (NBS) patients with high frequency of consecutive lymphoma formation. J Pathol. 2008;216(3):337–44. https://doi.org/10.1002/path.2418.
29. Shapiro RS. Malignancies in the setting of primary immunodeficiency: implications for hematologists/oncologists. Am J Hematol. 2011;86(1):48–55. https://doi.org/10.1002/ajh.21903.
30. Seidemann K, Henze G, Beck JD, Sauerbrey A, Kuhl J, Mann G, et al. Non-Hodgkin's lymphoma in pediatric patients with chromosomal breakage syndromes (AT and NBS): experience from the BFM trials. Ann Oncol. 2000;11(Suppl 1):141–5.
31. Rothblum-Oviatt C, Wright J, Lefton-Greif MA, McGrath-Morrow SA, Crawford TO, Lederman HM. Ataxia telangiectasia: a review. Orphanet J Rare Dis. 2016;11(1):159. https://doi.org/10.1186/s13023-016-0543-7.
32. Suarez F, Mahlaoui N, Canioni D, Andriamanga C, Dubois d'Enghien C, Brousse N, et al. Incidence, presentation, and prognosis of malignancies in ataxia-telangiectasia: a report from the French national registry of primary immune deficiencies. J Clin Oncol. 2015;33(2):202–8. https://doi.org/10.1200/JCO.2014.56.5101.
33. Chrzanowska KH, Gregorek H, Dembowska-Baginska B, Kalina MA, Digweed M. Nijmegen breakage syndrome (NBS). Orphanet J Rare Dis. 2012;7(1):13. https://doi.org/10.1186/1750-1172-7-13.
34. Dembowska-Baginska B, Perek D, Brozyna A, Wakulinska A, Olczak-Kowalczyk D, Gladkowska-Dura M, et al. Non-Hodgkin lymphoma (NHL) in children with Nijmegen breakage syndrome (NBS). Pediatr Blood Cancer. 2009;52(2):186–90. https://doi.org/10.1002/pbc.21789.
35. Ripperger T, Schlegelberger B. Acute lymphoblastic leukemia and lymphoma in the context of constitutional mismatch repair deficiency syndrome. Eur J Med Genet. 2016;59(3):133–42. https://doi.org/10.1016/j.ejmg.2015.12.014.
36. Alexander TB, McGee RB, Kaye EC, McCarville MB, Choi JK, Cavender CP, et al. Metachronous T-lymphoblastic lymphoma and Burkitt lymphoma in a child with constitutional mismatch repair deficiency syndrome. Pediatr Blood Cancer. 2016;63(8):1454–6. https://doi.org/10.1002/pbc.25989.
37. Lavoine N, Colas C, Muleris M, Bodo S, Duval A, Entz-Werle N, et al. Constitutional mismatch repair deficiency syndrome: clinical description in a French cohort. J Med Genet. 2015;52(11):770–8. https://doi.org/10.1136/jmedgenet-2015-103299.
38. Tichy A, Vavrova J, Pejchal J, Rezacova M. Ataxia-telangiectasia mutated kinase (ATM) as a central regulator of radiation-induced DNA damage response. Acta Med (Hradec Kralove). 2010;53(1):13–7.
39. Taylor AM, Metcalfe JA, Thick J, Mak YF. Leukemia and lymphoma in ataxia telangiectasia. Blood. 1996;87(2):423–38.
40. van Os NJ, Roeleveld N, Weemaes CM, Jongmans MC, Janssens GO, Taylor AM, et al. Health risks for ataxia-telangiectasia

mutated heterozygotes: a systematic review, meta-analysis and evidence-based guideline. Clin Genet. 2016;90(2):105–17. https://doi.org/10.1111/cge.12710.
41. Schuetz JM, MaCarthur AC, Leach S, Lai AS, Gallagher RP, Connors JM, et al. Genetic variation in the NBS1, MRE11, RAD50 and BLM genes and susceptibility to non-Hodgkin lymphoma. BMC Med Genet. 2009;10:117. https://doi.org/10.1186/1471-2350-10-117.
42. Tabori U, Hansford JR, Achatz MI, Kratz CP, Plon SE, Frebourg T, et al. Clinical management and tumor surveillance recommendations of inherited mismatch repair deficiency in childhood. Clin Cancer Res. 2017;23(11):e32–e7. https://doi.org/10.1158/1078-0432.CCR-17-0574.
43. Durno CA, Sherman PM, Aronson M, Malkin D, Hawkins C, Bakry D, et al. Phenotypic and genotypic characterisation of biallelic mismatch repair deficiency (BMMR-D) syndrome. Eur J Cancer. 2015;51(8):977–83. https://doi.org/10.1016/j.ejca.2015.02.008.
44. Wimmer K, Kratz CP, Vasen HF, Caron O, Colas C, Entz-Werle N, et al. Diagnostic criteria for constitutional mismatch repair deficiency syndrome: suggestions of the European consortium 'care for CMMRD' (C4CMMRD). J Med Genet. 2014;51(6):355–65. https://doi.org/10.1136/jmedgenet-2014-102284.
45. Vasen HF, Ghorbanoghli Z, Bourdeaut F, Cabaret O, Caron O, Duval A, et al. Guidelines for surveillance of individuals with constitutional mismatch repair-deficiency proposed by the European consortium "care for CMMR-D" (C4CMMR-D). J Med Genet. 2014;51(5):283–93. https://doi.org/10.1136/jmedgenet-2013-102238.
46. Wimmer K, Kratz CP. Constitutional mismatch repair-deficiency syndrome. Haematologica. 2010;95(5):699–701. https://doi.org/10.3324/haematol.2009.021626.
47. Picard C, Bobby Gaspar H, Al-Herz W, Bousfiha A, Casanova JL, Chatila T, et al. International Union of Immunological Societies: 2017 Primary Immunodeficiency Diseases Committee Report on Inborn Errors of Immunity. J Clin Immunol. 2018;38(1):96–128. https://doi.org/10.1007/s10875-017-0464-9.
48. Bousfiha A, Jeddane L, Picard C, Ailal F, Bobby Gaspar H, Al-Herz W, et al. The 2017 IUIS phenotypic classification for primary Immunodeficiencies. J Clin Immunol. 2018;38(1):129–43. https://doi.org/10.1007/s10875-017-0465-8.
49. Mayor PC, Eng KH, Singel KL, Abrams SI, Odunsi K, Moysich KB, et al. Cancer in primary immunodeficiency diseases: cancer incidence in the United States Immune Deficiency Network Registry. J Allergy Clin Immunol. 2018;141(3):1028–35. https://doi.org/10.1016/j.jaci.2017.05.024.
50. Verhoeven D, Stoppelenburg AJ, Meyer-Wentrup F, Boes M. Increased risk of hematologic malignancies in primary immunodeficiency disorders: opportunities for immunotherapy. Clin Immunol. 2018;190:22–31. https://doi.org/10.1016/j.clim.2018.02.007.
51. de Jong D, Roemer MG, Chan JK, Goodlad J, Gratzinger D, Chadburn A, et al. B-cell and classical Hodgkin lymphomas associated with immunodeficiency: 2015 SH/EAHP Workshop Report-Part 2. Am J Clin Pathol. 2017;147(2):153–70. https://doi.org/10.1093/ajcp/aqw216.
52. Latour S, Winter S. Inherited Immunodeficiencies with high predisposition to Epstein–Barr virus-driven lymphoproliferative diseases. Front Immunol. 2018;9:1103. https://doi.org/10.3389/fimmu.2018.01103.
53. Arjunaraja S, Angelus P, Su HC, Snow AL. Impaired control of Epstein-Barr virus infection in B-cell expansion with NF-kappaB and T-cell Anergy Disease. Front Immunol. 2018;9:198. https://doi.org/10.3389/fimmu.2018.00198.
54. Worth AJ, Houldcroft CJ, Booth C. Severe Epstein-Barr virus infection in primary immunodeficiency and the normal host. Br J Haematol. 2016;175(4):559–76. https://doi.org/10.1111/bjh.14339.
55. Parvaneh N, Filipovich AH, Borkhardt A. Primary immunodeficiencies predisposed to Epstein-Barr virus-driven haematological diseases. Br J Haematol. 2013;162(5):573–86. https://doi.org/10.1111/bjh.12422.
56. Voskoboinik I, Trapani JA. Perforinopathy: a spectrum of human immune disease caused by defective perforin delivery or function. Front Immunol. 2013;4:441. https://doi.org/10.3389/fimmu.2013.00441.
57. Trapani JA, Thia KY, Andrews M, Davis ID, Gedye C, Parente P, et al. Human perforin mutations and susceptibility to multiple primary cancers. Oncoimmunology. 2013;2(4):e24185. https://doi.org/10.4161/onci.24185.
58. Chaudhry MS, Gilmour KC, House IG, Layton M, Panoskaltsis N, Sohal M, et al. Missense mutations in the perforin (PRF1) gene as a cause of hereditary cancer predisposition. Oncoimmunology. 2016;5(7):e1179415. https://doi.org/10.1080/2162402X.2016.1179415.
59. Cetica V, Sieni E, Pende D, Danesino C, De Fusco C, Locatelli F, et al. Genetic predisposition to hemophagocytic lymphohistiocytosis: report on 500 patients from the Italian registry. J Allergy Clin Immunol. 2016;137(1):188–96 e4. https://doi.org/10.1016/j.jaci.2015.06.048.
60. Ciambotti B, Mussolin L, d'Amore ES, Pillon M, Sieni E, Coniglio ML, et al. Monoallelic mutations of the perforin gene may represent a predisposing factor to childhood anaplastic large cell lymphoma. J Pediatr Hematol Oncol. 2014;36(6):e359–65. https://doi.org/10.1097/MPH.0000000000000073.
61. Clementi R, Locatelli F, Dupre L, Garaventa A, Emmi L, Bregni M, et al. A proportion of patients with lymphoma may harbor mutations of the perforin gene. Blood. 2005;105(11):4424–8. https://doi.org/10.1182/blood-2004-04-1477.
62. Cannella S, Santoro A, Bruno G, Pillon M, Mussolin L, Mangili G, et al. Germline mutations of the perforin gene are a frequent occurrence in childhood anaplastic large cell lymphoma. Cancer. 2007;109(12):2566–71. https://doi.org/10.1002/cncr.22718.
63. Manso R, Rodriguez-Pinilla SM, Lombardia L, Ruiz de Garibay G, Del Mar Lopez M, Requena L, et al. An A91V SNP in the perforin gene is frequently found in NK/T-cell lymphomas. PLoS One. 2014;9(3):e91521. https://doi.org/10.1371/journal.pone.0091521.
64. Panchal N, Booth C, Cannons JL, Schwartzberg PL. X-linked Lymphoproliferative disease type 1: a clinical and molecular perspective. Front Immunol. 2018;9:666. https://doi.org/10.3389/fimmu.2018.00666.
65. Candotti F. Clinical manifestations and pathophysiological mechanisms of the Wiskott-Aldrich syndrome. J Clin Immunol. 2018;38(1):13–27. https://doi.org/10.1007/s10875-017-0453-z.
66. Buchbinder D, Nugent DJ, Fillipovich AH. Wiskott-Aldrich syndrome: diagnosis, current management, and emerging treatments. Appl Clin Genet. 2014;7:55–66. https://doi.org/10.2147/TACG.S58444.
67. Massaad MJ, Ramesh N, Geha RS. Wiskott-Aldrich syndrome: a comprehensive review. Ann N Y Acad Sci. 2013;1285:26–43. https://doi.org/10.1111/nyas.12049.
68. Ochs HD, Filipovich AH, Veys P, Cowan MJ, Kapoor N. Wiskott-Aldrich syndrome: diagnosis, clinical and laboratory manifestations, and treatment. Biol Blood Marrow Transplant. 2009;15(1 Suppl):84–90. https://doi.org/10.1016/j.bbmt.2008.10.007.
69. Blundell MP, Worth A, Bouma G, Thrasher AJ. The Wiskott-Aldrich syndrome: the actin cytoskeleton and immune cell function. Dis Markers. 2010;29(3-4):157–75. https://doi.org/10.3233/DMA-2010-0735.
70. Glocker EO, Kotlarz D, Boztug K, Gertz EM, Schaffer AA, Noyan F, et al. Inflammatory bowel disease and mutations affecting the interleukin-10 receptor. N Engl J Med. 2009;361(21):2033–45. https://doi.org/10.1056/NEJMoa0907206.

71. Shouval DS, CL Ebens, R Murchie, K McCann, R Rabah, C Klein, et al. Large B-cell lymphoma in an adolescent patient with Iinterleukin-10 receptor deficiency and history of infantile inflammatory bowel disease. J Pediatr Gastroenterol Nutr. 2016;63(1):e15–e17. https://doi.org/10.1097/MPG.0000000000000532.
72. Neven B, Mamessier E, Bruneau J, Kaltenbach S, Kotlarz D, Suarez F, et al. A Mendelian predisposition to B-cell lymphoma caused by IL-10R deficiency. Blood. 2013;122(23):3713–22. https://doi.org/10.1182/blood-2013-06-508267.
73. Kandiel A, Fraser AG, Korelitz BI, Brensinger C, Lewis JD. Increased risk of lymphoma among inflammatory bowel disease patients treated with azathioprine and 6-mercaptopurine. Gut. 2005;54(8):1121–5. https://doi.org/10.1136/gut.2004.049460.
74. Mitchell KJ. What is complex about complex disorders? Genome Biol. 2012;13(1):237. https://doi.org/10.1186/gb-2012-13-1-237.
75. Cerhan JR. Host genetics in follicular lymphoma. Best Pract Res Clin Haematol. 2011;24(2):121–34. https://doi.org/10.1016/j.beha.2011.02.004.
76. Knapke S, Nagarajan R, Correll J, Kent D, Burns K. Hereditary cancer risk assessment in a pediatric oncology follow-up clinic. Pediatr Blood Cancer. 2012;58(1):85–9. https://doi.org/10.1002/pbc.23283.
77. Zhang J, Walsh MF, Wu G, Edmonson MN, Gruber TA, Easton J, et al. Germline mutations in predisposition genes in pediatric cancer. N Engl J Med. 2015;373(24):2336–46. https://doi.org/10.1056/NEJMoa1508054.
78. Mack TM, Cozen W, Shibata DK, Weiss LM, Nathwani BN, Hernandez AM, et al. Concordance for Hodgkin's disease in identical twins suggesting genetic susceptibility to the young-adult form of the disease. N Engl J Med. 1995;332(7):413–8. https://doi.org/10.1056/NEJM199502163320701.
79. Crump C, Sundquist K, Sieh W, Winkleby MA, Sundquist J. Perinatal and family risk factors for Hodgkin lymphoma in childhood through young adulthood. Am J Epidemiol. 2012;176(12):1147–58. https://doi.org/10.1093/aje/kws212.
80. Friedman DL, Kadan-Lottick NS, Whitton J, Mertens AC, Yasui Y, Liu Y, et al. Increased risk of cancer among siblings of long-term childhood cancer survivors: a report from the childhood cancer survivor study. Cancer Epidemiol Biomark Prev. 2005;14(8):1922–7. https://doi.org/10.1158/1055-9965.EPI-05-0066.
81. Lu Y, Sullivan-Halley J, Cozen W, Chang ET, Henderson K, Ma H, et al. Family history of haematopoietic malignancies and non-Hodgkin's lymphoma risk in the California teachers study. Br J Cancer. 2009;100(3):524–6. https://doi.org/10.1038/sj.bjc.6604881.
82. Goldgar DE, Easton DF, Cannon-Albright LA, Skolnick MH. Systematic population-based assessment of cancer risk in first-degree relatives of cancer probands. J Natl Cancer Inst. 1994;86(21):1600–8. https://doi.org/10.1093/jnci/86.21.1600.
83. Goldin LR, McMaster ML, Ter-Minassian M, Saddlemire S, Harmsen B, Lalonde G, et al. A genome screen of families at high risk for Hodgkin lymphoma: evidence for a susceptibility gene on chromosome 4. J Med Genet. 2005;42(7):595–601. https://doi.org/10.1136/jmg.2004.027433.
84. Skibola CF, Bracci PM, Nieters A, Brooks-Wilson A, de Sanjose S, Hughes AM, et al. Tumor necrosis factor (TNF) and lymphotoxin-alpha (LTA) polymorphisms and risk of non-Hodgkin lymphoma in the InterLymph Consortium. Am J Epidemiol. 2010;171(3):267–76. https://doi.org/10.1093/aje/kwp383.
85. Nieters A, Conde L, Slager SL, Brooks-Wilson A, Morton L, Skibola DR, et al. PRRC2A and BCL2L11 gene variants influence risk of non-Hodgkin lymphoma: results from the InterLymph consortium. Blood. 2012;120(23):4645–8. https://doi.org/10.1182/blood-2012-05-427989.
86. Kuhlen M, Borkhardt A. Cancer susceptibility syndromes in children in the area of broad clinical use of massive parallel sequencing. Eur J Pediatr. 2015;174(8):987–97. https://doi.org/10.1007/s00431-015-2565-x.
87. Kuhlen M, Borkhardt A. Trio sequencing in pediatric cancer and clinical implications. EMBO Mol Med. 2018;10(4):e8641–7. https://doi.org/10.15252/emmm.201708641.
88. Al-Mousa H, Abouelhoda M, Monies DM, Al-Tassan N, Al-Ghonaium A, Al-Saud B, et al. Unbiased targeted next-generation sequencing molecular approach for primary immunodeficiency diseases. J Allergy Clin Immunol. 2016;137(6):1780–7. https://doi.org/10.1016/j.jaci.2015.12.1310.
89. Nijman IJ, van Montfrans JM, Hoogstraat M, Boes ML, van de Corput L, Renner ED, et al. Targeted next-generation sequencing: a novel diagnostic tool for primary immunodeficiencies. J Allergy Clin Immunol. 2014;133(2):529–34. https://doi.org/10.1016/j.jaci.2013.08.032.
90. Stoddard JL, Niemela JE, Fleisher TA, Rosenzweig SD. Targeted NGS: a cost-effective approach to molecular diagnosis of PIDs. Front Immunol. 2014;5(1):531. https://doi.org/10.3389/fimmu.2014.00531.
91. Sandoval C, Swift M. Treatment of lymphoid malignancies in patients with ataxia-telangiectasia. Med Pediatr Oncol. 1998;31(6):491–7.
92. Taylor AM. Ataxia telangiectasia genes and predisposition to leukaemia, lymphoma and breast cancer. Br J Cancer. 1992;66(1):5–9.
93. van Os NJH, Haaxma CA, van der Flier M, Merkus PJFM, van Deuren M, de Groot IJM, et al. Ataxia-telangiectasia: recommendations for multidisciplinary treatment. Dev Med Child Neurol. 2017;59(7):680–9.
94. Ben-Omran TI, Cerosaletti K, Concannon P, Weitzman S, Nezarati MM. A patient with mutations in DNA ligase IV: clinical features and overlap with Nijmegen breakage syndrome. Am J Med Genet A. 2005;137A(3):283–7. https://doi.org/10.1002/ajmg.a.30869.
95. Sharapova SO, Chang EY, Guryanova IE, Proleskovskaya IV, Fedorova AS, Rutskaya EA, et al. Next generation sequencing revealed DNA ligase IV deficiency in a "developmentally normal" patient with massive brain Epstein-Barr virus-positive diffuse large B-cell lymphoma. Clin Immunol. 2016;163:108–10. https://doi.org/10.1016/j.clim.2016.01.002.
96. Altmann T, Gennery AR. DNA ligase IV syndrome; a review. Orphanet J Rare Dis. 2016;11(1):137. https://doi.org/10.1186/s13023-016-0520-1.
97. Cunniff C, Bassetti JA, Ellis NA. Bloom's syndrome: clinical spectrum, molecular pathogenesis, and cancer predisposition. Mol Syndromol. 2017;8(1):4–23. https://doi.org/10.1159/000452082.
98. Kostjukovits S, Klemetti P, Valta H, Martelius T, Notarangelo LD, Seppanen M, et al. Analysis of clinical and immunologic phenotype in a large cohort of children and adults with cartilage-hair hypoplasia. J Allergy Clin Immunol. 2017;140(2):612–4 e5. https://doi.org/10.1016/j.jaci.2017.02.016.
99. Taskinen M, Ranki A, Pukkala E, Jeskanen L, Kaitila I, Makitie O. Extended follow-up of the Finnish cartilage-hair hypoplasia cohort confirms high incidence of non-Hodgkin lymphoma and basal cell carcinoma. Am J Med Genet A. 2008;146A(18):2370–5. https://doi.org/10.1002/ajmg.a.32478.
100. Ding Q, Yang LY. Perforin gene mutations in 77 Chinese patients with lymphomas. World J Emerg Med. 2013;4(2):128–32. https://doi.org/10.5847/wjem.j.1920-8642.2013.02.008.
101. Mehta PA, Davies SM, Kumar A, Devidas M, Lee S, Zamzow T, et al. Perforin polymorphism A91V and susceptibility to B-precursor childhood acute lymphoblastic leukemia: a report from the Children's Oncology Group. Leukemia. 2006;20(9):1539–41. https://doi.org/10.1038/sj.leu.2404299.
102. Koochakzadeh L, Hosseinverdi S, Hedayat M, Farahani F, Tofighi A, Eghbali M, et al. Study of SH2D1A gene mutation in paediatric

patients with B-cell lymphoma. Allergol Immunopathol (Madr). 2015;43(6):568–70. https://doi.org/10.1016/j.aller.2015.01.007.
103. Tangye SG. XLP: clinical features and molecular etiology due to mutations in SH2D1A encoding SAP. J Clin Immunol. 2014;34(7):772–9. https://doi.org/10.1007/s10875-014-0083-7.
104. Booth C, Gilmour KC, Veys P, Gennery AR, Slatter MA, Chapel H, et al. X-linked lymphoproliferative disease due to SAP/SH2D1A deficiency: a multicenter study on the manifestations, management and outcome of the disease. Blood. 2011;117(1):53–62. https://doi.org/10.1182/blood-2010-06-284935.
105. Arico M, Imashuku S, Clementi R, Hibi S, Teramura T, Danesino C, et al. Hemophagocytic lymphohistiocytosis due to germline mutations in SH2D1A, the X-linked lymphoproliferative disease gene. Blood. 2001;97(4):1131–3.
106. Linka RM, Risse SL, Bienemann K, Werner M, Linka Y, Krux F, et al. Loss-of-function mutations within the IL-2 inducible kinase ITK in patients with EBV-associated lymphoproliferative diseases. Leukemia. 2012;26(5):963–71. https://doi.org/10.1038/leu.2011.371.
107. Salzer E, Daschkey S, Choo S, Gombert M, Santos-Valente E, Ginzel S, et al. Combined immunodeficiency with life-threatening EBV-associated lymphoproliferative disorder in patients lacking functional CD27. Haematologica. 2013;98(3):473–8. https://doi.org/10.3324/haematol.2012.068791.
108. Abolhassani H, Edwards ES, Ikinciogullari A, Jing H, Borte S, Buggert M, et al. Combined immunodeficiency and Epstein-Barr virus-induced B cell malignancy in humans with inherited CD70 deficiency. J Exp Med. 2017;214(1):91–106. https://doi.org/10.1084/jem.20160849.
109. Izawa K, Martin E, Soudais C, Bruneau J, Boutboul D, Rodriguez R, et al. Inherited CD70 deficiency in humans reveals a critical role for the CD70-CD27 pathway in immunity to Epstein-Barr virus infection. J Exp Med. 2017;214(1):73–89. https://doi.org/10.1084/jem.20160784.
110. Winter S, Martin E, Boutboul D, Lenoir C, Boudjemaa S, Petit A, et al. Loss of RASGRP1 in humans impairs T-cell expansion leading to Epstein-Barr virus susceptibility. EMBO Mol Med. 2018;10(2):188–99. https://doi.org/10.15252/emmm.201708292.
111. Stray-Pedersen A, Jouanguy E, Crequer A, Bertuch AA, Brown BS, Jhangiani SN, et al. Compound heterozygous CORO1A mutations in siblings with a mucocutaneous-immunodeficiency syndrome of epidermodysplasia verruciformis-HPV, molluscum contagiosum and granulomatous tuberculoid leprosy. J Clin Immunol. 2014;34(7):871–90. https://doi.org/10.1007/s10875-014-0074-8.
112. Moshous D, Martin E, Carpentier W, Lim A, Callebaut I, Canioni D, et al. Whole-exome sequencing identifies Coronin-1A deficiency in 3 siblings with immunodeficiency and EBV-associated B-cell lymphoproliferation. J Allergy Clin Immunol. 2013;131(6):1594–603. https://doi.org/10.1016/j.jaci.2013.01.042.
113. Martin E, Palmic N, Sanquer S, Lenoir C, Hauck F, Mongellaz C, et al. CTP synthase 1 deficiency in humans reveals its central role in lymphocyte proliferation. Nature. 2014;510(7504):288–92. https://doi.org/10.1038/nature13386.
114. Verzegnassi F, Valencic E, Kiren V, Giurici N, Bianco AM, Marcuzzi A, et al. The challenge of next generation sequencing in a boy with severe mononucleosis and EBV-related lymphoma. J Pediatr Hematol Oncol. 2018;40(5):e323–6. https://doi.org/10.1097/MPH.0000000000001004.
115. Gineau L, Cognet C, Kara N, Lach FP, Dunne J, Veturi U, et al. Partial MCM4 deficiency in patients with growth retardation, adrenal insufficiency, and natural killer cell deficiency. J Clin Invest. 2012;122(3):821–32. https://doi.org/10.1172/JCI61014.
116. Li FY, Chaigne-Delalande B, Su H, Uzel G, Matthews H, Lenardo MJ. XMEN disease: a new primary immunodeficiency affecting Mg2+ regulation of immunity against Epstein-Barr virus. Blood. 2014;123(14):2148–52. https://doi.org/10.1182/blood-2013-11-538686.
117. Sherkat R, Sabri MR, Dehghan B, Bigdelian H, Reisi N, Afsharmoghadam N, et al. EBV lymphoproliferative-associated disease and primary cardiac T-cell lymphoma in a STK4 deficient patient: a case report. Medicine (Baltimore). 2017;96(48):e8852. https://doi.org/10.1097/MD.0000000000008852.
118. Crequer A, Troeger A, Patin E, Ma CS, Picard C, Pedergnana V, et al. Human RHOH deficiency causes T cell defects and susceptibility to EV-HPV infections. J Clin Invest. 2012;122(9):3239–47. https://doi.org/10.1172/JCI62949.
119. Skibola CF, Berndt SI, Vijai J, Conde L, Wang Z, Yeager M, et al. Genome-wide association study identifies five susceptibility loci for follicular lymphoma outside the HLA region. Am J Hum Genet. 2014;95(4):462–71. https://doi.org/10.1016/j.ajhg.2014.09.004.
120. Conde L, Halperin E, Akers NK, Brown KM, Smedby KE, Rothman N, et al. Genome-wide association study of follicular lymphoma identifies a risk locus at 6p21.32. Nat Genet. 2010;42(8):661–4. https://doi.org/10.1038/ng.626.
121. Smedby KE, Foo JN, Skibola CF, Darabi H, Conde L, Hjalgrim H, et al. GWAS of follicular lymphoma reveals allelic heterogeneity at 6p21.32 and suggests shared genetic susceptibility with diffuse large B-cell lymphoma. PLoS Genet. 2011;7(4):e1001378. https://doi.org/10.1371/journal.pgen.1001378.
122. Cerhan JR, Berndt SI, Vijai J, Ghesquieres H, McKay J, Wang SS, et al. Genome-wide association study identifies multiple susceptibility loci for diffuse large B cell lymphoma. Nat Genet. 2014;46(11):1233–8. https://doi.org/10.1038/ng.3105.
123. Tan DE, Foo JN, Bei JX, Chang J, Peng R, Zheng X, et al. Genome-wide association study of B cell non-Hodgkin lymphoma identifies 3q27 as a susceptibility locus in the Chinese population. Nat Genet. 2013;45(7):804–7. https://doi.org/10.1038/ng.2666.

Part III

Evaluation

Response Assessment in Pediatric Non-Hodgkin Lymphoma

9

Tony H. Truong and Veronique Minard-Colin

Introduction

Through national and international collaboration, the outcome of children with non-Hodgkin lymphoma (NHL) has greatly improved over the past half century [13]. Advances in risk stratification and response assessment have facilitated therapeutic decisions by maximizing therapy in those with the most advanced and resistant diseases, while sparing toxicity and late effects in those with more favorable ones. An ongoing challenge remains the accurate determination of response and remission status, such that subsequent therapy can be individually modified to the patient's disease based on their response to treatment.

Response assessment is the clinical, biopathological, and radiological evaluation of a patient to determine if active residual disease remains either at an interim time point during treatment or at the end of the therapy. The methods used for response assessment are closely linked to those used to assess extent of disease during staging at the time of initial diagnosis. Clinical examination of sites of disease such as residual lymphadenopathy, hepatosplenomegaly, and extra-nodal disease sites are useful at the bedside but lack sensitivity. Follow-up assessments often include repeat staging evaluations, such as imaging and, if applicable, bone marrow aspirates and biopsies and lumbar punctures for cerebrospinal fluid (CSF) involvement. Imaging modalities remain the primary method to assess response status since these tumors are often not evident by other means.

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If residual lesions are identified on follow-up imaging, a major dilemma is whether these represent sites of active residual disease or benign processes such as tumor necrosis and/or inflammatory fibrosis. If there is sufficient concern, the gold standard and often recommended approach is a biopsy.

Different study groups have evaluated the importance of response determination among the various NHL subtypes. In B-NHL, both the Société Française d'Oncologie Pédiatrique (SFOP) LMB and Berlin–Frankfurt–Münster (BFM) studies have demonstrated that residual disease following three cycles of therapy leads to an increased risk of relapse. Intensification of chemotherapy or mega-dose chemotherapy followed by hematopoietic stem cell rescue has resulted in improved outcomes [17, 20]. Among patients with lymphoblastic lymphoma (LBL), the COG A5971 study showed that a radiologic response at two weeks significantly correlated with event-free survival (EFS) and overall survival (OS) [25]. In BFM 90–95 studies, patients with <70% reduction in the size of their mediastinal mass by end of induction day 33 had therapy intensified [19]. In anaplastic large cell lymphoma (ALCL), early response assessment after one course by PCR evaluation may identify patients with a very high risk of treatment failure [10].

Based on a combination of radiographic and histological findings, conventional definitions of response use the designations of complete response (CR), partial response (PR), no response (NR), and progressive disease (PD). CR often refers to the complete absence of any disease detected clinically or radiographically by some pre-specified measure of residual size of the baseline lesion. Partial response encompasses a wide range of definitions between CR and stable disease (SD), also known as "no response." Progressive disease often refers to increasing size of the baseline mass or new sites of disease not present at diagnosis. These definitions are quite varied and often specific to certain diseases or collaborative groups, making comparison across diseases and clinical trials a challenge.

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Response Assessment by Pathology and Molecular Biology

Histological confirmation remains the gold standard to differentiate active residual disease from tumor necrosis or inflammatory scar tissue. In the SFOP LBM89 study, 126 out of 551 patients had radiologic evidence of residual masses. Of these, 113 patients underwent either biopsy or excision of the mass, but only 12 had viable tumor cells (10.6%) [17]. In LMB96, 23 of 657 patients (3.5%) had histologically proven residual disease at remission assessment [4]. For those with active residual disease after three courses of chemotherapy, the success of intensification of therapy suggests that repeat biopsy for these questionable masses may be justified [17].

Sometimes, the decision to resect or biopsy a residual mass may be complicated by several factors, including the patient's underlying condition, the location of the mass, the ease/difficulty of the procedure, and the risks involved. In general, a resection or biopsy should only be attempted if it will change the management approach. Resections are preferred to reduce tumor burden and improve diagnostic yield from pathology, but sometimes may not be feasible or dangerous (e.g., lesions in the gastrointestinal tract). Oopherectomy should be avoided and lesions in the visceral organs need only be sampled with a biopsy. If diagnostic tissue is not obtained, serial biopsies may be attempted if the benefit of knowing the result outweighs the risk involved.

Morphological assessment with the identification of tumor cells is the mainstay for determining residual disease. However, the evaluation of viability of residual cells may be challenging since necrotic tumor cells may still stain positive for B-cell markers such as CD20. The incorporation of highly sensitive measures such as immunophenotyping by flow cytometry, cytogenetics and FISH analysis, and molecular PCR methods have led to further improvement in the detection of minimal disseminated disease (MDD) at diagnosis or minimal residual disease (MRD) during response assessment.

The ability to detect MRD in acute lymphoblastic leukemia (ALL) has greatly informed the risk stratification, prognostication, and treatment for this disease [3, 15]. In NHL, MRD detection has been applied most commonly in lymphoblastic lymphoma using flow cytometry or molecular techniques based on clonal rearrangements of the immunoglobulin or T-cell receptor gene detected at the time of diagnosis [8]. Molecular methods have increased the sensitivity of disease detection with the use of PCR for immunoglobulin gene rearrangements for mature B-NHL (BL and DLBCL) [1]. In the AIEOP LNH-97 study, the Italian group used long-distance PCR for the t(8;14) for MDD detection in patients with Burkitt lymphoma (BL) and diffuse large B-cell lymphoma (DLBCL) [18].

In pediatric ALCL, over 90% of patients will have rearrangement of the *NPM* gene on 5q35 to the anaplastic lymphoma kinase gene *ALK* on 2p23, forming the translocation t(2;5) and the resulting fusion protein *NPM-ALK* [9]. When combined with the detection of antibodies to ALK protein, the BFM and Italian study groups showed that detection of *NPM-ALK* by PCR at diagnosis in blood and/or BM was highly predictive of outcome. High-risk patients with positive MDD and low ALK antibody titer had the lowest progression-free survival (PFS) of 28% compared to the low-risk group (MDD negative and high ALK titer) who had a PFS of 93% [14]. Moreover, detection of persistent *NPM-ALK* by PCR at the end of the first course of chemotherapy (MRD) was highly prognostic and associated with a high risk of relapse [10].

Novel methods such as next-generation sequencing using cell-free circulating tumor DNA are now being developed by many groups with potential future applications to various tissue types including the primary tumor mass, bone marrow, CSF, and/or blood at the time of follow-up [22]. To date, the role of MRD and MDD assessment in response evaluation and risk stratification remains investigational. A thorough review of minimal disseminated disease is presented in the following chapter.

Response Assessment by Imaging

The use of imaging modalities to detect response to treatment remains standard practice in pediatric NHL. The most commonly used modalities are computerized tomography (CT) and magnetic resonance imaging (MRI), each having their unique advantages and applications. CT is the most readily available modality at almost every center, is inexpensive, is fast to perform, and often does not require a general anesthetic in young children. However, exposure to radiation is major concern, especially in patients with predispositions that increase sensitivity to ionizing radiation. For detection and follow-up of pulmonary lesions, CT remains the best modality. For lymphoma patients, MRI is best used for the evaluation of CNS disease in the case of neurologic symptoms or parameningeal mass but is a lengthier procedure which often requires a general anesthetic in young children.

The definitions of imaging-defined response categories, CR, PR, PD, etc., were historically based on the measurement of tumors on cross-sectional imaging. Many measurement methods have been used to assess disease burden and calculate response, leading to variability in practices and difficulty in comparing responses across clinical trials. Currently, the general practice is to identify the most representative lesion and measure it using the longest diameter (LDi) and the

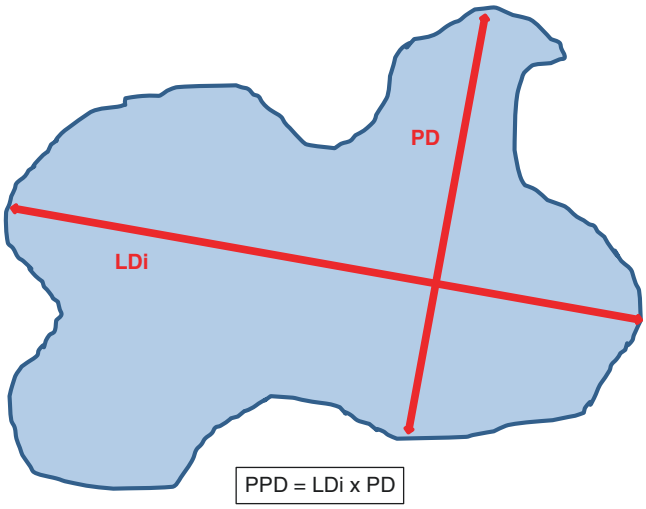

Fig. 9.1 Drawing of cross-sectional image and calculation of the sum of the product of the greatest perpendicular dimensions (SPD)

perpendicular diameter (PD). Multiplying these two diameters generates a product of the perpendicular diameter (PPD) (Fig. 9.1). If more than one lesion exists, as is often the case, then up to six of the most representative (often the largest) lesions are identified as "target" lesions and the sum of products of the largest diameter and the perpendicular diameter for each lesion (SPD) is calculated. The SPD is used as a measure to compare baseline disease burden to that at a later point in time [23]. Other ways to assess response have included measuring the change in transverse diameter or sum of the largest diameters and/or change in three-dimensional volume. Given the variability in response assessment, the need to establish uniform measurement criteria and standard definitions of response was well recognized.

A significant correlation has been observed between the size of the residual lesion and tumor viability. A residual mass measuring ≥5 cm in the largest diameter should be assessed by pathology while a lesion <2 cm is usually reassuring. For intermediate-sized residual lesions (i.e., 2–5 cm), pathological assessment is recommended either by biopsy or complete resection, if possible (Patte, personal communication). In clinical practice, a major challenge is also the assessment of extra-nodal residual disease, which is more frequent in children/adolescents with NHL than in adults [4, 16]. These include more frequent mediastinal residual masses, residual kidney lesions (very common), and residual hepatic and ovarian lesions. Imaging should be considered suspicious if the size of the organ is enlarged (as seen in ovarian masses) or if "stick out masses" are seen (in mediastinal masses). Cases of residual lesions detected on CT/MRI but not apparent on ultrasonography because of necrosis/fibrosis are generally more reassuring (e.g., kidney/hepatic lesions).

FDG-PET

Whole-body ^{18}F-2-fluoro-2-deoxy-D-glucose (^{18}F-FDG)-PET has become an invaluable tool in staging and response assessment of non-Hodgkin lymphoma therapy in adults but its value needs to be further evaluated in pediatric NHL. FDG is a glucose analog that is taken up by cells via glucose transporter proteins. It then undergoes phosphorylation by hexokinase where it does not undergo any further metabolism and is trapped within the cell. FDG uptake is increased in certain malignancies including NHL and Hodgkin lymphoma, and its use is being actively investigated in many other cancer types.

Functional imaging with FDG-PET is often used to assess response evaluation in childhood NHL, but the data to guide such practices are lacking. Limitations to PET include the lack of standardized imaging protocols and variable reporting criteria. This creates uncertainty about the interpretation of PET for use in interim assessment and end of therapy assessment.

PET scans generally have high sensitivity and negative predictive value (NPV) for ruling out disease when negative, but more variable and modest positive predictive value when the result is positive. In a single-center study, PET/CT was compared to conventional imaging and biopsy findings in 18 children with NHL who had biopsy results for evaluation of residual disease. Patients had mature B-NHL and ALCL. A score of 4 or 5 using the London criteria defined PET-positive status. The sensitivity and NPV for PET/CT was 100% but specificity was 60% and PPV was 25%. However, conventional imaging (mostly by CT and MRI) was no better than PET/CT with a sensitivity and NPV of 100% but lower specificity of 20% and PPV of 14% [2].

In a study of 24 pediatric patients with abdominal Burkitt lymphoma, 4 were found to have PET-positive scans at the end of treatment, leading to the need for histological confirmation. Three of these patients had no evidence of malignancy while one patient did, leading to 100% NPV and 25% PPV [21]. Overall, these data indicate that false positive findings by PET/CT are common in children with NHL. A negative scan is generally reassuring as a good indicator of complete response.

The reproducibility of PET interpretation has also been called into question. To address these concerns, standard PET imaging classifications have been adopted, such as the Deauville criteria, a 5-point visual-based criteria, similar to that used in the adult Lugano classification [5, 12]. The most intense FDG site is graded, as per the following Table 9.1.

Use of PET for treatment monitoring during the course of therapy is a common practice, but there is limited evidence to support its use in clinical decision-making. Therefore, this should only be used in a clinical trial or prospective registry study.

Table 9.1 Deauville score in assessing PET response

Score	Description	Interpretation
1	No uptake above background	Complete metabolic response
2	Uptake ≤ mediastinum	
3	Uptake > mediastinum but ≤ liver	1. Probable complete response (CR) 2. May be considered inadequate response to avoid under-treatment in a de-escalation trial
4	Uptake moderately higher than liver	1. Reduced uptake compared to baseline: partial metabolic response 2. No significant change from baseline: no response 3. Increase uptake from baseline: progressive metabolic disease
5	Uptake markedly higher than liver and/or new lesions	

Adapted from: Meignan et al. [12]

The presence of residual PET uptake on an end-of-treatment PET scan, also known as minimal residual uptake (MRU), is a troubling issue and often leads to further investigations to obtain histology or increased frequency of follow-up scans. A single institution study of patients with BL and DLBCL suggests that end of therapy surveillance imaging has low yield for relapse detection but exposure to unnecessary radiation. Only 3 of 44 patients (6.8%) relapsed, none of whom were identified from CT- or PET-based surveillance imaging [11]. In addition to active residual disease, a positive PET may be due to many benign processes including brown fat, rebound thymic hyperplasia, infection, or a benign inflammatory process [24].

Standardization of Response Assessment

Given the need to standardize the measurement and assessment of PET-avid malignancies, an international collaborative effort was initiated by the adult group known as the International Harmonization Project [6] which later produced updated recommendations [7]. The latter guidelines made a formal inclusion of FDG-PET, such that patients with a PET negative residual mass were now considered CR instead of CRu (CR-unconfirmed) on the predecessor guideline. In addition, bone marrow immunohistochemistry and flow cytometry were also incorporated in the response evaluation. A further update known as the Lugano classification emphasized the importance of PET as the gold standard for routine imaging of all FDG-avid, nodal lymphomas and obviated the need for a bone marrow biopsy (BMB) at least in the case of Hodgkin lymphoma when PET-CT is used [5]. This recommendation did not directly translate to NHL as the panel recognized the importance of a BMB in DLBCL when the PET is negative and in cases where knowing BM status would change patient management.

It is well recognized that pediatric NHL differs from adult NHL in several ways: only a few subtypes form the majority of pediatric NHL, most are high-grade lymphomas, and there is a predominance of advanced disease presentations, generally involving the bone marrow and CNS. The need for separate pediatric criteria led to a multidisciplinary collaboration of experts at the third and fourth International Symposia on Childhood, Adolescent, and Young Adult NHL in 2009 and 2012, respectively, resulting in the development of the International Pediatric NHL Response Criteria [23]. The new pediatric criteria incorporate the combination of imaging, tumor histology, bone marrow, and CSF, into five major categories of response (Table 9.2). In addition, the availability of newer techniques based on immunophenotype, cytogenetics, and molecular techniques are used as supporting criteria to more accurately describe the basis for response

Table 9.2 International pediatric NHL response criteria

Criterion	Definition
CR	Disappearance of all disease (three designations) CT or MRI reveals no residual disease or new lesions Resected residual mass that is pathologically (morphologically) negative for disease[a] BM and CSF morphologically free of disease
CRb	Residual mass has no morphologic evidence of disease from limited or core biopsy, with no new lesions by imaging examination[a] BM and CSF morphologically free of disease No new and/or progressive disease elsewhere
Cru	Residual mass is negative by FDG-PET (Deauville score 1, 2, or 3); no new lesions by imaging examination BM and CSF morphologically free of disease[a] No new and/or progressive disease elsewhere
PR	50% decrease in SPD on CT or MRI; FDG-PET may be positive (Deauville score or 4 or 5 with reduced lesional uptake compared with baseline); no new and/or PD; morphologic evidence of disease may be present in BM or CSF if present at diagnosis[a]; however, there should be 50% reduction in percentage of lymphoma cells
MR	Decrease in SPD > 25% but <50% on CT or MRI; no new and/or PD; morphologic evidence of disease may be present in BM or CSF if present at diagnosis[a]; however, there should be 25–50% reduction in percentage of lymphoma cells
NR	For those who do not meet CR, PR, MR, or PD criteria
PD	For those with >25% increase in SPD on CT or MRI, Deauville score 4 or 5 on FDG-PET with increase in lesional uptake from baseline, or development of new morphologic evidence of disease in BM or CSF

Adapted from Sandlund et al. [23].
Abbreviations: *BM* bone marrow, *CR* complete response, *CRb* complete response biopsy negative, *CRu* complete response unconfirmed, *CT* computed tomography, *FDG* 18-F-fluorodeoxyglucose, *MR* minor response, *MRI* magnetic resonance imaging, *NHL* non-Hodgkin lymphoma, *NR* no response, *PD* progressive disease, *PET* positron emission tomography, *PR* partial response; *SPD* sum of product of greatest perpendicular diameters
[a]Detection of disease with more sensitive techniques described as supporting data (Table 9.3)

Table 9.3 Supporting international pediatric NHL response criteria data

Supporting criterion	Description
BM involvement	Currently defined by morphologic evidence of lymphoma cells; this applies to any histologic subtype; type and degree of BM involvement should be specified[a]
BMm	BM positive by morphology (specify percentage of lymphoma cells)
BMi	BM positive by immunophenotypic methods (histochemical or flow cytometric analysis; specify percentage of lymphoma cells)
BMc	BM positive by cytogenetic or FISH analysis (specify percentage of lymphoma cells)
BMmol	BM positive by molecular techniques
CNS involvement	CSF positivity is based on morphologic evidence of lymphoma cells; CSF should be considered positive when any number of blasts is detected; CSF may be unknown; as with BM, type of CSF involvement should be described whenever possible
CSFm	CSF positive by morphology (specify No. of blasts/μL)
CSFi	CSF positive by immunophenotype methods (histochemical or flow cytometric analysis; specify percentage of lymphoma cells)
CSFc	CSF positive by cytogenetic or FISH analysis (specify percentage of lymphoma cells)
CSFmol	CSF positive by molecular techniques
Residual mass (RM)	
RMm	Tumor detected by standard morphologic evaluation
RMi	Tumor detected by immunophenotypic methods (immunohistochemical or flow cytometric analysis)
RMc	Tumor detected by cytogenetic or FISH analysis
RMmol	Tumor detected by molecular techniques

Adapted from Sandlund et al. [23]
Abbreviations: *BM* bone marrow, *FISH* fluorescent in situ hybridization, *NHL* non-Hodgkin lymphoma, *PB* peripheral blood, *RM* residual mass
[a]Same approach should be used for PB involvement (i.e. PBm, PBi, PBc, PBmol)

determination (Table 9.3). The inclusion of supporting response data, though not directly incorporated into the response evaluation, is forward thinking as these measures will in no doubt be integrated in future criteria.

A standardized response evaluation schema has many benefits but requires widespread acceptance and incorporation into clinical trials. It will allow for comparison of treatment efficacy across multiple regimens while facilitating clinical decision-making for the individual patient.

References

1. Agsalda M, Kusao I, Troelstrup D, Shiramizu B. Screening for residual disease in pediatric burkitt lymphoma using consensus primer pools. Adv Hematol. 2009;2009:412163.
2. Bhojwani D, McCarville MB, Choi JK, Sawyer J, Metzger ML, Inaba H, Davidoff AM, Gold R, Shulkin BL, Sandlund JT. The role of FDG-PET/CT in the evaluation of residual disease in paediatric non-Hodgkin lymphoma. Br J Haematol. 2015;168(6):845–53.
3. Borowitz MJ, Devidas M, Hunger SP, Bowman WP, Carroll AJ, Carroll WL, Linda S, Martin PL, Pullen DJ, Viswanatha D, Willman CL, Winick N, Camitta BM, Children's Oncology Group. Clinical significance of minimal residual disease in childhood acute lymphoblastic leukemia and its relationship to other prognostic factors: a Children's Oncology Group study. Blood. 2008;111(12):5477–85.
4. Cairo M, Gerrard M, Sposto R, et al. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. Blood. 2007;109(7):2736–43.
5. Cheson BD, Fisher RI, Barrington SF, Cavalli F, Schwartz LH, Zucca E, Lister TA, Alliance, Australasian Leukaemia and Lymphoma Group, Eastern Cooperative Oncology Group, European Mantle Cell Lymphoma Consortium, Italian Lymphoma Foundation, European Organisation for Research, Treatment of Cancer/Dutch Hemato-Oncology Group, Grupo Espanol de Medula Osea, German High-Grade Lymphoma Study Group, German Hodgkin's Study Group, Japanese Lymphorra Study Group, Lymphoma Study Association, NCIC Clinical Trials Group, Nordic Lymphoma Study Group, Southwest Oncology Group, United Kingdom National Cancer Research Institute. Recommendations for initial evaluation, staging, and response assessment of Hodgkin and non-Hodgkin lymphoma: the Lugano classification. J Clin Oncol. 2014;32(27):3059–68.
6. Cheson BD, Horning SJ, Coiffier B, Shipp MA, Fisher RI, Connors JM, Lister TA, Vose J, Grillo-Lopez A, Hagenbeek A, Cabanillas F, Klippensten D, Hiddemann W, Castellino R, Harris NL, Armitage JO, Carter W, Hoppe R, Canellos GP. Report of an international workshop to standardize response criteria for non-Hodgkin's lymphomas. NCI Sponsored International Working Group. J Clin Oncol. 1999;17(4):1244–53.
7. Cheson BD, Pfistner B, Juweid ME, Gascoyne RD, Specht L, Horning SJ, Coiffier B, Fisher RI, Hagenbeek A, Zucca E, Rosen ST, Stroobants S, Lister TA, Hoppe RT, Dreyling M, Tobinai K, Vose JM, Connors JM, Federico M, Diehl V, International Harmonization Project on Lymphoma. Revised response criteria for malignant lymphoma. J Clin Oncol. 2007;25(5):579–86.
8. Coustan-Smith E, Sandlund JT, Perkins SL, Chen H, Chang M, Abromowitch M, Campana D. Minimal disseminated disease in childhood T-cell lymphoblastic lymphoma: a report from the children's oncology group. J Clin Oncol. 2009;27(21):3533–9.
9. Damm-Welk C, Klapper W, Oschlies I, Gesk S, Rottgers S, Bradtke J, Siebert R, Reiter A, Woessmann W. Distribution of NPM1-ALK and X-ALK fusion transcripts in paediatric anaplastic large cell lymphoma: a molecular-histological correlation. Br J Haematol. 2009;146(3):306–9.
10. Damm-Welk C, Mussolin L, Zimmermann M, Pillon M, Klapper W, Oschlies I, d'Amore ES, Reiter A, Woessmann W, Rosolen A. Early assessment of minimal residual disease identifies patients at very high relapse risk in NPM-ALK-positive anaplastic large-cell lymphoma. Blood. 2014;123(3):334–7.
11. Eissa HM, Allen CE, Kamdar K, Simko S, Goradia P, Dreyer Z, Steuber P, McClain KL, Guillerman RP, Bollard CM. Pediatric Burkitt's lymphoma and diffuse B-cell lymphoma: are surveillance scans required? Pediatr Hematol Oncol. 2014;31(3):253–7.
12. Meignan M, Gallamini A, Meignan M, Gallamini A, Haioun C. Report on the First International Workshop on Interim-PET-Scan in Lymphoma. Leuk Lymphoma. 2009;50(8):1257–60.
13. Minard-Colin V, Brugieres L, Reiter A, Cairo MS, Gross TG, Woessmann W, Burkhardt B, Sandlund JT, Williams D, Pillon M, Horibe K, Auperin A, Le Deley MC, Zimmerman M, Perkins SL, Raphael M, Lamant L, Klapper W, Mussolin L, Poirel HA,

Macintyre E, Damm-Welk C, Rosolen A, Patte C. Non-Hodgkin lymphoma in children and adolescents: progress through effective collaboration, current knowledge, and challenges ahead. J Clin Oncol. 2015;33(27):2963–74.
14. Mussolin L, Damm-Welk C, Pillon M, Zimmermann M, Franceschetto G, Pulford K, Reiter A, Rosolen A, Woessmann W. Use of minimal disseminated disease and immunity to NPM-ALK antigen to stratify ALK-positive ALCL patients with different prognosis. Leukemia. 2013;27(2):416–22.
15. Paganin M, Fabbri G, Conter V, Barisone E, Polato K, Cazzaniga G, Giraldi E, Fagioli F, Arico M, Valsecchi MG, Basso G. Postinduction minimal residual disease monitoring by polymerase chain reaction in children with acute lymphoblastic leukemia. J Clin Oncol. 2014;32(31):3553–8.
16. Patte C, Auperin A, Gerrard M, Michon J, Pinkerton R, Sposto R, Weston C, Raphael M, Perkins SL, McCarthy K, Cairo MS, FAB/LMB96 International Study Committee. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. Blood. 2007;109(7):2773–80.
17. Patte C, Auperin A, Michon J, Behrendt H, Leverger G, Frappaz D, Lutz P, Coze C, Perel Y, Raphael M, Terrier-Lacombe MJ. The Societe Francaise d'Oncologie Pediatrique LMB89 protocol: highly effective multiagent chemotherapy tailored to the tumor burden and initial response in 561 unselected children with B-cell lymphomas and L3 leukemia. Blood. 2001;97(11):3370–9.
18. Pillon M, Mussolin L, Carraro E, Conter V, Arico M, Vinti L, Garaventa A, Piglione M, Buffardi S, Sala A, Santoro N, Lo Nigro L, Mura R, Tondo A, Casale F, Farruggia P, Pierani P, Cesaro S, d'Amore ES, Basso G. Detection of prognostic factors in children and adolescents with Burkitt and Diffuse Large B-Cell Lymphoma treated with the AIEOP LNH-97 protocol. Br J Haematol. 2016;175(3):467–75.
19. Reiter A, Schrappe M, Parwaresch R, Henze G, Muller-Weihrich S, Sauter S, Sykora KW, Ludwig WD, Gadner H, Riehm H. Non-Hodgkin's lymphomas of childhood and adolescence: results of a treatment stratified for biologic subtypes and stage – a report of the Berlin-Frankfurt-Munster Group. J Clin Oncol. 1995;13(2):359–72.
20. Reiter A, Schrappe M, Tiemann M, Ludwig WD, Yakisan E, Zimmermann M, Mann G, Chott A, Ebell W, Klingebiel T, Graf N, Kremens B, Muller-Weihrich S, Pluss HJ, Zintl F, Henze G, Riehm H. Improved treatment results in childhood B-cell neoplasms with tailored intensification of therapy: a report of the Berlin-Frankfurt-Munster Group Trial NHL-BFM 90. Blood. 1999;94(10):3294–306.
21. Riad R, Omar W, Sidhom I, Zamzam M, Zaky I, Hafez M, Abdel-Dayem HM. False-positive F-18 FDG uptake in PET/CT studies in pediatric patients with abdominal Burkitt's lymphoma. Nucl Med Commun. 2010;31(3):232–8.
22. Roschewski M, Dunleavy K, Pittaluga S, Moorhead M, Pepin F, Kong K, Shovlin M, Jaffe ES, Staudt LM, Lai C, Steinberg SM, Chen CC, Zheng J, Willis TD, Faham M, Wilson WH. Circulating tumour DNA and CT monitoring in patients with untreated diffuse large B-cell lymphoma: a correlative biomarker study. Lancet Oncol. 2015;16(5):541–9.
23. Sandlund JT, Guillerman RP, Perkins SL, Pinkerton CR, Rosolen A, Patte C, Reiter A, Cairo MS. International Pediatric non-Hodgkin lymphoma response criteria. J Clin Oncol. 2015;33(18):2106–11.
24. Shammas A, Lim R, Charron M. Pediatric FDG PET/CT: physiologic uptake, normal variants, and benign conditions. Radiographics. 2009;29(5):1467–86.
25. Termuhlen AM, Smith LM, Perkins SL, Lones M, Finlay JL, Weinstein H, Gross TG, Abromowitch M. Disseminated lymphoblastic lymphoma in children and adolescents: results of the COG A5971 trial: a report from the Children's Oncology Group. Br J Haematol. 2013;162(6):792–801.

Minimal Disseminated and Minimal Residual Disease in Pediatric Non-Hodgkin Lymphoma

10

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Introduction

Minimal residual disease (MRD) has been established as the most powerful independent prognostic parameter for children and adults with acute lymphoblastic leukemia (ALL). It has been introduced into standard clinical practice for treatment stratification for ALL both during initial therapy and in relapse worldwide [1–7]. Different techniques with international quality control for sensitivity, specificity, and the quantitative range are in place [7]. Patient-specific immunoglobulin or T-cell receptor rearrangements measured by PCR and aberrant immunophenotypes detected by flow cytometry are the methods most widely standardized. Both methods have their specific application time points, advantages, and disadvantages.

MRD using the standard methods has also been established as prognostic factor for disease monitoring for some subtypes of Non-Hodgkin lymphoma in adults, especially indolent lymphomas like chronic lymphatic leukemia, mantle cell lymphoma, or follicular lymphoma. In addition, newer next-generation sequencing-based methods targeting both patient-specific and disease-specific markers have been started to be studied in adults with diffuse large B-cell lymphoma, mantle cell lymphoma, and follicular lymphoma [8, 9].

The knowledge from ALL and adult lymphoma can, however, not easily be transferred to Non-Hodgkin lymphoma in children. Comprising a heterogeneous group of different diseases, few patients are available in each biological subgroup to evaluate the possible prognostic value of MRD. The different NHL entities need different approaches and techniques to measure minimal disease. As a major difference to leukemia, initial tumor material often is limited in NHL and needs to be used for assurance of an accurate histopathological diagnosis. In most instances, there is no fresh tumor material available for establishment of MRD markers. Bone marrow (BM) is involved in only a part of the patients, so that it can only be used to establish MRD markers and measurement of MRD in Burkitt leukemia (B-AL), stage IV lymphoblastic lymphoma (LBL), or some peripheral T-cell lymphomas (PTCL). One prerequisite to study MRD in NHL in most children, therefore, is to determine minimal disseminated disease (MDD) in the BM and/or blood at diagnosis. Only among those children with detectable cytological or minimal disease in BM at diagnosis the prognostic meaning of MRD can be studied. The associated question arising is whether the detection of disseminated lymphoma cells at diagnosis may already be of prognostic value comparable to micro metastases in pediatric solid tumors or adults with DLBCL [10–14].

Further special features of pediatric NHL need to be considered when studying MRD. One is the high probability of event-free survival (EFS) with current chemotherapeutic strategies for the most frequent subtypes which asks for prognostic factors with a very high predictive value [15]. In addition, relapses occur very early, usually within a few weeks to months after the end of therapy or even as progression so that MRD—with the intention to use it for stratification of patients to different treatment—needs to be measured at very early time points during therapy which leaves a short time for establishment of MRD markers.

In this chapter, we are going to describe the techniques currently used for minimal disease detection in the different lymphoma subtypes. We then summarize the available clini-

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cal data on the possible prognostic meaning of MDD and MRD in the larger pediatric NHL subtypes. In addition to the use as prognostic marker, we discuss possible other applications of MRD, like longitudinal disease monitoring in individual patients or status before consolidation by stem cell transplantation in relapse.

Techniques Used for MDD/MRD Detection in Lymphoma Subtypes

Four basic methods currently are available for the detection of rare lymphoblastic leukemia and lymphoma cells. Specific rearrangements of the T-cell receptor or immunoglobulin genes can be used as patient-specific markers which need initial tumor material for establishment. This method has almost no risk of cross-contamination and strict quality control has been established in leukemia trials. However, this method is both labor consuming and expensive. Secondly, the expression of a leukemia/lymphoma-specific immunophenotype allows for quantification of few tumor cells by flow cytometry. Initial tumor material is not necessary for every lymphoma type using this technique. It is less expensive compared to the first method, but the sensitivity is somewhat lower using the current approach. As a third method, lymphoma-specific fusion genes can be measured either on the DNA or RNA level if translated into a fusion protein (type 1 fusions), or on the DNA level only in lymphomas without expression of fusion transcripts (type 2 fusions). These approaches require initial tumor material in some but not all cases. Measurement on the RNA level has a high sensitivity, however, bears the risk of cross-contamination with false-positive results. Lastly, next-generation sequencing of initial lymphoma material may detect aberrations that can be used as a tumor- and patient-specific MRD marker.

These techniques have different sensitivities and specificities that are not only dependent on the method but also vary according to the lymphoma subtype.

Genetically, Burkitt lymphoma (BL) and mature B-cell leukemia (B-AL) are characterized by the presence of chromosomal translocations involving the *C-MYC* gene on chromosome 8 and the immunoglobulin heavy- or light-chain genes on chromosome 14, 22 or 2 [16]. The most common translocation, accounting for about 75% of all cases, is the t(8;14)(q24;q32) which juxtaposes the *C-MYC* gene to the immunoglobulin heavy chain (*IGH@*) locus on chromosome 14 in divergent orientation. The breakpoint locations vary considerably depending on the geographic distribution of the disease and likely on the presence or absence of Epstein-Barr virus (EBV) genome. On chromosome 8, breakpoints are usually located upstream of the *C-MYC* gene in the endemic (African) BL and within exon 1 or intron 1 in the sporadic (Caucasian) BL. The breakpoints in the *IGH@* locus are distributed over a region of at least 100 kb, but they are preferentially located in the *IGH@* joining region (JH) or in the switch regions in the endemic and the sporadic BL, respectively [17]. The *MYC-IGH@* rearrangement is detectable only at the genomic DNA level because no fusion transcript originates from this genetic alteration. In the great majority of sporadic BL, the t(8;14) translocation can be detected by a long-distance polymerase chain reaction (LD-PCR) assay which relies on the use of one primer specific for *C-MYC* exon 2 combined, in different reactions, with four primers for the *IGH@* locus [17, 18]. The LD-PCR assay is useful not only for the molecular characterization of the primary disease, but it can also be applied to detect MRD because the LD-PCR product detecting the *MYC-IGH@* rearrangements is specific for each individual tumor. The assay has a sensitivity approaching $10^{-3}/10^{-4}$ both *in vitro* and *in vivo* [19]. Notably, it is not possible to study endemic BL by this technique due to the wide chromosomal region involved.

In addition to the t(8;14), another marker for MDD/MRD detection in BL and mature B-AL is represented by clonal rearrangement of immunoglobulin (IG) genes, which consist of variable, constant, and junctional regions; the latter are unique to the lymphatic/leukemic clone. Investigators screen for these rearrangements at diagnosis in each case, and after identifying the lymphoma-specific junctional sequences, design junctional region-specific oligonucleotides, also called allele-specific oligonucleotides, which will be used as primers for the PCR assay to MDD/MRD during treatment [19]. These assays were performed according to the guidelines of the European Study Group on MRD detection in ALL (ESG-MRD-ALL) [20]. In most of the targets, a sensitivity of at least 10^{-4} was achieved and at least one sensitive target was available. Because access to original tumor is usually required to design the patient-specific primers, the feasibility of using IgV(H) primer pools to detect disease in clinical specimens was assessed. IgV(H) primer pools from IgV(H1) to IgV(H7) regions were tested to detect MDD/MRD, thus eliminating the need for original tumor. Until now only small cohorts of patients have been analyzed [21, 22]. MDD/MRD detection by IgV(H) primer pools needs further investigation to establish the potential role as a real-time assessment tool to monitor pediatric BL and possibly other B-cell Non-Hodgkin lymphoma.

In almost 90% of cases, ALK-positive anaplastic large cell lymphoma (ALCL) is associated with the tumor-specific t(2;5)(p23;q35) chromosomal translocation, which gives rise to the fusion gene *NPM-ALK* [23–25]. In the last decades, several variant rearrangements at the 2p23 *ALK* locus other than t(2;5) have been identified in ALCL, in which partner genes other than *NPM* are involved [26–31]. The fusion gene transcript can be readily and sensitively detected by PCR [32–34]. In experiments testing mixtures of the translocation-

positive Karpas-299 ALCL cell line with the translocation-negative Jurkat T-cell line a sensitivity of 10^{-6} for this RT-PCR assay could be demonstrated [34]. A quantitative RQ-PCR for *NPM-ALK* was developed according to the standardized protocol of the EAC (Europe Against Cancer) program for TaqMan-based RQ-PCR [32]. The *NPM-ALK* fusion transcript copy number is normalized to the copy numbers of *ABL* that was selected as the control gene to compensate for variations in RNA integrity. The normalized copy number is expressed as copy numbers of *NPM-ALK* per 10^4 copies *ABL*. In serial cell dilution experiments with Karpas 299 cells in NPM-ALK-negative DG75 cells, the method detected fewer than 10 NPM-ALK-positive cells/10^6 control cells, indicating a sensitivity at least 10^{-5} at the cellular level. However, the method is difficult to transfer to other laboratories and an international cut-off value would need to be defined. Quantification at the lowest end of the standard curve is necessary which hampers inter-laboratory comparison. For the international biological NHL Study Group, establishment of a quality controlled quantitative real-time PCR for *NPM-ALK* is one of our major efforts at present. In terms of feasibility, qualitative RT-PCR currently is the 'gold standard' because it is easily reproducible and less expensive. In addition, a flow cytometric (FCM) assay for quantification of ALK- and CD30-positive ALCL cells has been developed that showed a high specificity and a sensitivity of 10^{-4} to 10^{-5} [35]. The results of the FCM assay and quantitative PCR for *NPM-ALK* correlated but the sensitivity of the PCR exceeded that of the FCM by at least one log. Quantitative PCR was more time-consuming and expensive than FCM. The FCM method needs to be tested in a larger cohort of patients to determine whether it has sufficient sensitivity to be used as a substitute for quantitative PCR.

Lymphoblastic T-cell lymphoma (T-LBL) and T-acute lymphoblastic leukemia (T-ALL) are often considered to be part of a spectrum of a single disease [36]. The malignant cells in T-ALL and T-LBL are morphologically indistinguishable, and immunophenotype as well as genetic abnormalities of the cells are similar. The sensitive and specific methodologies used for MRD monitoring in T-ALL, such as PCR amplification of specific genetic abnormalities and clonal *IG/TCR* gene rearrangements, can be used to detect submicroscopic disseminated disease also in patients with T-LBL. Like *IG* genes, *T-cell receptor (TCR)* genes consist of variable, constant, and junctional regions. After screening for these rearrangements at diagnosis in each case with identification of lymphoma-specific junctional sequences, junctional region-specific oligonucleotides need to be designed which will be used as primers for the PCR assay for MRD evaluation. These methods require the identification of a clone-specific molecular target in each patient through the initial analysis of tumor cells [37]. To bypass this problem multiparametric FCM could be used for MDD/MRD analysis [38, 39] that can detect one T-LBL cell among 10,000 normal cells. The immunophenotype of LBL tumor cells at diagnosis is classified according to the European Group for the Immunological Characterization of Leukemias (EGIL) classification [40]. Firstly, as for the blasts in ALL, lymphoma cells are identified using an immunological gate, based on a lineage-defined antigen (CD7 or CD19) expression associated with light scatter (Side Scatter or SSC). LBL cells are recognized for co-expression of cell markers not found in normal lymphocytes or typical of lymphocytes normally confined to the thymus. PCR assay and FCM results were compared for T-ALL and the investigators obtained a very good correlation between these two methods [39].

Of note, standardized procedures are warranted to apply MDD/MRD evaluation within multicenter therapeutic trials or to introduce new more sensitive molecular assay (e.g. next-generation sequencing analysis).

In 2012, five university laboratories from about 15 European and extra-European countries designed a joint project to develop standardization and quality control analysis for MDD/MRD studies in pediatric lymphoma patients. For example, for ALCL, a common set of primers was established, and standard operating procedures were implemented; quality control is performed regularly.

Clinical Impact of MDD and MRD in Lymphoblastic Lymphoma

FCM and RQ-PCR for *TCR* gene rearrangements have been shown to identify minimal disseminated disease in T-LBL with a sensitivity of 0.01%. A retrospective study of 17 stage III T-LBL patients, analyzed at diagnosis, showed MRD positivity in the BM of 88% and 80% of the cases when evaluated by FCM and RQ-PCR, respectively [37]. The concordance between the two methods was 67% at the highest sensitivity level, with 5/17 cases with discordant results. The first data on the prognostic impact of minimal disease at diagnosis, evaluated by FCM, was reported by Coustan-Smith et al. in 99 pediatric T-LBL patients [38]. Submicroscopic disease was detected in 72% of BM studied (71/99), with T-LBL blasts ranging from 0.01% to about 32%. Detection of T-LBL cells in BM was more frequent among younger patients (<10 years; $p = 0.046$) and among patients with lower LDH ($p = 0.0027$), but was not significantly related to other features, such as gender, CNS involvement, or stage. In 90/99 patients a PB sample at diagnosis was also studied. Every patient with detectable disease in the BM had also detectable disease in blood ($r = 0.86$, $p < 0.0001$). In eight T-LBL patients, cells were found in PB but not in BM, suggesting that examination of PB might allow a more sensitive detection of disseminated disease. Two-year EFS was 52% for patients with >5% T-LBL cells

by FCM in BM versus 89% for the remaining patients ($p = 0.009$). Using a cut-off level of 1%, the 2-year EFS was 68% for patients with higher levels of disease dissemination versus 91% for those with lower levels. The 2-year EFS of patients with 0.001–0.1% did not differ from the EFS of patients with negative MDD. In multivariate analysis, detection of T-LBL cells by FCM or stage IV did not retain an independent prognostic value.

The AIEOP study group examined BM and PB samples from a series of 65 children affected by T- (52) and B-lineage (13) LBL using FCM. MDD was detected in 49% (32/65) of BM samples, whereas only 21% (14/65) were positive by standard morphological evaluation. Findings from FCM analyses of paired BM and PB samples were highly concordant [39]. Using an MDD cut-off level of 3% by FCM (75th percentile), 5-year event-free survival (EFS) was 60 ± 22% for patients with MDD >3% LBL cells versus 83 ± 6% for the remaining patients ($p = 0.04$). No statistically significant difference in EFS was observed among LBL patients considering the following parameters at diagnosis: sex, median age, B and T immunophenotype, mediastinal involvement, serum lactate dehydrogenase, morphological infiltration of BM, CNS involvement, stage of disease, and treatment protocol. These results cumulatively indicate that MDD assessment in LBL by FCM is at least 100-fold more sensitive compared to cytology. FCM can be performed faster and is less expensive compared to a RQ-PCR assay and does not necessarily need initial tumor tissue. However, in the studies reported so far, the MDD cutoff was at the level of 3–5%, whereas cytological BM infiltration has not been shown to have a prognostic meaning in several trials [41, 42]. Therefore, whether MDD could be used for patient stratification in clinical studies needs to be investigated in a larger patient cohort together with other risk factors like the molecular profile [43–45].

Clinical Impact of MDD and MRD in Burkitt Lymphoma and Leukemia

The Italian Association of Pediatric Hematology-Oncology Group (AIEOP) used a LD-PCR-based assay for the *MYC-IGH* fusion to prospectively study 78 BL patients [46, 47]. A specific *MYC-IGH*@ PCR marker could be established in 52 patients. MDD was positive in 36% of the patients. The study of a larger cohort of patients (134 BL specimens; 84 of them had both a *MYC-IGH* breakpoint and BM available) confirmed that more than 30% of patients were MDD positive whereas only 12 patients (14%) were positive by morphology. Most of the patients with molecular detection of disease in the bone marrow at diagnosis (22/26, 85%) belonged to the R4 Risk Group according to the BFM definition (stage III or stage IV according to St. Jude staging classification and LDH >1000 U/l). The 3-year progression-free survival (PFS) was 68 ± 10% for MRD-positive R4 patients compared with 93 ± 5% for MRD-negative R4 patients ($p = 0.03$) (Fig. 10.1) [47], whereas there was no significant difference in PFS between children with morphological BM involvement at diagnosis and those who had negative BM (PFS 67 ± 14% vs. 87 ± 6%, respectively, $p = 0.12$). By multivariate analysis only MDD was predictive of higher risk of failure among R4 patients (hazard ratio, 4.7; $P = 0.04$) [47]. Busch et al. reported on 18 patients with t(8;14)-positive BL without cytological BM infiltration using a PCR assay that combined the LD-PCR with a nested PCR [48]. The sensitivity range of this approach was 10^{-3}–10^{-5}. In eight patients the specific *C-MYC-IGH*@ rearrangement was detected either in BM (4/18) or in PB samples (6/15). In two of the four BM-positive cases, the blood was also positive; in the remaining two

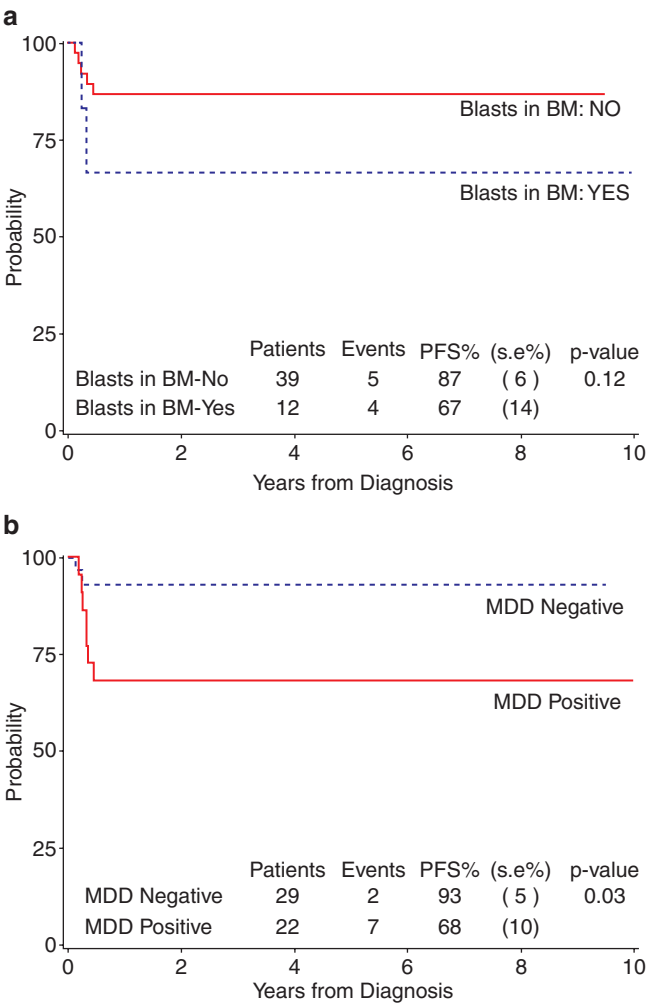

Fig. 10.1 Progression-free survival (PFS) analysis in patients with Burkitt lymphoma at high risk (BFM group R4) according to (**a**) morphological bone marrow status at diagnosis and (**b**) minimal disseminated disease (MDD) status in BM at diagnosis. (From Mussolin et al. [47], used with the permission from the American Society of Clinical Oncology)

cases, no blood was available. In contrast, in three of the six PB-positive cases the BM was negative, thus suggesting that analysis of both BM and blood might be relevant for MRD monitoring in BL patients.

The application of the LD-PCR assay has some limitations. The sensitivity limit does not exceed 10^{-4} in most patients [47]. It is applicable in about 80% of patients with t(8;14) but not for patients with t(2;8) or t(8;22). The latter obstacle can be at least in part overcome using clone-specific immunoglobulin gene rearrangements as MDD target [19]. Overall, 36 B-AL and 19 BL cases were analyzed by this method in an AIEOP study. In 88% of the cases, a sensitivity of at least 10^{-4} was achieved. Molecular BM involvement at diagnosis was detected in 6/19 BLs using this assay. MRD positivity persisted during chemotherapy in 6/36 children affected by B-AL. In most patients, LD-PCR and *IG* gene rearrangement-based methods detected MRD with similar results. Thus, both methods can be used for MDD/MRD analysis in mature B-AL and BL patients and each has advantages and disadvantages. The LD-PCR method is fast and relatively inexpensive, but the t(8;14) translocation cannot be detected in all the cases. *IG* rearrangements are near-universal targets for MRD studies in B-cell malignancies and provide accurate quantification of MRD, but their detection is laborious, can be expensive, and requires initial tumor material [49].

Agsalda et al. studied MRD on follow-up specimens from B-NHL cases, relying on monoclonality defined by *IGVH* gene rearrangements, rather than on the identification of lymphoma clone-specific *IG* rearrangements that needs the availability of primary tumor tissue [21]. They hypothesized that MRD could be screened in specimens using primer pools made up of IGV_H oligomers from respective V_{H1} to V_{H7} families. The study was limited to 14 patients, but their findings support the feasibility of this approach to analyze MRD because a previous study using patient-specific primers on the same cohort of children gave concordant results.

MDD/MRD was assessed by real-time polymerase chain reaction at the end of induction and consolidation with patient-specific primers from 10 children/adolescents with B-AL ± central nervous system disease who were treated with French-British-American/Lymphome Malins de Burkitt (FAB/LMB) 96 C1 therapy augmented with rituximab. MRD after induction and consolidation was positive in 7/10 and 5/7 patients, respectively. However, there was no relapse in this small cohort and subsequent therapy appeared to eliminate MRD [22].

Despite the large number of MRD studies conducted in childhood ALL, there is very limited information on the prognostic relevance of MRD in B-AL, possibly due to the rarity of B-AL as compared with B-cell precursor ALL and to the difficulty to conduct prospective studies on a significant number of cases. The AIEOP group studied 68 BM at diagnosis from children affected by B-AL for the presence of t(8;14) by LD-PCR [50]. Sixty-nine percent were positive. MRD response was determined in 39 patients. The 3-year relapse-free survival (RFS) was 38 ± 7% for patients who were MRD-positive after the first chemotherapy cycle compared with 84 ± 7% for MRD-negative patients ($p = 0.0005$). An extension of this study with 128 patients confirmed these results. MRD positivity after the first course of chemotherapy remained the only unfavorable prognostic factor for progression-free survival in multivariate analysis [51].

In conclusion, the analysis of MDD and MRD in BL and B-AL may contribute to design better risk-adapted therapies for high-risk subgroups of BL and B-AL patients.

Clinical Experiences of MDD/MRD in ALK-Positive Anaplastic Large Cell Lymphoma (ALCL)

MDD and MRD in ALK-positive ALCL have been studied using the sensitive and specific detection of *NPM-ALK* transcripts by RT-PCR in BM and blood cells. Initial fresh frozen tumor material often is not available to screen for *NPM-ALK* transcripts. However, the presence of NPM-ALK can reliably be determined using immune histology or FISH using dual-color probes. NPM-ALK is expressed in the cytoplasm as well as the nucleus, whereas all variant ALK (X-ALK) fusion proteins described so far are expressed exclusively in the cytoplasm or at the cell membrane (MSN-ALK) [23, 25, 52, 53]. Since more than 80% of pediatric ALK-positive ALCL are positive for *NPM-ALK*, a nuclear and cytoplasmic ALK stating or a *t(2;5)*-specific positive dual-color fluorescence in situ hybridisation ascertain NPM-ALK positivity thereby allowing for the investigation of MDD without the need of fresh tumor [23, 25].

The first analysis of MDD in ALCL using a qualitative *NPM-ALK* RT-PCR with a sensitivity of 10^{-6} was reported by the AIEOP group in 2005 [34]. 25/41 (61%) of analyzed patients had MDD detectable in the BM at the time point of diagnosis, whereas only 15% of the corresponding BM smears were positive by microscopy. Outcome analysis was performed in 35 of the analyzed patients. PFS at 5 years was 41 ± 11% for the MDD-positive patients compared to 100% for the MDD-negative patients ($p = 0.001$) [34]. The prognostic meaning of MDD evaluation in the BM was confirmed by the BFM group in an analysis of 80 *NPM-ALK*-positive ALCL patients. Forty-eight percent of these patients were MDD positive in the BM. The cumulative incidence of relapses (CI-R) was 50 ± 10% for MDD-positive patients compared to a 15 ± 7% for MDD-negative patients ($p < 0.001$). The detection of MDD in the BM was associated with higher stage, visceral or mediastinal organ involvement, and a not common histological subtype [32].

MDD could be compared between bone marrow and blood in 52 of those patients. The results correlated well with concordant result obtained in both media in 45/52 patients. Six of the seven remaining patients were positive in blood but not in bone marrow, one patient was positive in bone marrow but not blood [32]. A collaborative study of the AIOP and BFM group evaluated MDD by qualitative PCR for *NPM-ALK* in either blood or BM in 180 uniformly treated NPM-ALK-positive ALCL patients [54, 55]. Fifty-seven percent of the analyzed patients were MDD positive. The association of a positive MDD with not common histomorphology and clinical risk factors could be confirmed. The prognostic meaning of MDD analyzed by RT-PCR for the fusion gene transcript could be validated with an EFS at 5 years of 51 ± 5% for MDD-positive and 83 ± 5% for MDD-negative patients ($p < 0.001$) (Fig. 10.2). In multivariate analysis, only MDD, histological subtype, and ALK-antibody titers retained an independent prognostic value. These studies cumulatively established MDD as the most powerful and independent prognostic factor in ALK-positive ALCL.

To enable stratification of patients according to MDD as measured by qualitative RT-PCR for NPM-ALK, five reference laboratories for MDD in the EICNHL started a quality control system. Standard operating procedures (SOPs) were established and common sets of reagents, PCR primers, and controls were introduced. Blinded test samples are distributed centrally twice yearly. Until 2014, five *NPM-ALK* RT-PCR quality control rounds were performed. All participating laboratories detected *NPM-ALK* transcripts with sensitivities between 10^{-5} and 10^{-6} by using the SOP-RT-PCR protocol [56]. Inter-laboratory quality control as an indispensable prerequisite for patient stratification according to MDD in international studies could be successfully established.

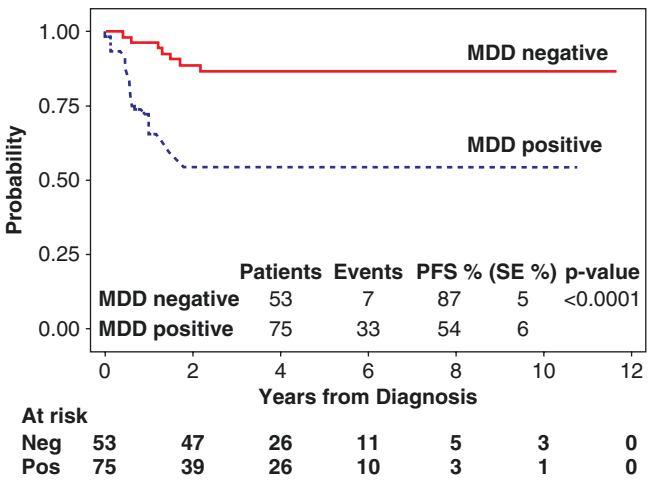

Fig. 10.2 5-year progression-free survival of patients with NPM-ALK-positive anaplastic large cell lymphoma ALCL according to minimal disseminated disease (MDD) in bone marrow or blood measured by qualitative PCR for *NPM-ALK*. (From Mussolin et al. [55], with permission)

The level of MRD predicts the risk of relapse in patients with ALL. With the aim to investigate whether a group of patients with a so-called "high" MDD load might have a very high risk of relapse, the BFM group quantified *NPM-ALK* transcripts using an *NPM-ALK*-specific RQ-PCR assay with *ABL* as control gene [32]. Quantification of transcripts was performed in BM samples of 74 patients, and blood of 52 of them. A cutoff of 10 copies *NPM-ALK* transcript per 10,000 copies of *ABL* (normalized copy numbers = NCN) was applied for outcome analysis and only samples with adequate copy numbers of the control gene (2000 copies *ABL*) were called quantifiable. 16/74 patients (22%) had more than 10 NCN detectable in BM. They had a cumulative incidence of relapse of 71 ± 14% compared to 18 ± 6% of the remaining 58 patients ($p < 0.001$). Quantification in blood showed a comparable result [32]. In summary, quantitative RQ-PCR allowed identifying patients with a very high risk of relapse already at diagnosis in this study. The Japanese NHL study group investigated the prognostic impact of *NPM-ALK* transcript quantification in blood and/or BM applying the same cutoff in 59 patients. The percentage of patients with more than 10 NCN in blood or BM was higher compared to the study of the BFM group (37% versus 22%) approaching the percentage of MDD-positive patients detectable by RT-PCR in the BFM group (50–55%). The PFS for the 22 patients with more than 10 NCN in blood and BM was 57.8 ± 12% compared to 84.9 ± 6% for patients with <10NCN ($p = 0.0016$) [57]. The difference between the studies of the BFM and Japanese study group emphasizes the need of international quality control for quantitative RQ-PCR to enable comparison of results in multinational studies. The quantitative PCR assay is not only expensive and difficult to perform with quality control in several laboratories. The most important challenge for its widespread use is the low cutoff of 10 NCN, which would imply reliable measurement of NPM-ALK copies at the detection limit of the quantitative PCR assay.

The prognostic meaning of MRD in NPM-ALK-positive ALCL was evaluated in a collaborative AIEOP and BFM group analysis. MRD was assessed before the second course of BFM-type chemotherapy using the qualitative *NPM-ALK* RT-PCR [54]. MRD could be analyzed in 52 of 103 MDD-positive patients. The 26 MRD-positive patients had a 5-year CI-R of 81 ± 6% compared to 31 ± 9% for MDD-positive/MRD-negative patients and 16 ± 5% for MDD-negative patients ($p < 0.0001$) (Fig. 10.3). The overall survival of MRD-positive patients was significantly lower compared to both other groups (OS 65 ± 9% compared to 92 ± 5% and 91 ± 4% for MDD-positive/MRD-negative and MDD-

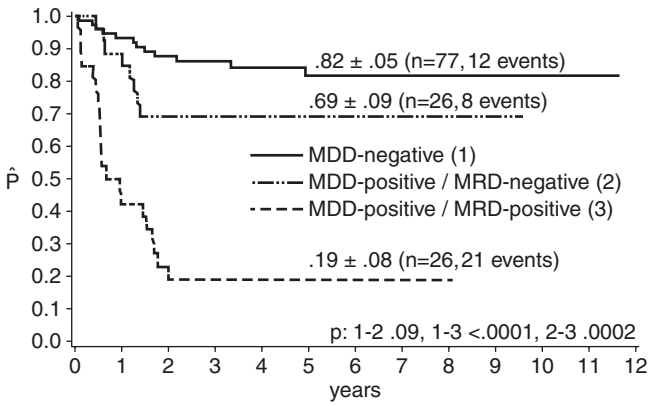

Fig. 10.3 5-year event-free survival of patients with NPM-ALK-positive anaplastic large cell lymphoma ALCL according to minimal residual disease (MRD) in bone marrow or blood measured by qualitative PCR for *NPM-ALK* before the second course of chemotherapy. (From Damm-Welk et al. [54], with permission)

negative patients, respectively; $p < 0.001$). MRD was associated with non-common histopathological subtype.

Quantitative MRD measurement for the longitudinal evaluation of very high-risk patients as tool to detect a relapse early or to judge the efficacy of a therapeutic element has not been analyzed systemically so far but reported in single cases. Quantitative PCR for *NPM-ALK* was used to monitor disease progression in two patients with relapsed ALCL after discontinuation of treatment with the ALK kinase inhibitor Crizotinib who had reached a clinical remission with MRD negativity. Rapid increase of MRD confirmed clinical progression and further MRD demonstrated the efficacy of a second treatment with Crizotinib [58]. In another case report of a patient with a very high-risk ALCL progressing during initial treatment, longitudinal MRD assessment allowed to estimate the efficacy of different therapeutic approaches (crizotinib, brentuximab, or nivolumab) and confirmed disease progression at occasions at which differentiation between infection and tumor progression was clinically not possible [59]. Kalinova et al. reported the course of MRD by *NPM-ALK*-specific RQ-PCR in ten ALCL patients. Five of these patients relapsed, with one patient having four relapses, and one patient experienced two relapses. All investigated samples of relapsed patients had detectable *NPM-ALK* copy numbers in the BM at the time point of relapse or closely before with five relapses showing an increase of at least half a log level of normalized *NPM-ALK* copy numbers. Measurement of *CD30* transcripts by RQ-PCR did not prove suitable for MRD monitoring in that study [60].

Measuring *NPM-ALK* transcripts in blood or BM cells currently is the standard method to assess circulating tumor cells in ALK-positive ALCL. Two other techniques of quantitative minimal disease measurement for ALK-positive ALCL have been compared to the standard RQ-PCR method. Krumbholz et al. assessed the possibility to use the patient-specific *NPM-ALK* fusion on the DNA level for minimal disease measurement. Analysis of the genomic *NPM-ALK* fusion sequence by a multiplex PCR assay allowed establishing individual fusion site-specific droplet digital (dd)-PCR assays for eight NPM-ALK-positive ALCL patients. Parallel quantification of *NPM-ALK* on 45 blood or BM samples with the fusion site-specific dd-PCR assay (DNA) and the established *NPM-ALK*-specific RQ-PCR assay (RNA), respectively, showed a high concordance between DNA and RNA-based MDD and MRD measurement. The DNA-fusion site dd-PCR assay was additionally applied to parallel plasma samples to quantify circulating tumor-specific DNA of the same patients. A significant correlation of the ctDNA values with the cellular *NPM-ALK* values on both the RNA and DNA level was observed [61]. Measurement of MDD/MRD using cellular or cell-free DNA as an additional tool for MRD monitoring may allow the detection of transcriptional quiescent ALCL cells. However, DNA-based MRD measurement needs initial tumor material for the establishment of the individual fusion site. Secondly, a flow cytometric assay to detect circulating ALCL cells in blood or BM has been developed and compared to quantitative PCR for NPM-ALK fusion transcripts [35]. Simultaneous measurement of intracellular ALK protein and surface CD30 expression allowed for the detection of one ALCL cell in 100,000 normal cells in serial dilutions of NPM-ALK-positive cell lines in blood. Despite concordance of the results in 11 patients, RQ-PCR for *NPM-ALK* was one log level more sensitive compared to FCM. Advantages of FCM MDD measurement over RQ-PCR would be the applicability to all ALK-positive ALCL. A possible prognostic meaning of the DNA-based and the FCM-based minimal disease quantification in addition to the standard technique needs to be analyzed prospectively in a larger cohort of ALK-positive ALCL patients.

Summary: Present Status of Minimal Disseminated and Minimal Residual Disease in Childhood NHL

The following tables (Tables 10.1 and 10.2) summarize the established techniques for the detection of MDD and MRD in childhood Non-Hodgkin lymphomas and the prognostic meaning of MDD and MRD during first-line treatment according to the NHL subtypes. Currently, no studies have been published on the prognostic value of MDD and MRD in PTCL or relapsed NHL. Since the studies on MRD in relapsed acute lymphoblastic leukemia also included systemic relapses of lymphoblastic lymphomas, the poor prognostic meaning of a high MRD after re-induction and before as well as after stem cell transplantation can likely be

Table 10.1 Techniques established to detect minimal disease in childhood Non-Hodgkin lymphoma and their sensitivities

	LBL	BL/B-AL	ALCL	PTCL
NHL-specific marker	–	67% t(8;14) with MYC-IgH	>85% t(2;5) with NPM-ALK	–
PCR for Ig/TCR rearrangement (DNA) Initial tumor?	+ (10^{-5}–10^{-6}) Needed	+[a] (10^{-4}) Needed	–	+ (10^{-5}–10^{-6}) Needed
FCM for aberrant immunophenotype Initial tumor?	+[b] (10^{-4}) Not needed	–	+[b] (10^{-4}) Not needed	–
LD-PCR for MYC-IgH (DNA) Initial tumor?		+ (10^{-3}–10^{-5}) Needed		
RT/RQ-PCR for NPM-ALK transcripts Initial tumor?			+[b] (10^{-5}–10^{-6}) Not needed	
Multiplex PCR followed by patient-specific primers for NPM-ALK (DNA) Initial tumor?			+[b] (10^{-5}–10^{-6}) Needed	

FCM flow cytometry, *LBL* lymphoblastic lymphoma, *BL* Burkitt lymphoma, *B-AL* Burkitt leukemia, *ALCL* anaplastic large cell lymphoma, *PTCL* peripheral T-cell lymphoma, *LD-PCR* long-distance PCR
[a]IgV(H) primer pools may eliminate the need for initial tumor material
[b]blood can substitute for bone marrow

Table 10.2 Clinical data on the prognostic impact of minimal disseminated disease (MDD) and minimal residual disease (MRD) in childhood Non-Hodgkin lymphoma

	T-LBL[a]		BL/B-AL		ALCL	
NHL-specific marker			67% t(8;14) with MYC-IgH		>85% t(2;5) with NPM-ALK	
	MDD	MRD	MDD	MRD	MDD	MRD
PCR for Ig/TCR rearrangement (DNA)	n.d.	n.d.	n.d.	(+)	–	–
FCM for aberrant immunophenotype	+	(+)	–	–	(+)	n.d.
LD-PCR for MYC-IgH (DNA)	–	–	+	+[b]		
RT/RQ-PCR for NPM-ALK transcripts	–	–	–	–	++	+[b]
Multiplex PCR for NPM-ALK (DNA)	–	–	–	–	n.d.	n.d.

[a]There are no data on pB-LBL
[b]measurement before the second course of BFM-type chemotherapy
n.d. no data, – not applicable, (+) hints toward a possible prognostic impact, + at least one study demonstrating a prognostic meaning, ++ validated independent prognostic factor

transferred from ALL to LBL [1, 2, 4, 5]. The potential application of MRD for disease monitoring at the end of therapy and as surveillance after therapy has not been studied systematically so far so that this is not included in the tables. Furthermore, NGS-based techniques for MDD and MRD measurement have not yet been studied in children.

Future Directions

Considered their method-specific pitfalls, MDD and MRD provide highly sensitive and specific assessments of subclinical minimal disease in lymphoma.

Technically, NGS-based methods for the detection of patient-specific or disease-specific markers bare the potential for broader applicability, especially for DLBCL and BL, but also for LBL and definition of the DNA breakpoints in ALK-positive ALCL and BL. In addition, preliminary data indicate the potential use of circulating tumor-derived DNA in plasma or serum as minimal disease marker.

Quality control must be established for each method among minimal disease laboratories to enable comparability and inclusion of MDD and MRD measurements into multinational clinical trials. Also, tissue, blood, and bone marrow samples have to be collected, processed, and stored in a comparable optimized fashion in clinical trials.

So far, the prognostic meaning of MDD only has been validated for patients with ALK-positive ALCL and is, therefore, ready to be used for stratification in clinical trials. Further trials need to establish the clinical meaning of MDD for the other lymphoma entities in children, especially BL and T-LBL.

The possible use of early MRD for a change of treatment for individual patients (either intensification or reduction) should be validated for both B-AL and ALCL. Furthermore, end-of-treatment MRD, MDD at diagnosis of relapse, and

MRD as potential substitute for radiological disease monitoring are areas of minimal disease measurement which still need to be explored for pediatric NHL. Preemptive MRD-guided therapy of an impending relapse and MRD as endpoint for clinical studies are unexplored areas for MRD in pediatric NHL.

References

1. Bader P, Kreyenberg H, Henze GH, Eckert C, Reising M, Willasch A, Barth A, Borkhardt A, Peters C, Handgretinger R, Sykora KW, Holter W, Kabisch H, Klingebiel T, von Stackelberg A, Group A-RBS. Prognostic value of minimal residual disease quantification before allogeneic stem-cell transplantation in relapsed childhood acute lymphoblastic leukemia: the ALL-REZ BFM Study Group. J Clin Oncol. 2009;27(3):377–84.
2. Bader P, Kreyenberg H, von Stackelberg A, Eckert C, Salzmann-Manrique E, Meisel R, Poetschger U, Stachel D, Schrappe M, Alten J, Schrauder A, Schulz A, Lang P, Muller I, Albert MH, Willasch AM, Klingebiel TE, Peters C. Monitoring of minimal residual disease after allogeneic stem-cell transplantation in relapsed childhood acute lymphoblastic leukemia allows for the identification of impending relapse: results of the ALL-BFM-SCT 2003 trial. J Clin Oncol. 2015;33(11):1275–84. https://doi.org/10.1200/JCO.2014.58.4631.
3. Campana D, Pui CH. Minimal residual disease-guided therapy in childhood acute lymphoblastic leukemia. Blood. 2017;129(14):1913–8. https://doi.org/10.1182/blood-2016-12-725804.
4. Eckert C, Biondi A, Seeger K, Cazzaniga G, Hartmann R, Beyermann B, Pogodda M, Proba J, Henze G. Prognostic value of minimal residual disease in relapsed childhood acute lymphoblastic leukaemia. Lancet. 2001;358(9289):1239–41. https://doi.org/10.1016/S0140-6736(01)06355-3.
5. Eckert C, Hagedorn N, Sramkova L, Mann G, Panzer-Grumayer R, Peters C, Bourquin JP, Klingebiel T, Borkhardt A, Cario G, Alten J, Escherich G, Astrahantseff K, Seeger K, Henze G, von Stackelberg A. Monitoring minimal residual disease in children with high-risk relapses of acute lymphoblastic leukemia: prognostic relevance of early and late assessment. Leukemia. 2015;29(8):1648–55. https://doi.org/10.1038/leu.2015.59.
6. Schrappe M. Detection and management of minimal residual disease in acute lymphoblastic leukemia. Hematology Am Soc Hematol Educ Program. 2014;2014(1):244–9. https://doi.org/10.1182/asheducation-2014.1.244.
7. van Dongen JJ, van der Velden VH, Bruggemann M, Orfao A. Minimal residual disease diagnostics in acute lymphoblastic leukemia: need for sensitive, fast, and standardized technologies. Blood. 2015;125(26):3996–4009. https://doi.org/10.1182/blood-2015-03-580027.
8. Chase ML, Armand P. Minimal residual disease in non-Hodgkin lymphoma – current applications and future directions. Br J Haematol. 2018;180(2):177–88. https://doi.org/10.1111/bjh.14996.
9. Herrera AF, Armand P. Minimal residual disease assessment in lymphoma: methods and applications. J Clin Oncol. 2017;35(34):3877–87. https://doi.org/10.1200/JCO.2017.74.5281.
10. Kuroda T, Morikawa N, Matsuoka K, Fujino A, Honna T, Nakagawa A, Kumagai M, Masaki H, Saeki M. Prognostic significance of circulating tumor cells and bone marrow micrometastasis in advanced neuroblastoma. J Pediatr Surg. 2008;43(12):2182–5. https://doi.org/10.1016/j.jpedsurg.2008.08.046.
11. Kurtz DM, Green MR, Bratman SV, Scherer F, Liu CL, Kunder CA, Takahashi K, Glover C, Keane C, Kihira S, Visser B, Callahan J, Kong KA, Faham M, Corbelli KS, Miklos D, Advani RH, Levy R, Hicks RJ, Hertzberg M, Ohgami RS, Gandhi MK, Diehn M, Alizadeh AA. Noninvasive monitoring of diffuse large B-cell lymphoma by immunoglobulin high-throughput sequencing. Blood. 2015;125(24):3679–87. https://doi.org/10.1182/blood-2015-03-635169.
12. Roschewski M, Dunleavy K, Pittaluga S, Moorhead M, Pepin F, Kong K, Shovlin M, Jaffe ES, Staudt LM, Lai C, Steinberg SM, Chen CC, Zheng J, Willis TD, Faham M, Wilson WH. Circulating tumour DNA and CT monitoring in patients with untreated diffuse large B-cell lymphoma: a correlative biomarker study. Lancet Oncol. 2015;16(5):541–9. https://doi.org/10.1016/S1470-2045(15)70106-3.
13. Schleiermacher G, Peter M, Oberlin O, Philip T, Rubie H, Mechinaud F, Sommelet-Olive D, Landman-Parker J, Bours D, Michon J, Delattre O. Increased risk of systemic relapses associated with bone marrow micrometastasis and circulating tumor cells in localized Ewing tumor. J Clin Oncol. 2003;21(1):85–91.
14. Vo KT, Edwards JV, Epling CL, Sinclair E, Hawkins DS, Grier HE, Janeway KA, Barnette P, McIlvaine E, Krailo MD, Barkauskas DA, Matthay KK, Womer RB, Gorlick RG, Lessnick SL, Mackall CL, DuBois SG. Impact of two measures of micrometastatic disease on clinical outcomes in patients with newly diagnosed ewing sarcoma: a report from the Children's Oncology Group. Clin Cancer Res. 2016;22(14):3643–50. https://doi.org/10.1158/1078-0432.CCR-15-2516.
15. Minard-Colin V, Brugieres L, Reiter A, Cairo MS, Gross TG, Woessmann W, Burkhardt B, Sandlund JT, Williams D, Pillon M, Horibe K, Auperin A, Le Deley MC, Zimmerman M, Perkins SL, Raphael M, Lamant L, Klapper W, Mussolin L, Poirel HA, Macintyre E, Damm-Welk C, Rosolen A, Patte C. Non-Hodgkin lymphoma in children and adolescents: progress through effective collaboration, current knowledge, and challenges ahead. J Clin Oncol. 2015;33(27):2963–74. https://doi.org/10.1200/JCO.2014.59.5827.
16. Dalla-Favera R, Bregni M, Erikson J, Patterson D, Gallo RC, Croce CM. Human c-myc onc gene is located on the region of chromosome 8 that is translocated in Burkitt lymphoma cells. Proc Natl Acad Sci U S A. 1982;79(24):7824–7.
17. Basso K, Frascella E, Zanesco L, Rosolen A. Improved long-distance polymerase chain reaction for the detection of t(8;14)(q24;q32) in Burkitt's lymphomas. Am J Pathol. 1999;155(5):1479–85. https://doi.org/10.1016/S0002-9440(10)65463-6.
18. Akasaka T, Muramatsu M, Ohno H, Miura I, Tatsumi E, Fukuhara S, Mori T, Okuma M. Application of long-distance polymerase chain reaction to detection of junctional sequences created by chromosomal translocation in mature B-cell neoplasms. Blood. 1996;88(3):985–94.
19. Lovisa F, Mussolin L, Corral L, Pillon M, Cazzaniga G, Biondi A, Rosolen A. IGH and IGK gene rearrangements as PCR targets for pediatric Burkitt's lymphoma and mature B-ALL MRD analysis. Lab Investig. 2009;89(10):1182–6. https://doi.org/10.1038/labinvest.2009.81.
20. van Dongen JJ, Langerak AW, Bruggemann M, Evans PA, Hummel M, Lavender FL, Delabesse E, Davi F, Schuuring E, Garcia-Sanz R, van Krieken JH, Droese J, Gonzalez D, Bastard C, White HE, Spaargaren M, Gonzalez M, Parreira A, Smith JL, Morgan GJ, Kneba M, Macintyre EA. Design and standardization of PCR primers and protocols for detection of clonal immunoglobulin and T-cell receptor gene recombinations in suspect lymphoproliferations: report of the BIOMED-2 Concerted Action BMH4-CT98-3936. Leukemia. 2003;17(12):2257–317. https://doi.org/10.1038/sj.leu.2403202.
21. Agsalda M, Kusao I, Troelstrup D, Shiramizu B. Screening for residual disease in pediatric burkitt lymphoma using consensus primer pools. Adv Hematol. 2009;2009:412163. https://doi.org/10.1155/2009/412163.

22. Shiramizu B, Goldman S, Smith L, Agsalda-Garcia M, Galardy P, Perkins SL, Frazer JK, Sanger W, Anderson JR, Gross TG, Weinstein H, Harrison L, Barth MJ, Mussolin L, Cairo MS. Impact of persistent minimal residual disease post-consolidation therapy in children and adolescents with advanced Burkitt leukaemia: a Children's Oncology Group Pilot Study Report. Br J Haematol. 2015;170(3):367–71. https://doi.org/10.1111/bjh.13443.
23. Damm-Welk C, Klapper W, Oschlies I, Gesk S, Rottgers S, Bradtke J, Siebert R, Reiter A, Woessmann W. Distribution of NPM1-ALK and X-ALK fusion transcripts in paediatric anaplastic large cell lymphoma: a molecular-histological correlation. Br J Haematol. 2009;146(3):306–9.
24. Lamant L, Meggetto F, al Saati T, Brugieres L, de Paillerets BB, Dastugue N, Bernheim A, Rubie H, Terrier-Lacombe MJ, Robert A, Rigal F, Schlaifer D, Shiuta M, Mori S, Delsol G. High incidence of the t(2;5)(p23;q35) translocation in anaplastic large cell lymphoma and its lack of detection in Hodgkin's disease. Comparison of cytogenetic analysis, reverse transcriptase-polymerase chain reaction, and P-80 immunostaining. Blood. 1996;87(1):284–91.
25. Perkins SL, Pickering D, Lowe EJ, Zwick D, Abromowitch M, Davenport G, Cairo MS, Sanger WG. Childhood anaplastic large cell lymphoma has a high incidence of ALK gene rearrangement as determined by immunohistochemical staining and fluorescent in situ hybridisation: a genetic and pathological correlation. Br J Haematol. 2005;131(5):624–7.
26. Hernandez L, Pinyol M, Hernandez S, Bea S, Pulford K, Rosenwald A, Lamant L, Falini B, Ott G, Mason DY, Delsol G, Campo E. TRK-fused gene (TFG) is a new partner of ALK in anaplastic large cell lymphoma producing two structurally different TFG-ALK translocations. Blood. 1999;94(9):3265–8.
27. Lamant L, Dastugue N, Pulford K, Delsol G, Mariame B. A new fusion gene TPM3-ALK in anaplastic large cell lymphoma created by a (1;2)(q25;p23) translocation. Blood. 1999;93(9):3088–95.
28. Ma Z, Cools J, Marynen P, Cui X, Siebert R, Gesk S, Schlegelberger B, Peeters B, De Wolf-Peeters C, Wlodarska I, Morris SW. Inv(2)(p23q35) in anaplastic large-cell lymphoma induces constitutive anaplastic lymphoma kinase (ALK) tyrosine kinase activation by fusion to ATIC, an enzyme involved in purine nucleotide biosynthesis. Blood. 2000;95(6):2144–9.
29. Meech SJ, McGavran L, Odom LF, Liang X, Meltesen L, Gump J, Wei Q, Carlsen S, Hunger SP. Unusual childhood extramedullary hematologic malignancy with natural killer cell properties that contains tropomyosin 4--anaplastic lymphoma kinase gene fusion. Blood. 2001;98(4):1209–16.
30. Tort F, Pinyol M, Pulford K, Roncador G, Hernandez L, Nayach I, Kluin-Nelemans HC, Kluin P, Touriol C, Delsol G, Mason D, Campo E. Molecular characterization of a new ALK translocation involving moesin (MSN-ALK) in anaplastic large cell lymphoma. Lab Investig. 2001;81(3):419–26.
31. Touriol C, Greenland C, Lamant L, Pulford K, Bernard F, Rousset T, Mason DY, Delsol G. Further demonstration of the diversity of chromosomal changes involving 2p23 in ALK-positive lymphoma: 2 cases expressing ALK kinase fused to CLTCL (clathrin chain polypeptide-like). Blood. 2000;95(10):3204–7.
32. Damm-Welk C, Busch K, Burkhardt B, Schieferstein J, Viehmann S, Oschlies I, Klapper W, Zimmermann M, Harbott J, Reiter A, Woessmann W. Prognostic significance of circulating tumor cells in bone marrow or peripheral blood as detected by qualitative and quantitative PCR in pediatric NPM-ALK-positive anaplastic large-cell lymphoma. Blood. 2007;110(2):670–7.
33. Downing JR, Shurtleff SA, Zielenska M, Curcio-Brint AM, Behm FG, Head DR, Sandlund JT, Weisenburger DD, Kossakowska AE, Thorner P. Molecular detection of the (2;5) translocation of non-Hodgkin's lymphoma by reverse transcriptase-polymerase chain reaction. Blood. 1995;85(12):3416–22.
34. Mussolin L, Pillon M, d'Amore ES, Santoro N, Lombardi A, Fagioli F, Zanesco L, Rosolen A. Prevalence and clinical implications of bone marrow involvement in pediatric anaplastic large cell lymphoma. Leukemia. 2005;19(9):1643–7.
35. Damm-Welk C, Schieferstein J, Schwalm S, Reiter A, Woessmann W. Flow cytometric detection of circulating tumour cells in nucleophosmin/anaplastic lymphoma kinase-positive anaplastic large cell lymphoma: comparison with quantitative polymerase chain reaction. Br J Haematol. 2007;138(4):459–66.
36. Burkhardt B. Paediatric lymphoblastic T-cell leukaemia and lymphoma: one or two diseases? Br J Haematol. 2010;149(5):653–68. https://doi.org/10.1111/j.1365-2141.2009.08006.x.
37. Stark B, Avigad S, Luria D, Manor S, Reshef-Ronen T, Avrahami G, Yaniv I. Bone marrow minimal disseminated disease (MDD) and minimal residual disease (MRD) in childhood T-cell lymphoblastic lymphoma stage III, detected by flow cytometry (FC) and real-time quantitative polymerase chain reaction (RQ-PCR). Pediatr Blood Cancer. 2009;52(1):20–5. https://doi.org/10.1002/pbc.21823.
38. Coustan-Smith E, Sandlund JT, Perkins SL, Chen H, Chang M, Abromowitch M, Campana D. Minimal disseminated disease in childhood T-cell lymphoblastic lymphoma: a report from the children's oncology group. J Clin Oncol. 2009;27(21):3533–9. https://doi.org/10.1200/JCO.2008.21.1318.
39. Mussolin L, Buldini B, Lovisa F, Carraro E, Disaro S, Lo Nigro L, d'Amore ES, Pillon M, Basso G. Detection and role of minimal disseminated disease in children with lymphoblastic lymphoma: the AIEOP experience. Pediatr Blood Cancer. 2015;62(11):1906–13. https://doi.org/10.1002/pbc.25607.
40. Dworzak MN, Gaipa G, Ratei R, Veltroni M, Schumich A, Maglia O, Karawajew L, Benetello A, Potschger U, Husak Z, Gadner H, Biondi A, Ludwig WD, Basso G. Standardization of flow cytometric minimal residual disease evaluation in acute lymphoblastic leukemia: multicentric assessment is feasible. Cytometry B Clin Cytom. 2008;74(6):331–40. https://doi.org/10.1002/cyto.b.20430.
41. Burkhardt B, Woessmann W, Zimmermann M, Kontny U, Vormoor J, Doerffel W, Mann G, Henze G, Niggli F, Ludwig WD, Janssen D, Riehm H, Schrappe M, Reiter A. Impact of cranial radiotherapy on central nervous system prophylaxis in children and adolescents with central nervous system-negative stage III or IV lymphoblastic lymphoma. J Clin Oncol. 2006;24(3):491–9. https://doi.org/10.1200/JCO.2005.02.2707.
42. Reiter A, Schrappe M, Ludwig WD, Tiemann M, Parwaresch R, Zimmermann M, Schirg E, Henze G, Schellong G, Gadner H, Riehm H. Intensive ALL-type therapy without local radiotherapy provides a 90% event-free survival for children with T-cell lymphoblastic lymphoma: a BFM group report. Blood. 2000;95(2):416–21.
43. Balbach ST, Makarova O, Bonn BR, Zimmermann M, Rohde M, Oschlies I, Klapper W, Rossig C, Burkhardt B. Proposal of a genetic classifier for risk group stratification in pediatric T-cell lymphoblastic lymphoma reveals differences from adult T-cell lymphoblastic leukemia. Leukemia. 2016;30(4):970–3. https://doi.org/10.1038/leu.2015.203.
44. Bonn BR, Rohde M, Zimmermann M, Krieger D, Oschlies I, Niggli F, Wrobel G, Attarbaschi A, Escherich G, Klapper W, Reiter A, Burkhardt B. Incidence and prognostic relevance of genetic variations in T-cell lymphoblastic lymphoma in childhood and adolescence. Blood. 2013;121(16):3153–60. https://doi.org/10.1182/blood-2012-12-474148.
45. Callens C, Baleydier F, Lengline E, Ben Abdelali R, Petit A, Villarese P, Cieslak A, Minard-Colin V, Rullier A, Moreau A, Baruchel A, Schmitt C, Asnafi V, Bertrand Y, Macintyre E. Clinical impact of NOTCH1 and/or FBXW7 mutations, FLASH deletion, and TCR status in pediatric T-cell lymphoblastic lymphoma. J Clin Oncol. 2012;30(16):1966–73. https://doi.org/10.1200/JCO.2011.39.7661.

46. Mussolin L, Basso K, Pillon M, d'Amore ES, Lombardi A, Luzzatto L, Zanesco L, Rosolen A. Prospective analysis of minimal bone marrow infiltration in pediatric Burkitt's lymphomas by long-distance polymerase chain reaction for t(8;14)(q24;q32). Leukemia. 2003;17(3):585–9.
47. Mussolin L, Pillon M, d'Amore ES, Conter V, Piglione M, Lo Nigro L, Garaventa A, Buffardi S, Arico M, Rosolen A. Minimal disseminated disease in high-risk Burkitt's lymphoma identifies patients with different prognosis. J Clin Oncol. 2011;29(13):1779–84.
48. Busch K, Borkhardt A, Wossmann W, Reiter A, Harbott J. Combined polymerase chain reaction methods to detect c-myc/IgH rearrangement in childhood Burkitt's lymphoma for minimal residual disease analysis. Haematologica. 2004;89(7):818–25.
49. Cazzaniga G, Biondi A. Molecular monitoring of childhood acute lymphoblastic leukemia using antigen receptor gene rearrangements and quantitative polymerase chain reaction technology. Haematologica. 2005;90(3):382–90.
50. Mussolin L, Pillon M, Conter V, Piglione M, Lo Nigro L, Pierani P, Micalizzi C, Buffardi S, Basso G, Zanesco L, Rosolen A. Prognostic role of minimal residual disease in mature B-cell acute lymphoblastic leukemia of childhood. J Clin Oncol. 2007;25(33):5254–61.
51. Pillon M, Mussolin L, Carraro E, Conter V, Arico M, Vinti L, Garaventa A, Piglione M, Buffardi S, Sala A, Santoro N, Lo Nigro L, Mura R, Tondo A, Casale F, Farruggia P, Pierani P, Cesaro S, d'Amore ES, Basso G. Detection of prognostic factors in children and adolescents with Burkitt and Diffuse Large B-Cell Lymphoma treated with the AIEOP LNH-97 protocol. Br J Haematol. 2016;175(3):467–75. https://doi.org/10.1111/bjh.14240.
52. Chiarle R, Voena C, Ambrogio C, Piva R, Inghirami G. The anaplastic lymphoma kinase in the pathogenesis of cancer. Nat Rev Cancer. 2008;8(1):11–23.
53. Pulford K, Lamant L, Espinos E, Jiang Q, Xue L, Turturro F, Delsol G, Morris SW. The emerging normal and disease-related roles of anaplastic lymphoma kinase. Cell Mol Life Sci. 2004;61(23):2939–53.
54. Damm-Welk C, Mussolin L, Zimmermann M, Pillon M, Klapper W, Oschlies I, d'Amore ES, Reiter A, Woessmann W, Rosolen A. Early assessment of minimal residual disease identifies patients at very high relapse risk in NPM-ALK-positive anaplastic large-cell lymphoma. Blood. 2014;123(3):334–7.
55. Mussolin L, mm-Welk C, Pillon M, Zimmermann M, Franceschetto G, Pulford K, Reiter A, Rosolen A, Woessmann W. Use of minimal disseminated disease and immunity to NPM-ALK antigen to stratify ALK-positive ALCL patients with different prognosis. Leukemia. 2013;27(2):416–22.
56. Damm-Welk C, Yamashita Y, Bench A, Turner S, Lamant L, Verge V, Tosato E, Schieferstein J, Mussolin L. Quality control of standardized methods for NPM-ALK RT-PCR and anti-ALK-antibody-measurement for anaplastic large cell lymphoma -a report of the EICNHL reference laboratory group. Br J Haematol. 2015;171(S1):56.
57. Iijima-Yamashita Y, Mori T, Nakazawa A, Fukano R, Takimoto T, Tsurusawa M, Kobayashi R, Horibe K. Prognostic impact of minimal disseminated disease and immune response to NPM-ALK in Japanese children with ALK-positive anaplastic large cell lymphoma. Int J Hematol. 2018;107(2):244–50. https://doi.org/10.1007/s12185-017-2338-6.
58. Gambacorti-Passerini C, Mussolin L, Brugieres L. Abrupt relapse of ALK-positive lymphoma after discontinuation of crizotinib. N Engl J Med. 2016;374(1):95–6. https://doi.org/10.1056/NEJMc1511045.
59. Hebart H, Lang P, Woessmann W. Nivolumab for refractory anaplastic large cell lymphoma: a case report. Ann Intern Med. 2016;165(8):607–8. https://doi.org/10.7326/L16-0037.
60. Kalinova M, Krskova L, Brizova H, Kabickova E, Kepak T, Kodet R. Quantitative PCR detection of NPM/ALK fusion gene and CD30 gene expression in patients with anaplastic large cell lymphoma--residual disease monitoring and a correlation with the disease status. Leuk Res. 2008;32(1):25–32. https://doi.org/10.1016/j.leukres.2007.01.002.
61. Krumbholz M, Woessmann W, Zierk J, Seniuk D, Ceppi P, Zimmermann M, Singh VK, Metzler M, Damm-Welk C. Characterization and diagnostic application of genomic NPM-ALK fusion sequences in anaplastic large-cell lymphoma. Oncotarget. 2018;9:26543–55.

Prognostic Factors in Childhood and Adolescent Non-Hodgkin Lymphoma

11

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Introduction

Non-Hodgkin lymphoma (NHL) represents the third most common tumor in children less than 15 years of age in North America and Western Europe. The prognosis has improved dramatically over the past 40 years and more than 85% of patients in North America and Western Europe are cured with upfront multiagent chemotherapy. The four most common histologies that occur in children and adolescents include anaplastic large cell lymphoma (ALCL), lymphoblastic lymphoma (LL), Burkitt lymphoma (BL), and diffuse large B-cell lymphoma (DLBCL). A number of risk or prognostic factors have been identified in each of these subtypes including histology, stage, extent of disease, minimal disseminated disease (MDD), minimal residual disease (MRD), disease burden, cytogenetics, and molecular genetics, among others. In this chapter, we will outline the known prognostic factors in each of the common histologies, including ALCL, LL, BL, and DLBCL.

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Prognostic Factors in T-Cell Lymphomas

Anaplastic Large Cell Lymphoma

Anaplastic large cell lymphoma (ALCL) is one of the most common pediatric T-cell malignancies [1]. First described in 1985, ALCL is characterized by large pleomorphic tumor cells that tend to grow cohesively, consistently expressing CD30 in their surface and Golgi region [2, 3]. Previously named Ki-1, CD30 is an activation-induced antigen that belongs to the tumor necrosis factor (TNF) receptor superfamily [4–6]. Stimulation of CD30 in ALCL has been shown to trigger competing cellular effects, including activation of caspase and NK-κB-mediated cell survival pathways [6, 7]. Furthermore, the majority of pediatric ALCL cases are associated with nonrandom chromosomal rearrangements involving the anaplastic lymphoma kinase (*ALK*), a tyrosine kinase gene localized to chromosome 2p23 [2, 8]. Juxtaposition of *ALK* and other gene coding regions result in gene fusion and the generation of novel chimeric proteins, an oncogenic mechanism not very common in lymphomas. Most frequently, the chimeric protein includes the catalytic domain of ALK and the oligomerization motif of nucleophosmin (*NPM1*, localized to 5q35), a nuclear phosphoprotein [9]. The presence of NPM1/ALK protein leads to ectopic expression of ALK and has shown to mediate malignant transformation in vitro and in vivo, by activation of different downstream effectors [10, 11]. The importance of ALK is underlined by the fact that its expression defines two separate ALCL entities (ALK-positive and ALK-negative ALCL) according to the most recent edition of the *WHO Classification of Tumors of Haematopoietic and Lymphoid Tissues* [3].

Although insights into disease biology have expanded in recent years, still about one-third of the pediatric patients with ALCL will have an unfavorable outcome, regardless of the treatment strategy used [12–15]. Understanding the heterogeneity of this group of patients, and predicting which subset of patients will not respond to primary treatment or

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Table 11.1 Prognostic factors in children and adolescents with anaplastic large lymphoma

Prognostic factor	Impact	Risk/outcome	Frequency	Reference
Lymphoma-related				
"Non-common" morphology	Unfavorable	HR[c] 2.0 (95% CI 1.3–3.0) $P = 0.002$	30%	[16]
ALK protein expression	Favorable	5y OS 78.8% vs. 32.9%, $P < 0.01$	>90%	[27]
DUSP22 rearrangements[a]	Favorable	5y OS 90%, $P < 0.0001$	30%	[35]
TP63 rearrangements[a]	Unfavorable	5y OS 17%, $P < 0.0001$	8%	[35]
CD8 expression	Unfavorable	HR 2.6 (95% CI 1.43–7.65) $P < 0.01$	5–35%	[39]
CD56 expression	Unfavorable	$P = 0.02$	18%	[29]
Survivin expression	Unfavorable	5y FFS[d] 34% vs. 100%, $P = 0.009$[g]	50%[b]	[42]
Clinical characteristics				
Visceral, skin, mediastinal involvement	Unfavorable	3y EFS < 45%, $P < 0.01$	30–40%	[19]
Advanced stage	Unfavorable	3y EFS 94% vs. 55%, $P = 0.006$	70%	[19]
B symptoms	Unfavorable	5y EFS 67%, $P = 0.04$	50%	[14]
Bone marrow involvement	Unfavorable	$P = 0.03$	13%	[13]
MDD positive[e]	Unfavorable	CI-R[f] 50% vs. 15%, $P < 0.001$	47%	[20]
Patient immune status				
Elevated ALK antibody titers	Favorable	CI-R 11% vs. 31% vs. 63%, $P < 0.001$[h]	Variable	[59]
Elevated cytokine (IL6) levels	Favorable	EFS 85.7% vs. 44.6%, $P < 0.001$	N/A	[58]

[a]*DUSP22* and *TP63* rearrangements have been described in ALK-negative ALCL only
[b]Adult data only
[c]*HR* hazard ratio
[d]*FFS* failure-free survival
[e]*MDD* minimal disseminated disease detected by PCR (NPM-ALK transcripts)
[f]*CI-R* cumulative incidence of relapse
[g]FS for patients with ALK-positive ALCL. Surviving expression seems to be prognostic independent of ALK expression [42]
[h]Numbers correspond to lower CI-R for ALK antibody titers ≥1/60,750 (11% ± 6%); titers 1/2025–1/60,750 (31% ± 8%); and titers ≤1/750 (63% ± 10%)

will suffer a relapse, in the context of standard or new treatment approaches are crucial. It may help us contextualize results of previous and experimental interventions and identify patients who will have a favorable outcome or the ones for whom novel therapies should be immediately offered. A variety of lymphoma- and host-related factors that may interfere with the prognosis of patients with ALCL will be discussed here (Table 11.1).

Lymphoma-Related Prognostic Factors

Morphology

Under the same broad category, ALCL patients may differ according to morphologic features. The WHO classification recognizes five different histological patterns: "common pattern," "lymphohistiocytic pattern," "small cell pattern," "Hodgkin-like pattern," and "composite pattern" (Fig. 11.1) [3]. In all subtypes, cells with eccentric horseshoe- or reniform-shaped nucleus with prominent nucleoli, or "hallmark cells," can be found. The "common pattern" is characterized by the presence of cells with abundant cytoplasm with multiple irregular nuclei, which sometimes resemble Hodgkin's lymphoma (HL) Reed-Sternberg cells. The "lymphohistiocytic pattern" is a mixture of CD30+ lymphoma cells in a large background of reactive histiocytes, while the "small cell pattern" is characterized by the presence of small/medium-sized lymphoma cells with irregular nuclei, similar to morphologic features of peripheral T-cell lymphoma (PTCL). Bone marrow (BM) involvement seems to be more common in patients with "small cell variant." Less commonly, histological findings resemble nodular sclerosis HL in the "Hodgkin-like pattern," and patients with the "composite pattern" will have a combination of more than one morphological subtype [3]. ALK-positive and ALK-negative ALCL share the same morphological features, being distinguishable only by the presence or absence of ALK expression [3].

Some studies have tried to tie morphologic pattern to prognosis [16–20]. Patients with "small cell variant" are thought to present with more aggressive lymphoma with multi-organ involvement and guarded prognosis [17, 18]. In a large series of 375 pediatric patients with ALK-positive ALCL, not only the "small cell variant" but the "lymphohistiocytic subtype" was linked to outcome (Fig. 11.2) [16]. Both variants accounted for 32% of the cases, and their presence was significantly associated with a high risk of failure (hazard ratio, 2.0; 95% CI, 1.3–3.0; $p = 0.002$) in the multivariate analysis controlling for clinical characteristics [16]. There seems to be an association between detection of NPM1/ALK fusion protein by polymerase chain reaction (PCR) in the BM and non-"common pattern" histology, in addition to the presence of clinical risk features of mediastinal

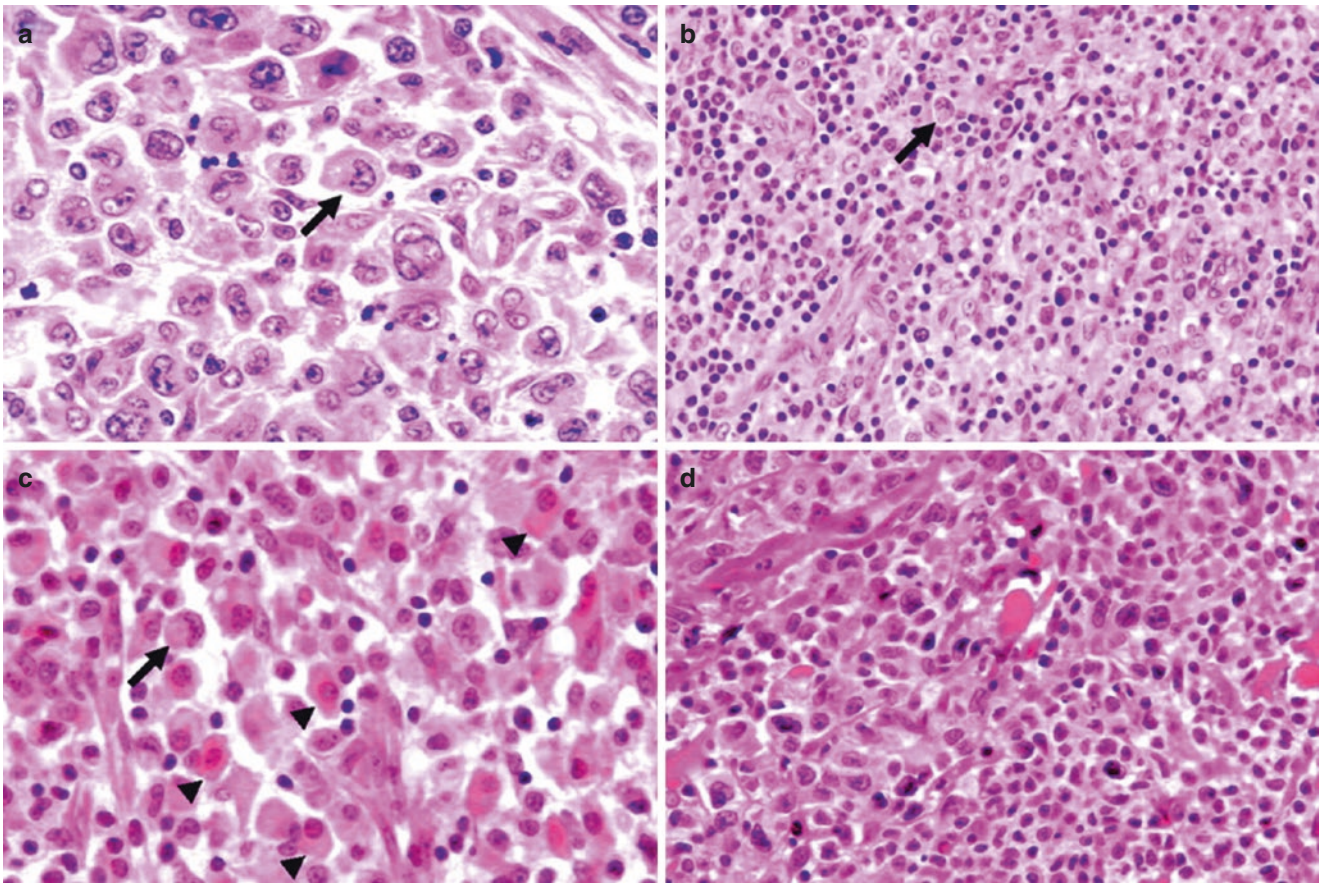
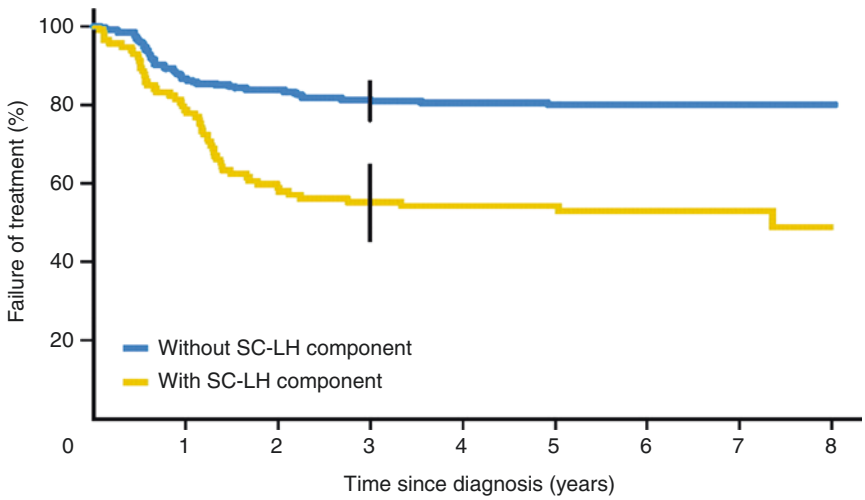


Fig. 11.1 (**a**) ALCL, common variant. Predominant population of large cells. Note: hallmark cells, with eccentric, horseshoe, or kidney-shaped nuclei, with a juxtanuclear eosinophilic region (arrow; hematoxylin and eosin [HE]; magnification, ×400). (**b**) ALCL, small cell variant. Predominant population of small cells with irregular nuclei and scattered hallmark cells (arrow; HE; magnification, ×200). (**c**) ALCL, lymphohistiocytic variant. Malignant cells (arrow) are admixed with a predominant population of reactive histiocytes (arrowhead) and are sometimes difficult to defect (HE; magnification, ×400). (**d**) Composite ALCL. Association in a single biopsy of areas of common pattern (left side) and small cell pattern (right side; HE; magnification, ×400). (Reprinted from Lamant et al. [16], with permission)



No. at risk									
Without SC-LH component	247	214	206	197	179	130	81	47	23
With SC-LH component	114	88	65	61	50	42	31	17	8

Fig. 11.2 Time to treatment failure curve according to the presence of a small cell (SC) and/or lymphohistiocytic (LH) component. SC-LH component was associated with high risk of failure (HR 2.0, 95% C.I. 1.3–3.0, $P = 0.02$). (Reprinted from Lamant et al. [16], with permission)

and visceral organ involvement, and advanced-stage disease, resulting in worse outcomes in non-"common pattern" of pediatric ALCL [20]. However, such association was not confirmed in another study [19]. Instead, visceral or mediastinal involvement and higher lactic dehydrogenase (LDH) were shown to be predictive of a higher risk of failure among 82 children treated in two consecutive trials [19].

It is possible that the different morphologic patterns described in patients with ALCL represent variants of the same disease and are not related to outcome [21]. Interestingly, however, ALK-positive ALCL have been shown to have distinct molecular signatures according to its morphologic features [22]. Lamant et al. found 248 genes that were overexpressed in morphologic variants compared with common pattern ALCL, including genes involved in cell adhesion and migration [22]. In this series, patients with a morphologic variant of ALCL had advanced-stage disease, and a significant proportion of them experienced early relapse [22].

Anaplastic Lymphoma Kinase Protein Expression

In normal tissues, the ALK protein function is still somewhat obscure, although its expression in neural and endothelial cells, and pericytes suggest a role in neurodevelopment [23]. When rearranged, the entire *ALK* intracellular domain at the 3′-end is fused to various 5′-end partner genes, leading to ligand-independent constitutive activation of the downstream ALK pathway. The most recurrent *ALK* partner in ALCL is *NPM1* [10]. Other partners include tropomyosin 3 (*TPM3*, 1q25) and less commonly TRK-fused gene (*TGF*, 3q21) and clathrin heavy chain like 1 (CLTC, 17q23), among others [10]. The different ALK fusion proteins seem to have a direct oncogenic effect as their transfection into xenograft models led to tumor development via multiple oncogenic mechanisms [10, 11]. Immunohistochemistry is routinely used to detect ALK expression in lymphoid neoplasms [24]. Its subcellular labeling pattern is informative as nuclear and diffuse cytoplasmic ALK staining is typical of NPM-ALK versus other patterns such as granular cytoplasmic (CTCL-ALK) or diffuse cytoplasmic (other fusion proteins) [24]. Translocations involving the *ALK* gene are usually detected by different techniques, including conventional cytogenetics, fluorescent in situ hybridization (FISH), or polymerase chain reaction (PCR) [25, 26]. There is no relationship between different ALK fusion proteins and morphologic subtypes [2].

ALK expression has been recognized as having a prognostic value in ALCL. In 1995, Shiota et al. retrospectively analyzed 105 ALCL cases using immune-stained paraffin-embedded sections with ALK antibody [27]. They not only found that ALK-positive cases were more common among children and adolescents with lymphoma but also a sharp contrast in outcome, with a 5-year survival rate of 80% among ALK-positive cases, versus 33% in the ALK-negative group [27]. Subsequent cohort of adult patients with ALCL confirmed the favorable outcome of ALK-positive cases, with long-term overall survival superior to 70% [28–30].

The poor prognosis associated with lack of ALK expression in ALCL is less understood. ALK-positive and ALK-negative ALCL have highly similar genome-wide DNA methylation profiles for genes involved in T-cell differentiation and immune response [31]. Until recently, no recurrent cytogenetic abnormality had been identified in ALK-negative ALCL cases. However, the use of next-generation sequencing uncovered the presence of significant genetic heterogeneity, including the presence of numerous rearrangements [32, 33]. About 30% of adults with ALK-negative cases have rearrangements involving the *DUSP22* (dual-specificity phosphatase) and *IRF4* (interferon regulatory factor 4) locus (6p25.3), and 10% have abnormalities involving a p53-related gene, *TP63* (localized to 3q28) [33–35]. Lymphoma cases having ALK-negative expression and *DUSP22* rearrangements have survival curves similar to ALK-positive patients, while patients with ALK-negative and *TP63* rearrangements have guarded outcomes [35–37]. The presence of *DUSP22*, *TP63* rearrangements, or any other form of predictive biomarker has not been described in pediatric ALK-negative ALCL.

Immunophenotyping

ALK-positive ALCL cells express T-cell antigens and have T-cell receptor gene rearrangements, including cytotoxic molecules (perforin and/or granzyme B) [38]. A smaller proportion of ALK-positive cases do not express T-cell antigens (null phenotype) [38]. The large presence of inflammatory cells in the tissue makes the immunophenotype characterization of this lymphoma challenging, making the association between specific immunophenotypic expression and outcome difficult to prove [16]. In a large cohort of pediatric patients with ALK-positive ALCL, the investigators used immunofluorescence multi-staining combining antibodies for ALK to specifically identify lymphoma cells with antibodies against CD30, CD3, CD5, CD8, Ki67, and phosphorylated STAT3 [39]. CD8 expression was more frequent among non-common patterns of ALCL, and it was associated with a poorer outcome. Interestingly, CD8 expression was independently associated with prognosis in a multivariate analysis (hazard ratio for survival 3.38, $p = 0.042$) [39]. Expression of CD56, a cell adhesion molecule (OMIN 116930) normally expressed in NK cells and in a small proportion of T cell, has been detected in ALCL cases, more specifically in morphologic variant subtypes [29, 38]. CD56 expression has been investigated as a potential prognostic marker in ALCL. In a cohort of 143 patients (58% ALK positive), 25 (18%) expressed CD56. This subgroup of patients had a significantly inferior outcome overall in both ALK-positive and ALK-negative cases, and CD56 expression was

independently associated with prognosis by multivariate analysis [29]. No correlation with outcome was found in a pediatric cohort of patients, but the number of CD56-positive cases was very small (7% of the cases) [40].

In addition, activation and subsequent cytoplasmic expression of the signal transduce and activator of transcription 3 (stat3) has been demonstrated in ALCL [40]. Persistent activation of stat3 signaling induces survivin and tissue inhibitor of metalloprotease 1 (TIMP1) gene expression and apoptosis inhibition in cancer [41, 42]. Survivin expression has been detected in about half of ALCL sample tumors and has been independently correlated with worse prognosis in ALCL [42]. In a cohort of pediatric patients from the Pediatric Oncology Group with predominantly ALK-positive ALCL, stat3 activation was demonstrated in the majority of tumors. However, cytoplasmic localization of survivin and TIMP1 was not frequent, suggesting that these features may explain the good prognosis of pediatric ALCL [40].

Disease Staging and Clinical Presentation

Most patients with ALCL present with B symptoms and extranodal involvement [12–15, 19, 24, 29, 43, 44]. Pediatric lymphomas are usually staged using the St. Jude staging system, instead of the Ann Arbor staging system, widely used in adult non-Hodgkin lymphoma (NHL) [45, 46]. The St. Jude staging system was developed taking into account the fact that children with NHL most commonly present with disseminated disease including extranodal, central nervous system (CNS), BM involvement by morphology, and noncontinuous spread of the disease [47]. Interestingly, certain clinical features at presentation seem to be related to risk of progression and prognosis in ALCL. However, there is variation among different studies of which clinical features are of prognostic relevance in ALCL. In a large study of pediatric ALCL conducted by the European Intergroup for Childhood Non-Hodgkin Lymphoma, St. Jude stage III or IV (presence of extensive intra-thoracic or intra-abdominal disease, presence of disease in both sides of the diaphragm, presence of central nervous system (CNS), and/or BM involvement by morphology) was significantly associated with visceral involvement but was independent of skin lesions [43]. Ann Arbor stage III or IV (presence of disease in both sides of the diaphragm or presence of any extralymphatic organ involvement) was by definition associated with visceral involvement but was not significantly associated with mediastinal involvement [43]. B symptoms; mediastinal mass; spleen, liver, or lung involvement; skin lesions; elevated LDH; and advanced stage (by Ann Arbor or St. Jude) significantly increased the risk of relapse in univariate analysis [43]. However, in multivariate analysis, only visceral involvement (lung, liver, or spleen), skin lesions, and mediastinal mass correlated with risk of progression/relapse. Advanced stage had no additional prognostic value when the three factors were taken into account [43]. Mediastinal mass, any visceral involvement, and elevated LDH were also associated with high risk of failure among 82 children treated on two consecutive studies conducted by the French Society of Pediatric Oncology [19]. In fact, the combination of those three parameters allowed the definition of two risk groups: patients who presented with none of those features had a very high 3-year event-free survival (EFS; 95%, 75–99%) versus those with at least one feature who had significantly reduced 3-year EFS (47%, 32–62%). Staging at presentation also correlated with outcome (stage I and II, 3-year EFS 94%, 74–99%; stage III and IV, 3-year EFS 55%, 41–68%) [19]. The prognostic significance of skin involvement was not demonstrated in the French cohort [19]. The United Kingdom Children's Cancer Study Group found only visceral involvement and mediastinal involvement to correlate with prognosis among 72 children with ALCL [48]. Among 89 pediatric patients treated with short pulse B-NHL-type chemotherapy, only presence of B symptoms at presentation was proved to be significantly associated with high risk of failure [14]. Bone marrow involvement by morphology was the only predictor of poor EFS in another cohort of 86 pediatric ALCL patients treated with intensive chemotherapy (by univariate analysis only) [13]. The impact of CNS disease in the outcome of pediatric ALCL is less understood as those patients are either excluded from the analysis or are present in very small numbers [13, 14, 19, 43, 48].

Bone marrow involvement in ALCL is considered uncommon at presentation, although conventional BM evaluation for lymphoma detection lacks sensitivity in this subtype of lymphoma [49]. The presence and extent of circulating lymphoma cells in the BM and peripheral blood (minimal disseminated disease, MDD) by reverse transcriptase (RT)-PCR or flow cytometry are more sensitive and specific as diagnostic tools and have also been evaluated for possible prognostic significance in ALCL. In 2005, the Italian Association of Paediatric Haematology and Oncology (AIEOP) used RT-PCR to validate this technique as a valuable tool to detect MDD [50]. With a level of detection of 10^{-6} tumor cells, the authors detected presence of MDD in 61% of ALCL patients at diagnosis, while conventional microscopy detected presence of marrow disease in only 15% of the cases [50]. In another cohort of 80 pediatric patients with ALCL, lymphoma cells were detected in the BM by PCR for NPM1/ALK transcripts in nearly half of the patients, and findings significantly correlated with clinical stage, mediastinal and visceral involvement, and histologic subtype (non-common subtypes) [20]. There was a strong correlation between detection of lymphoma cells in the BM and peripheral blood, and BM PCR positive was associated with a higher cumulative incidence of relapse. Detection of MDD by quantitative PCR in BM or peripheral blood allowed the identification of

20% of patients experiencing 60% of all relapses, representing the group with the lowest EFS in that cohort [20]. In addition, there seems to be a good correlation between detection of MDD by PCR and flow cytometry and CD30 or ALK expression in ALCL [51]. Interestingly, the combined use of MDD by RT-PCR and anti-ALK immune response (see Figs. 11.3 and 11.4) has been used for risk stratification in a cohort of 128 pediatric patients with ALK-positive ALCL [52]. Patients with MDD-positive and antibody titer ≤1/750 had a progression-free survival (PFS) of only 28% (biological high-risk group), while patients with MDD-negative and antibody titer >1/750 (biological low-risk group) and all remaining (biological intermediate risk group) had PFS of 93% and 68%, respectively [52]. Larger studies are necessary to confirm if detection of MDD can be used to stratify newly diagnosed patients.

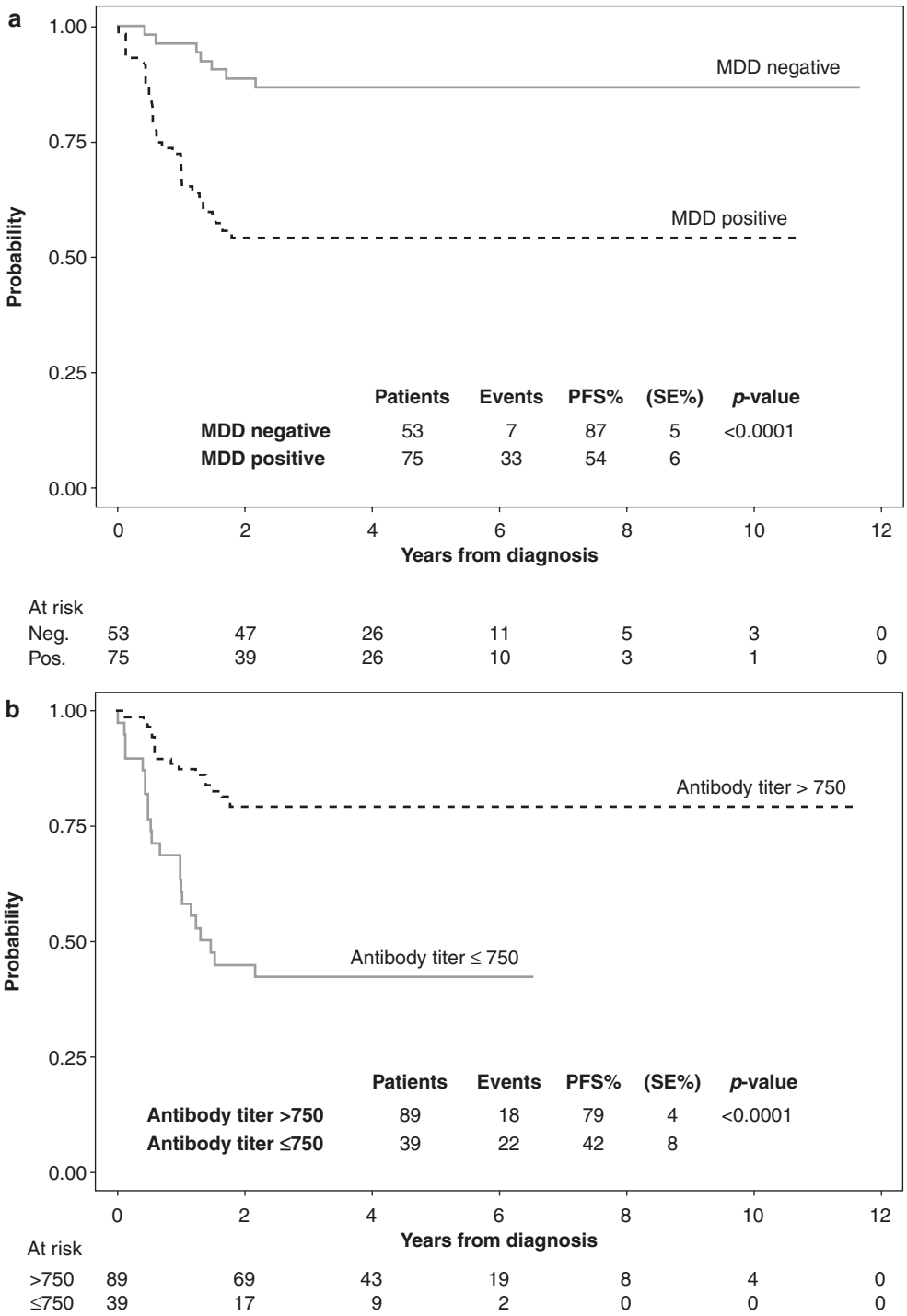


Fig. 11.3 Five-year progression-free survival of patients with NPM-ALK-positive ALCL according to (**a**) MDD in blood or BM and (**b**) anti-ALK antibody titer at diagnosis. (Reprinted from Mussolin et al. [52], with permission)

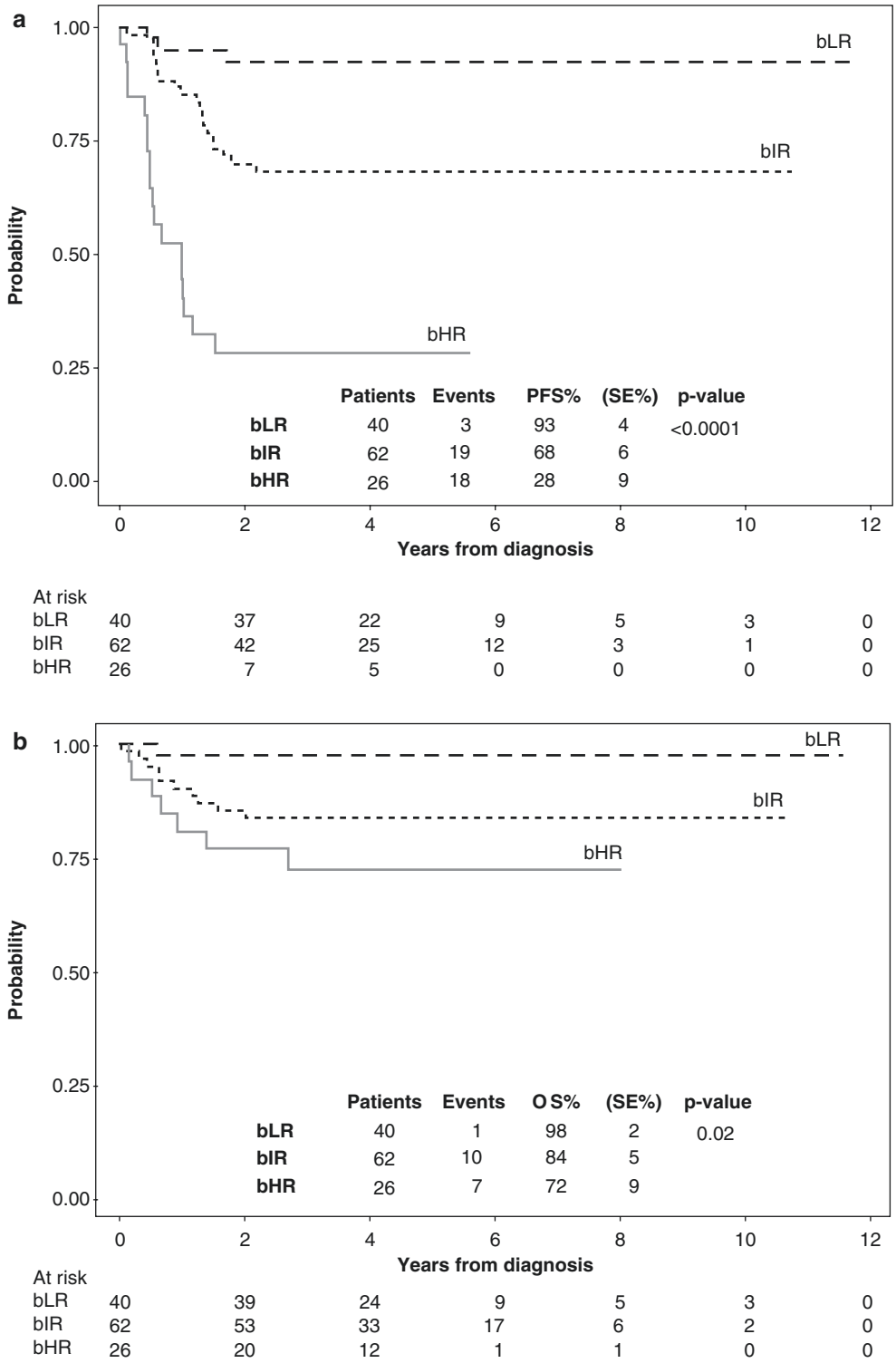


Fig. 11.4 Five-year progression-free survival (**a**) and overall survival (**b**) of patients with NPM-ALK-positive ALCL according to the combination of anti-ALK antibody titer and MDD at diagnosis: biological high risk (bHR), MDD-positive and antibody titer ≤1/750; biological intermediate risk (bIR), MDD-negative and antibody titer ≤1/750 or MDD-positive and antibody titer >1/750; biological low risk (bLR), MDD-negative and antibody titer >1/750. (Reprinted from Mussolin et al. [52], with permission)

Host-Related Prognostic Factors

Age, Gender, and Ethnicity

Age may be an important factor contributing to survival differences between ALK-positive and ALK-negative cases. ALK-positive ALCL mostly occurs in the first three decades of life with most of the affected patients being between 10 and 19 years of age at presentation [24, 28, 30, 32, 53]. ALK-negative ALCL patients are typically much older with peak incidence around the sixth decade of life [24, 28, 30, 32, 53]. In addition, survival was similar between ALK-positive and ALK-negative patients with higher international prognostic

index (IPI; one point assigned for each category: age greater than 60 years; stage III or IV disease; elevated serum LDH; Eastern Cooperative Oncology Group (ECOG) performance status of 2, 3, or 4; and more than one extranodal site) [30]. The majority of pediatric patients with ALCL have expression of ALK protein. In a large cohort of childhood ALCL, ALK-negative lymphoma accounted for only about 10% of the cases [43]. In analyzing risk of progression or relapse, the authors found no correlation with ALK positivity by multivariate analysis, suggesting that age indeed is driving the outcome discrepancies in this condition [43]. ALCL prognosis does not seem to correlate with gender [13]. A Surveillance Epidemiology and End Results (SEER) study of 1604 adult patients with ALCL found higher mortality among Blacks by multivariate analysis (HR, 1.37; 95% CI, 1.14–1.65; $p < 0.01$) [54].

Patient Immune Status

The development of an anti-tumor immune response with generation of anti-ALK autoantibodies and cytokine release has been demonstrated in ALK-positive ALCL patients [55–57]. Knorr et al. studied pretreatment concentrations of 25 cytokines in 119 pediatric patients with ALCL and found elevation of interleukin (IL)-9, IL-10, IL17a, hepatocyte growth factor, soluble IL-2 receptor, and soluble CD30 in the initial sera in comparison to the controls [58]. In addition, levels of IL-6, interferon-γ, interferon γ-induced protein, and soluble IL-2 receptor correlated with stage, initial general condition, MDD, ALK-antibody titers, and risk of relapse, suggesting a correlation between cytokine levels, tumor burden, and patient's immune response [58]. The greatest difference in survival was seen in patients with elevated levels of IL-6 vs. no detectable IL-6 (EFS 85.7% [95% CI 77.5–94.8] vs. 44.6% [95% CI 33.4–59.8], $p < 0.001$) and IL-6 independently correlated with prognosis in multivariate analyzes [58]. Additional studies are necessary to confirm those findings.

The prognostic significance of anti-ALK autoantibodies has also been investigated. Pretreatment levels of ALK autoantibodies were analyzed in 95 patients treated between 1996 and 2007 [59]. The ALK autoantibodies were detected in the majority of patients. Interestingly, the titers inversely correlated with stage and amount of circulating tumor cells. High antibody titers correlated with significantly lower cumulative incidence of relapse, suggesting that ALK autoantibodies may be a surrogate marker for the degree of immune-mediated destruction of lymphoma cells [59]. The combined use of biological markers such as MDD and ALK autoantibodies has been used to stratify pediatric ALCL patients according to risk of relapse, including patients with very low risk of relapse (Figs. 11.3 and 11.4) [52]. Interestingly, patients with high levels of ALK autoantibodies before treatment and whose levels persisted during and after completion of therapy represented a group of patients with higher EFS [60].

High amounts of soluble CD30 antigen have been demonstrated to be present in the serum of patients with CD30-positive lymphomas including ALCL [61–63]. In a cohort of 24 ALCL cases, soluble CD30 antigen values returned to normal range in patients in complete remission (CR), while it remained elevated in one patient who achieved only partial remission (PR). Subsequent increases in levels of soluble CD30 were subsequently detected in patients who relapsed [63]. Pretreatment levels of soluble CD30 were significantly higher in patients with ALCL in comparison to patients with Hodgkin lymphoma or normal controls and correlated with lower relapse-free survival rates, suggesting that soluble CD30 antigen is of prognostic significance in ALCL [64].

T-Cell Lymphoblastic Lymphoma

Lymphoblastic (or precursor) lymphoma (LL) is one of the most common types of pediatric lymphomas, with T-cell subtype being more common than the B-cell counterpart in children [1, 3]. T-cell LL (T-LL) is characterized by small- to medium-sized lymphoblast cells with scant cytoplasm, moderately condensed to dispersed chromatin and inconspicuous nucleoli [3]. T-LL is believed to originate in the thymus with potential to disseminate to any tissue including the bone marrow [65]. In fact, T-LL and T-cell acute lymphoblastic leukemia (T-ALL) are morphologically considered the same disease differentiated by the degree of bone marrow involvement (≥25% lymphoblast bone marrow infiltration defines ALL). The immature nature of those neoplastic cells is defined by the expression of non-lineage-specific markers, such as TdT, CD99, CD34, or CD1a, while the lineage of origin is defined by variable expression of T-cell markers such as CD2, CD3, CD4, CD5, CD7, and CD8 [3]. In fact, cell marker expression patterns can indicate the stage of T-cell differentiation from which the neoplasm originated: [1] early or pro-T cell (cytoplasmic CD3+, CD7+, CD2-, CD1a-, CD4-, CD8-, and CD34±), [2] pre-T (cytoplasmic CD3+, CD7+, CD2+, CD1a-, CD4-, CD8-, and CD34±), [3] cortical-T (cytoplasmic CD3+, CD7+, CD2+, CD1a+, CD4+, CD8+, and CD34-), and [4] medullary (cytoplasmic CD3+, CD7+, CD2+, CD1a-, CD4±, CD8+, and CD34- and surface CD3+) [65]. Myeloid markers such as CD13 and CD33 can also be expressed in up to 30% of the cases, as well as NK-related antigens such as CD16 and CD57 [3]. The genetic background of T-LL is complex. In over half of the cases, abnormal karyotype with pseudodiploidy, hypodiploidy, chromosomal deletions, or translocations can be found [66]. Abnormalities involving chromosome 14 (to include the T-cell receptor alpha [TCRA]/delta [TCRD]), in addition to abnormalities involving chromosomes 9, 10, and

Table 11.2 Prognostic factors in children and adolescents with T-cell lymphoblastic lymphoma

Prognostic factor	Impact	Risk/outcome	Frequency	References
Biology-related				
NOTCH1/FBXW7 mutation	Favorable	pEFS[b] 84% vs. 66%, $P < 0.021$	60%	[72]
LOH6q (FLASH) mutation	Unfavorable	pEFS 27% vs. 86%, $P < 0.001$	12%	[72]
TCR rearrangements[c]	Unfavorable	$P = 0.02$	7%	[75]
PTEN mutations	Unfavorable	pEFS 59% vs. 82%, $P = 0.014$	15%	[78]
PIK3R1/PIK3CA mutations	Unfavorable	pEFS 64% vs. 82%, $P = 0.025$	4%/6%	[78]
Treatment-related				
MDD[a]	>1% unfavorable	EFS 68.1% vs. 90.7%; $P = 0.031$	Variable	[86]

[a]*MDD* minimal disseminated disease detected by flow cytometry
[b]*pEFS*: probability of event-free survival
[c]Biallelic TCRγ deletion

11, are present in the majority of T-LL cases [65]. Similar to clonal rearrangements involving immunoglobulin (Ig) heavy and light chain in B-LL, T-LL is also characterized by T-cell receptor (TCR) clonal gene rearrangements [67]. Interestingly, additional analysis of the genetic background of T-LL has exposed distinctive patterns from T-ALL. Gene expression profile studies have suggested the presence of a signature profile for pediatric patients with T-LL with upregulation of genes that are associated with angiogenesis and chemotaxis, among other alterations [68–71].

Differently from T-ALL, a few clinical or laboratory features have been found to be of prognostic significance in pediatric T-LL. In the following section, we will review the current knowledge in prognostication of T-LL (Table 11.2).

Biology-Related Prognostic Factors

Genetics

Activating mutations involving *NOTCH1* (localized to 9q34.3; OMIN 190198) and *FBXW7* (localized to 4q31.3, OMIN 606278) has been described in patients with T-cell malignancies [72]. In fact, activation of *NOTCH1*, a gene encoding a transmembrane receptor that regulates normal T-cell development, has been directly implicated in the pathogenesis of T-ALL, while inactivation of *FBXW7* causes premature depletion of hematopoietic cells and development of T-cell leukemia [73, 74]. In T-ALL, presence of *NOTCH1* and/or *FBXW7* has been associated with good prognosis [75]. A cohort of 116 pediatric patients with T-LL treated with Berlin-Frankfurt-Münster (BFM)-type treatment were analyzed for *NOTCH1* and *FBXW7* mutations and their prognostic significance [72]. *NOTCH1* mutations were found in 60% of the patients and were associated with favorable prognosis [probability of EFS (pEFS) 84% ± 5% vs. 66% ± 7%, $p = 0.021$]. *FBXW7* mutations were found in 18% of the cohort, but only *NOTCH1* mutation was independently associated with prognosis [72].

In addition, the same BFM cohort of patients was evaluated for the presence of loss of heterozygosity (LOH) at chromosome 6q (LOH6q). Presence of LOH6q has been linked to possible deletion of *FLASH* (localized to 6q15–16.1), a gene that encodes an apoptosis-mediating protein determinant of glucocorticoid signaling [75]. Presence of LOH6q had been previously associated with higher risk of relapse in children with T-LL, while *FLASH* mutations have been associated with poor prognosis in T-ALL [75, 76]. LOH6q was found in 12% of the cases and was associated with a significantly inferior 5-year pEFS (27% ± 9% compared with 86% ± 3% for LOH6q-negative cases, $p < 0.0001$) [72]. Interestingly, LOH6q was rarely found among patients with *NOTCH1* mutations [72]. Another study investigating the frequency and prognostic value of *NOTCH1/FBXW7*, *FLASH* deletions, and *TCR* rearrangements in pediatric T-LL also found *NOTCH1/FBXW7* to be associated with improved EFS and OS [75]. FLASH mutations were found in 18% of the patients and were associated with inferior prognosis [75]. In a cohort of adult patients with T-LL, analysis of *NOTCH1/FBXW7*, *RAS/PTEN*, and *TRC* rearrangement (four-gene oncogenetic classifier) was found to be an independent prognostic indicator of EFS, OS, and disease-free survival (DFS) [77].

Balbach et al. studied the prognostic relevance of *NRAS*, *KRAS*, *PTEN*, *PIK3R1*, and *PIK3CA* mutations in a cohort of 114 pediatric T-LL treated as per NHL-BFM [78]. PTEN mutations were found in 15% of the patients, while *NRAS*, *KRAS*, *PIK3R1*, and *PIK3CA* mutations were present in 7%, 3%, 4%, and 6% of the cases, respectively. *PTEN* mutations were associated with poorer prognosis (pEFS 59% ± 12% vs. 82% ± 4%, $P = 0.014$). Interestingly, the prognostic impact of mutations involving PI3K-AKT pathway (other than *PTEN* mutations) was weaker (pEFS 64% ± 10% vs. 82% ± 4%, $P = 0.025$). In addition, NRAS and KRAS mutations did not have a statistically significant impact on pEFS (Fig. 11.5) [78].

There is some evidence in the literature supporting an association between clinical course and *TCR* rearrangement displayed in the malignant clone. In analyzing 41 pediatric and adult T-LL samples, Baleydier et al. found 3 subgroups

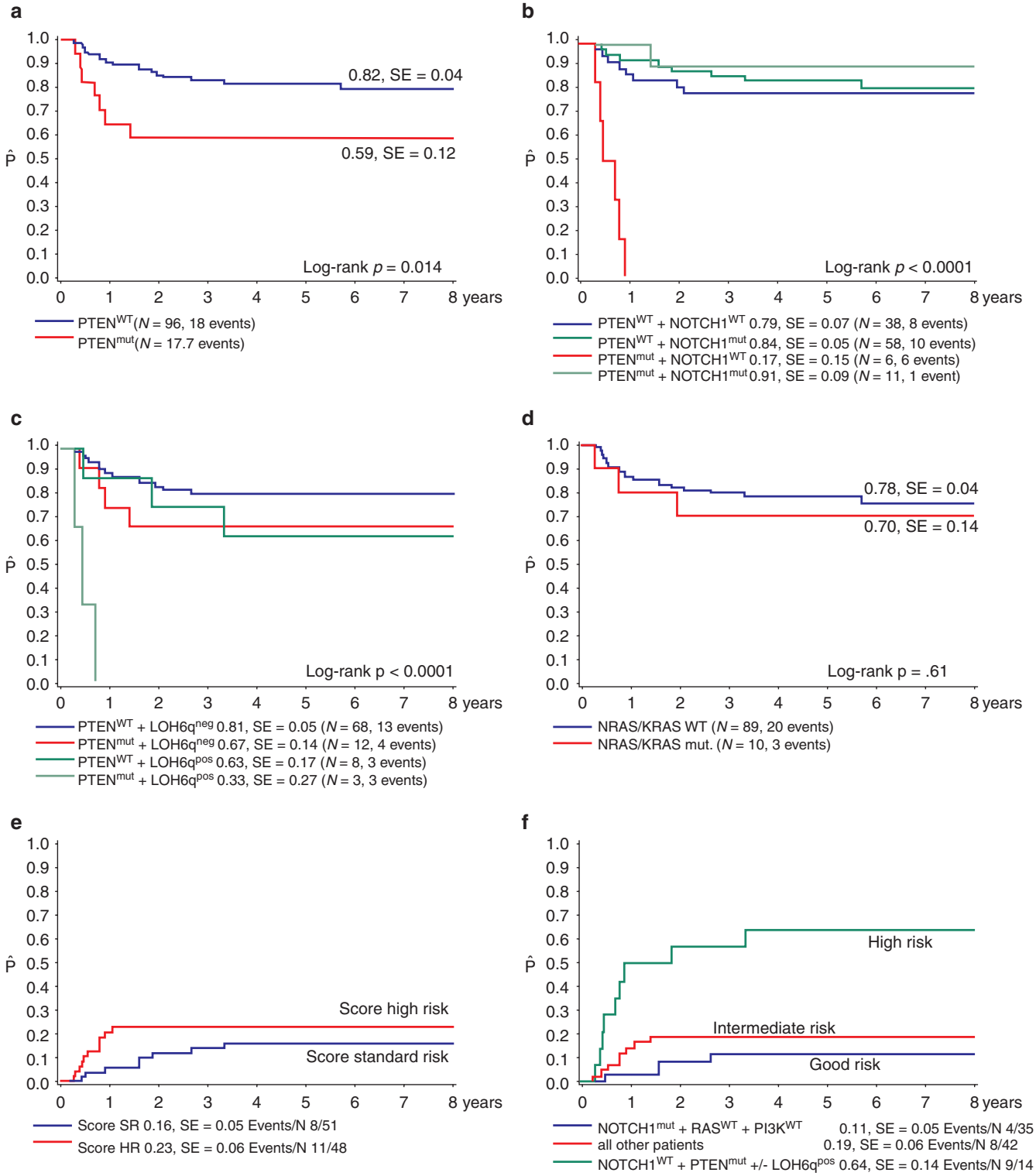


Fig. 11.5 Probability of event-free survival according to *PTEN* mutational status (**a**), to the combined genotype of *PTEN* and *NOTCH1* (**b**), to PTEN mutation and LOH6q (**c**), and to the mutational status of *NRAS* and *KRAS* (**d**). CIR according to the published classifier for adult T-ALL (**e**) and to the authors proposed classifier (**f**). (Reprinted from Balbach et al. [78], with permission)

of patients: "immature" group characterized by no TCR or incomplete TCRD rearrangement, "mature" group that showed biallelic TCRD deletion and both TCR gamma (TCRG) and TCR beta (TCRB) rearrangements (consistent with TCR alpha/beta lineage restriction), and an "intermediate" group with TCRD, TCRG, and TCRB rearrangements [79]. Overexpression of *HOX11/TLX1* and *HOXA9* was also found in the intermediate group [79]. TCR immature immunophenotyping was only found in adult patients with extrathymic disease and bone marrow involvement, while the other subsets were found among children and adults with predominantly thymic disease [79]. Adult patients (n = 6) with intermediate genotype had superior outcome; the addition of pediatric patients with intermediate subtype reinforced the improved OS [79]. In another cohort of pediatric T-LL, the absence of biallelic TCRD deletion was present in about 7% of the cases (all patients had *NOTCH1/FBXW7* mutations) and was associated with poor prognosis [75].

Clinical-Related Prognostic Factors

Disease Staging and Clinical Characteristics

Differently from other lymphoma subtypes, the large majority of patients with T-LL will present with advanced stage, making disease staging a not so helpful tool to stratify treatment. In a BFM cohort of 105 pediatric patients with T-LL, only 4 patients had either stage I or stage II [80]. Patients received ALL-type therapy, and the pEFS was 90% ± 3% and 95% ± 5% for patients with stage III and IV, respectively. Staging did not define a group with superior or inferior EFS [80]. Another study of almost 100 pediatric LL patients (80% T-LL, 10% null type, 10% B-LL) conducted at the Memorial Sloan-Kettering Cancer Center using LSA_2-L_2 type of therapy investigated the staging as a prognostic marker [81]. Patients with stage I–II disease (n = 8) were treated with a cumulative dose of 8400 mg/m^2 of cyclophosphamide and 240 mg/m^2 of daunorubicin for 2 years, while stage III or IV patients received up to 15,600 mg/m^2 of cyclophosphamide and 300 mg/m^2 of daunorubicin for 2–3 years (depending on the era of treatment) [81]. The OS and EFS rates for patients with stage I/II disease were 87% and 87%, respectively. Patients with stage III disease had an OS of 90% and EFS of 85%. Patients with stage IVA disease (bone marrow involvement <25%) had an OS and EFS of 79% and 73%, respectively, while patients with stage IVB (bone marrow involvement >25%) had OS of 74% and EFS of 70%. Univariate analysis did not link survival to presence of stage III/IV, in addition to other characteristics, such as age, gender, LDH level, duration of induction therapy, mediastinal involvement, primary tumor size, or phenotype [81]. Stage at diagnosis, LDH levels, and age did not influence the outcome of 27 adult patients with T-LL treated as per LMT-89 protocol [82]. However, bone marrow involvement was associated with a more guarded prognosis, with OS of 37% ± 30% vs. 85% ± 20% among patients without bone marrow involvement [82].

Treatment-Related Prognostic Factors

Speed of tumor resolution or assessment of early response to treatment has been investigated for potential prognostic significance. Among 101 evaluable pediatric patients with T-LL treated with ALL-type of therapy, 64 patients had a complete tumor response (TR), 35 patients had partial but 70% or more TR, and 2 patients had less than 70% TR [80]. Complete tumor response at day 33 of induction (in a 9-week induction course) was associated with a trend toward better survival with 5-year pEFS (95% ± 2%) in comparison with patients with residual tumor (5-year pEFS 89% ± 5%, p = 0.37) [80]. pEFS at 5 years was 95% ± 2% for the 80 patients with complete TR at the end of induction and 89% ± 5% for the 19 patients with residual tumor after completion of induction (p = 0.58), suggesting that incomplete tumor regression may not indicate poor prognosis [80]. However, another study investigating early resolution of mediastinal mass by chest radiography in pediatric T-LL found significant differences in both EFS and OS when the chest radiograph of 50 patients returned to normal within the 60 days of induction treatment compared with the 18 patients with persistent mediastinal mass [83]. In T-ALL patients, residual mediastinal mass by chest radiograph has not been shown to be of prognostic value [84].

More recently, assessment of early response to treatment by measuring levels of minimal residual disease (MRD) in BM or peripheral blood (PB) has become an important prognostic tool in the management of hematological malignancies. Stark et al. evaluated the feasibility of using real-time quantitative PCR (RQ-PCR) for TCR B/G/D gene rearrangement and flow cytometry (FC) in a cohort of 17 pediatric patients with stage III T-LL [85]. Bone marrow MDD at diagnosis of ≥0.01% was detected by FC and RQ-PCR in 88% and 80% of patients, respectively. MRD levels significantly decreased to very low levels on day 33 in nine out of ten patients studied. The only patient that remained MRD positive relapsed. Although a high prevalence of microscopic BM involvement was found, no prognostic correlation could be determined [85]. A larger series from Children's Oncology Group reported the use of FC to detect MDD (1 lymphoma cell among 10,000 normal cells) in the BM and PB of 99 children with T-LL at diagnosis and/or during therapy [86]. T-LL cells were detected in 71.7% of the BM samples obtained at diagnosis (level of detection from 0.01% to 31.3%); the majority of samples were from patients with stage II/III disease. Importantly, the 2-year EFS was 68% ± 11.1% for patients with ≥1% T-LL cells in BM vs. 90.7% ± 4.4% for those with lower levels of involvement (p = 0.031). The EFS for patients with ≥5% MDD was 51.9% ± 18% (p = 0.009), suggesting that MDD at diagnosis

may represent an important prognostic tool for T-LL patients [86]. More recently, similar results were found in a cohort of 65 pediatric patients with LL (T-LL in 52 patients) [87]. Using a MDD cutoff level of 3% by FC, 5-year EFS was 60% ± 22% for patients with MDD > 3% vs. 83% ± 6% for patients with lower levels of MDD (p = 0.04). No other analyzed clinical characteristic was found to be of prognostic significance [87].

Prognostic Factors in B-Cell Lymphomas

B-cell NHL is the most common subgroup of NHL in childhood and adolescence. Among B-NHLs, the most frequent histological subtypes are represented by Burkitt lymphoma (BL), diffuse large B-cell lymphoma (DLBCL), and primary mediastinal large B-cell lymphoma (PMBCL), which account for approximately 80%, 15%, and 5% of the cases, respectively [88–90]. Over the last few decades, B-NHL therapies have achieved substantial improvements in terms of survival. In the USA, SEER data have shown significant survival increase among NHL patients younger than 19 years: from 1975 to 2000, OS has improved from 42% to 79% in males and from 60% to 82% in females [91]. In some therapeutic groups, treatment has resulted in 100% survival, as reported by different studies [90, 92–94]. Stratification of treatment protocol in risk groups, defined on the basis of some clinical and pathological characteristics, was aimed not only at optimizing the treatment but also at reducing acute and long-term effects of chemotherapy, especially for patients with localized disease. To this goal, high-intensity and short-duration therapeutics blocks of chemotherapy were used, CNS prophylaxis was reduced, and cranial radiotherapy (RT) was abolished.

The therapeutic strategy of many international study groups, including BFM, Société Francaise d'Oncologie Pédiatrique (SFOP), the United Kingdom Children's Cancer Study Group (UKCCSG), Children's Cancer Group (CCG), and Associazione Italiana di Ematologia Oncologia Pediatrica (AIEOP), was to treat all subtypes of mature B-cell NHL with the same therapy, with the exception of PMBCL, a rather rare histology in children in which a worse prognosis has been demonstrated [88]. Thus, pediatric and adolescent patients with DLBCL and BL are currently treated with the same therapeutic regimens and are based on a stratified polychemotherapy strategy according to risk group. Although relapses usually occur at a later time in DLBCL, the outcome of patients with DLBCL was similar to the outcome of patients with BL. Many international studies had, in fact, confirmed that the histological subtype did not represent an unfavorable prognostic factor. Although the risk group definition is slightly different among study groups, the current overall strategy is based on the extent of the disease at diagnosis and the value of LDH [93, 95].

Complete staging is essential for optimal therapy. Numerous staging systems have been used for pediatric NHL over time. The St. Jude Children's Research Hospital staging system, described by Murphy in 1980 is the staging system currently used in the care of pediatric NHL patients [47]. It considers the sites commonly involved in pediatric NHL, i.e., extranodal involvement, metastatic spread to the BM and CNS, and involvement of noncontiguous sites. However, St. Jude staging system does not clearly define the extent of primary disease, especially if the disease is completely removed at diagnosis. To better define the stage of pediatric patients with NHL, a new risk group classification was subsequently proposed by French-American-British/Lymphoma Malins B (FAB/LMB) and subsequently adopted by CCG, SFOP, and UKCCSG [96–98]. This staging system expanded to include outcome from surgery in three groups A, B, and C. Other prognostic features used to stratify therapy include LDH levels at diagnosis, rapid response to therapy, and extension of disease (localized vs. disseminated), as in the risk classification used by the BFM and AIEOP groups [94, 99, 100].

In the last 30 years, the advances in the field of pediatric NHL diagnosis, especially through the use of cytogenetics and molecular and immunophenotypic characterization, as well as determination of the MDD and MRD, and the adoption of modern radiological techniques have highlighted additional limitations to the St. Jude staging system [101–105]. An international group of experts met in 2009 in Frankfurt, during the Third International Symposium of Childhood, Adolescent, and Young Adult Non-Hodgkin Lymphoma, to develop a new international staging system, the International Pediatric NHL Staging System (IPNHLSS) [106]. In the IPNHLSS, the authors recognized the prognostic role of some specific disease characteristics, including MDD, and suggested new techniques for the quantification of MDD in BM and cerebrospinal fluid (CSF) for some subtypes of NHL [107]. In the future, the IPNHLSS could allow the stratification of patients according to MDD levels, in order to potentially enhance therapy for high-risk and reduce toxicity for low-risk patients [106]. In this section, we will review the main prognostic factors in aggressive B-cell lymphomas (Table 11.3).

Preexisting Diseases

Constitutional molecular defects may play a role in oncogenesis. In particular, in patients with immunodeficiency, both congenital and acquired, there is an increased risk to develop B-NHL, and their outcome resulted to be worse than the patients without cancer predisposition syndrome [108]. Intensity of therapy should be adjusted to individual risk factors and tolerance.

Histology and Biology

No differences in survival have been observed between BL and DLBCL patients. Patients have been treated with the same treatment protocols. The exception is represented by

Table 11.3 Prognostic factors in children and adolescents with B-cell non-Hodgkin lymphomas

Prognostic factor	Impact	Risk/outcome	Frequency	References
Preexisting diseases				
Immunodeficiency	Unfavorable	pEFS 37%	8.5–10%	[108]
Cancer predisposition syndrome	Unfavorable	pEFS 40%	8.5–10%	[108]
Histology/biology				
PMBCL subtype	Unfavorable	pEFS 70%	5%	[88]
7q gain	Unfavorable	pEFS 72.2% vs. 83.6%	15%	[112]
13q deletion	Unfavorable	pEFS 63.6% vs. 84.9%	14%	[112]
More than three cytogenetic abnormalities	Unfavorable	pEFS 72.1% vs. 87.4%	37%	[112]
TNF-308 (G → A) and LT-a + 252 (A → G) polymorphisms	Unfavorable	pEFS 81% vs. 92%, $P = 0.018$	35%	[113]
Increased LDH at diagnosis	Unfavorable	RFR 2.0 (95% CI 1.3–3.2) $P = 0.003$	41%	[114]
Age				
Adolescent	Unfavorable	RR 6.7 (95% CI 2.2–20.4), $P = 0.01$	5.3%	[98]
Primary site at diagnosis				
CNS and/or BM involvement	Unfavorable	pEFS 60% vs. 81% $P = 0.001$	8.8%	[115]
Mediastinal involvement	Unfavorable	RFR 4.5 (95% CI 1.2–17) $P = 0.012$	2–19%	[114]
MDD at diagnosis	Unfavorable	HR 2.6 (95% CI 1.1–6.5)	30%	[94]
Response to CT				
Poor response after pre-phase	Unfavorable	RR 12.3 (3.2–47.1), $P = 0.006$	4.4%	(98)

pEFS probability of event-free survival, *PMBCL* primary mediastinal B-cell lymphoma, *LDH* lactate dehydrogenase, *RFR* relative failure rate, *CNS* central nervous system, *BM* bone marrow, *MDD* minimal disseminated disease, *HR* hazard ratio, *RR* relative risk, *CT* computed tomography

PMBCL subtype that has an inferior outcome with the standard B-cell treatment regimens. The rarity of PMBCL has not allowed identification of prognostic factors in pediatric patients [88]. Phenotypically, pediatric DLBCL has a higher proliferation index and is more frequently positive for MYC, CD10, and BCL6 expression in comparison to adult DLBCL [95, 109]. This profile justified the clear prevalence of GBC (germinal center B-cell) forms with GBC/ABC (activated B-cell like) ratio variable ration from 3:1 to 5:1 [95, 110]. Unlike in adults, the stratification GBC vs. ABC did not seem to have a prognostic impact, with excellent survival curves for both forms [111].

Few data are available on cytogenetic abnormalities in childhood B-NHL and their prognostic value. The major review on this topic was performed by GAB/LMB International Study Committee which highlighted that the BL and DLBCL gain of 7q or deletion of 13q and more than four abnormalities appeared to have an inferior EFS [112]. The BFM group showed that the TNF-308 (G → A) and the lymphotoxin alpha [LT-a + 252 (A → G)] polymorphism had a negative impact on the outcome of BL (Fig. 11.6) [113].

Age at Diagnosis

The role of age at diagnosis still remains controversial. Some studies highlighted that adolescents had outcomes inferior to those of younger children [89, 98, 100, 114]. However, for patients with BL who were treated on the FAB/LMB-96 clinical trial, adolescent age (≥15 years) was not a predictor for poor outcome. In the AIEOP LNH-97 for B-NHL, both in BL and DLBCL, the age was not confirmed to be of prognostic value [94].

Primary Site of Disease

Patients with localized disease have very good prognosis, whereas patients with advanced disease (stage III and IV) have outcomes that still need to be improved. In pediatric NHL literature, it is possible to find evidence that some sites of disease appear to have prognostic value (Table 11.4) [114].

Central Nervous System and Bone Marrow Involvement

The reported percentage of CNS-positive patients with BL/B-ALL and DLBCL was 8.8% and 2.6%, respectively [115]. Many studies have shown inferior outcomes in patients with CNS involvement, alone or in combination with BM involvement, for newly diagnosed pediatric B-NHL. BFM group analyzed the data of three consecutive trials (NHL-BFM86, NHL-BFM90, and NHL-BFM95) and pointed out that CNS-positive patients with BL/B-ALL had worse outcome than CNS-negative patients with stage IV BL/B-ALL (60% vs. 81%, respectively) [115]. In the LMB89 protocol, among the 123 patients belonging to risk group C, the only bad prognostic factor was CNS involvement [98]. Collaborative FAB/LMB96 study highlighted that patients with B-NHL with combined BM/CNS involvement had a significantly inferior EFS (60%) compared to patients with isolated CNS disease at presentation (83%) [96].

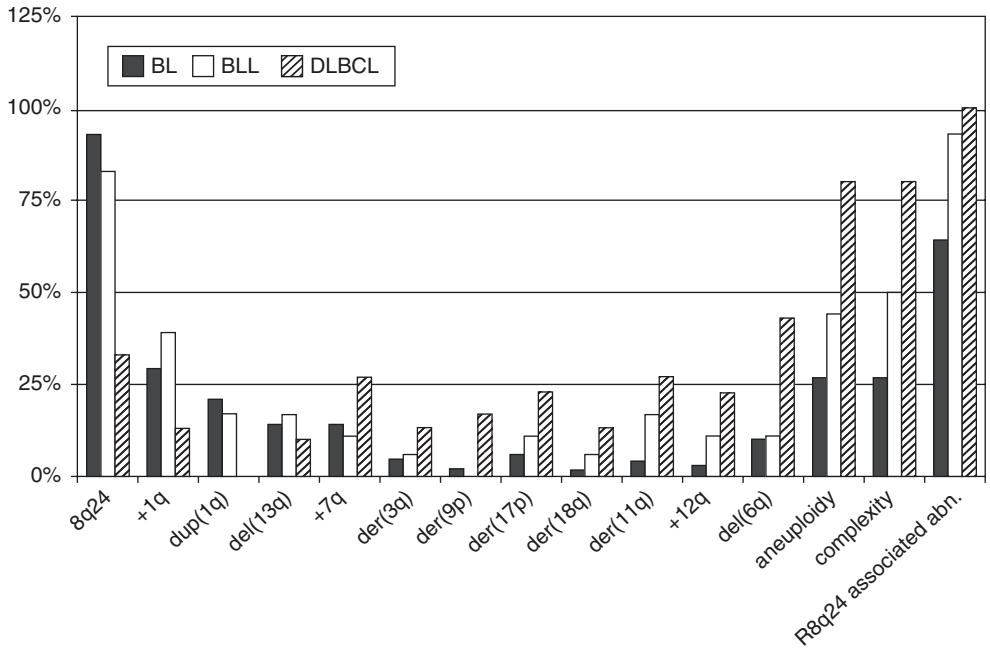


Fig. 11.6 Prognostic significance of individual cytogenetic abnormalities. (Reprinted from Poirel et al. [118], with permission)

Table 11.4 Significant risk factors associated with relapse/progression on FAB/LMB96 Study: univariate and multivariate analysis

	Univariate analysis	Multivariate analysis			
Risk factor	3-year EFS (% ± SE)	Log-rank P	RFR	95% CI	*P*
Age, years		0.15			0.58
<15	89 ± 1.0		1.0		
≥15	84 ± 3.4		1.2	0.70–1.9	
Prognostic group		<0.001			0.90
A	99 ± 0.75		1.0		
B	89 ± 1.2		2.0	0.38–11	
C	79 ± 2.7		2.6	0.36–19	
Stage (Murphy)		<0.001			0.082
I/II	98 ± 1.1		1.0		
III/IV	84 ± 1.4		2.4	0.90–6.4	
Primary site		<0.001			0.012
Peripheral node	97 ± 2.0		1.0		
Mediastinal	72 ± 6.2		4.5	1.2–17	
Abdominal/retroperitoneal	87 ± 1.4		2.7	0.83–9.0	
Head and neck	94 ± 2.0		1.2	0.32–4.4	
Other	85 ± 2.8		1.2	0.35–4.3	
Pathology		0.92			0.24
BL/BLL	89 ± 1.1		1.0		
DLBCL	87 ± 2.5		1.6	0.92–2.7	
Other	87 ± 4.2		1.0	0.49–2.1	
BM/CNS		<0.001			
BM −/CNS −	91 ± 1.1		1.0		<0.001
BM +/CNS −	88 ± 2.6		1.1	0.43–2.7	
BM −/CNS +	83 ± 5.6		1.8	0.50–6.6	
BM +/CNS +	61 ± 6.0		4.9	1.6–15	
LDH		<0.001			0.003
<2 × institutional ULN	94 ± 1.1		1.0		
≥2 × institutional ULN	81 ± 1.9		2.0	1.3–3.2	

FAB/LMB96 French-American-British Mature B-Cell Lymphoma 96, *SE* standard error, *RFR* relative failure rate, *CI* confidence interval, *BL/BLL* Burkitt lymphoma/Burkitt-like lymphoma, *DLBCL* diffuse large B-cell lymphoma, *BM* bone marrow, *CNS* central nervous system, *ULN* upper limit of normal

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Mediastinum

Patients with large mediastinal masses are at risk of complications related to compression of the trachea, upper vena cava syndrome, and pleural and/or pericardial effusions. Among B-cell NHL patients, the vast majority that presented primary mediastinal involvement at diagnosis were in fact PMBCL [88, 116]. In the most recent NHL-BFM95 study, the 3-year EFS once again showed inferiority of outcome for this subgroup of patients (53%). Also in the FAB/LMB96 international protocol, patients with PMBCL showed lower survival than patients with non-mediastinal DLBCL (4-year EFS 72% vs. 93%, respectively) [97, 114]. A small percentage of pediatric B-NHL with mediastinal involvement at diagnosis is represented by BL subtype. Despite the rarity of mediastinal BL, the few published cohorts reported higher risk of failure compared to non-mediastinal BL [117].

Lactate Dehydrogenase (LDH) Levels

It is well known that LDH levels are of prognostic importance in B-cell lymphomas. LDH levels have been correlated with dissemination of disease and have been used to identify different risk groups among patients with same lymphoma stage. In recent decades, the role of LDH value has been evaluated by the major European groups. In particular, the BFM group in the NHL-BFM90 study divided the patients into three risk groups, R1, R2, and R3, based on the stage and LDH. For patients in the R3 group, a predictor of disease progression turned out to be an LDH pretreatment value of ≥1000 IU/L. This parameter was the basis for further stratification of the R3 group into two new risk groups, R3 and R4, in the NHL-BFM95 study [100]. The excellent results confirmed the prognostic role of LDH. The LMB89 trial, conducted from 1989 to 1996, stratified a total of 561 pediatric patients in 3 therapeutic risk groups (A, B, and C) based exclusively on the sites involved at diagnosis [98]. Compared to other international groups, the value of LDH was not considered in the assignment of the risk group. Although this study did not stratify therapy based on the LDH value, the results showed a lower 5-year EFS for patients with elevated LDH, i.e., > 2 times the upper limit of normal value (87% vs. 95%, $p < 0.001$). In multivariate analysis, the high value of LDH together with age ≥15 years and poor response to cytoreductive chemotherapy with COP (cyclophosphamide, vincristine, prednisone) were independent prognostic factors for a worse outcome in the LMB89 study [98]. In multivariate analysis performed on a cohort of 442 patients treated in the AIEOP LNH-97 study, the higher LDH value above median confirmed to be the only clinical parameter significantly associated with increased risk of failure [94].

Minimal Disseminated Disease (MDD)

BL is characterized by the presence of chromosomal translocations involving *MYC* on chromosome 8 and the immunoglobulin (Ig) heavy- or light-chain genes on chromosome 14, 22, or 2 [102]. The t(8;14)(q24;q32) chromosomal translocation is detectable in almost 75% of BL/B-ALL samples and thus can be used as a marker to study minimal BM infiltration and identify a poor prognostic subgroup among BL patients (Table 11.5) [50, 102–105]. Refer to Chap. 9 for more details on MDD in pediatric NHL.

Table 11.5 Univariate and multivariate analysis restricted to patients studied for MDD

Characteristics	Categories	# Pts	Events	5-year PFS % (SE%)	Univariate *p*-value	Multivariate *p*-value	Hazard ratio (95% CI)
Age	<7.9 years	64	7	89 (4)	0.20	n.s.	
	≥7.9 years	64	12	81 (5)			
Gender	Male	113	15	87 (3)	0.16	n.s.	
	Female	15	4	73 (11)			
Median LDH	≤1009 IU/L	64	6	91 (4)	0.08	n.s.	
	>1009 IU/L	64	13	79 (5)			
Stage	I + II	26	2	92 (5)	0.27		
	III + IV	102	17	83 (4)			
Risk group	R1 + R2	34	2	94 (4)	0.09	n.s.	
	R3 + R4	94	17	82 (4)			
BM involvement	Yes	10	3	70 (14)	0.14	n.s.	
	No	118	16	86 (3)			
CNS involvement	Yes	7	2	71 (17)	0.22		
	No	121	17	86 (3)			
MDD	Pos	39	10	74 (7)	0.03	0.04	2.6 (1.1–6.5)
	Neg	89	9	90 (3)			

MDD minimal disseminated disease, *SE* standard error, *CI* confidence interval, *ns* not significant, *LDH* lactate dehydrogenase, *BM* bone marrow, *CNS* central nervous system

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Response to Chemotherapy

Poor response to chemotherapy or refractory disease was associated with unfavorable OS and EFS. Patte et al. reported that poor response to cytoreductive chemotherapy with COP (defined as less than 20% tumor reduction) was associated with inferior outcome in multivariate analysis ($p = 0.006$) [98]. The FAB/LMB96 analysis of results confirmed that the probability of 4-year EFS was only 30% for non-responder patients [96, 97]. Refer to Chap. 8 for more details on response to therapy in pediatric NHL.

Conclusions

In the field of pediatric B-NHL, conventional diagnostic techniques had identified clinical significant prognostic markers such as CNS involvement at diagnosis, elevated LDH, cytogenetics, extent of disease (stage), and poor response after pre-phase chemotherapy. Recently, by adopting innovative diagnostic tools, it is possible to provide physicians with new important tools to stage and monitor those patients, such as MDD. International collaboration and additional studies on molecular biology and gene expression will improve the treatment of pediatric and adolescence NHL.

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References

1. Sandlund JT, Downing JR, Crist WM. Non-Hodgkin's lymphoma in childhood. N Engl J Med. 1996;334(19):1238–48.
2. Stein H, Foss HD, Durkop H, Marafioti T, Delsol G, Pulford K, et al. CD30(+) anaplastic large cell lymphoma: a review of its histopathologic, genetic, and clinical features. Blood. 2000;96(12):3681–95.
3. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, et al. WHO classification of tumours of haematopoietic and lymphoid tissues. IARC: Lyon; 2008.
4. Schwab U, Stein H, Gerdes J, Lemke H, Kirchner H, Schaadt M, et al. Production of a monoclonal antibody specific for Hodgkin and Sternberg-Reed cells of Hodgkin's disease and a subset of normal lymphoid cells. Nature. 1982;299(5878):65–7.
5. Horie R, Watanabe T. CD30: expression and function in health and disease. Semin Immunol. 1998;10(6):457–70.
6. Wright CW, Duckett CS. The aryl hydrocarbon nuclear translocator alters CD30-mediated NF-kappaB-dependent transcription. Science. 2009;323(5911):251–5.
7. Hirsch B, Hummel M, Bentink S, Fouladi F, Spang R, Zollinger R, et al. CD30-induced signaling is absent in Hodgkin's cells but present in anaplastic large cell lymphoma cells. Am J Pathol. 2008;172(2):510–20.
8. Kaneko Y, Frizzera G, Edamura S, Maseki N, Sakurai M, Komada Y, et al. A novel translocation, t(2;5)(p23;q35), in childhood phagocytic large T-cell lymphoma mimicking malignant histiocytosis. Blood. 1989;73(3):806–13.
9. Morris SW, Kirstein MN, Valentine MB, Dittmer KG, Shapiro DN, Saltman DL, et al. Fusion of a kinase gene, ALK, to a nucleolar protein gene, NPM, in non-Hodgkin's lymphoma. Science. 1994;263(5151):1281–4.
10. Chiarle R, Voena C, Ambrogio C, Piva R, Inghirami G. The anaplastic lymphoma kinase in the pathogenesis of cancer. Nat Rev Cancer. 2008;8(1):11–23.
11. Werner MT, Zhao C, Zhang Q, Wasik MA. Nucleophosmin-anaplastic lymphoma kinase: the ultimate oncogene and therapeutic target. Blood. 2017;129(7):823–31.
12. Rosolen A, Pillon M, Garaventa A, Burnelli R, d'Amore ES, Giuliano M, et al. Anaplastic large cell lymphoma treated with a leukemia-like therapy: report of the Italian Association of Pediatric Hematology and Oncology (AIEOP) LNH-92 protocol. Cancer. 2005;104(10):2133–40.
13. Lowe EJ, Sposto R, Perkins SL, Gross TG, Finlay J, Zwick D, et al. Intensive chemotherapy for systemic anaplastic large cell lymphoma in children and adolescents: final results of Children's Cancer Group Study 5941. Pediatr Blood Cancer. 2009;52(3):335–9.
14. Seidemann K, Tiemann M, Schrappe M, Yakisan E, Simonitsch I, Janka-Schaub G, et al. Short-pulse B-non-Hodgkin lymphoma-type chemotherapy is efficacious treatment for pediatric anaplastic large cell lymphoma: a report of the Berlin-Frankfurt-Munster Group Trial NHL-BFM 90. Blood. 2001;97(12):3699–706.
15. Brugieres L, Le Deley MC, Rosolen A, Williams D, Horibe K, Wrobel G, et al. Impact of the methotrexate administration dose on the need for intrathecal treatment in children and adolescents with anaplastic large-cell lymphoma: results of a randomized trial of the EICNHL Group. J Clin Oncol. 2009;27(6):897–903.
16. Lamant L, McCarthy K, d'Amore E, Klapper W, Nakagawa A, Fraga M, et al. Prognostic impact of morphologic and phenotypic features of childhood ALK-positive anaplastic large-cell lymphoma: results of the ALCL99 study. J Clin Oncol. 2011;29(35):4669–76.
17. Youd E, Boyde AM, Attanoos RL, Dojcinov SD. Small cell variant of anaplastic large cell lymphoma: a 10-year review of the All Wales Lymphoma Panel database. Histopathology. 2009;55(3):355–8.
18. Kinney MC, Collins RD, Greer JP, Whitlock JA, Sioutos N, Kadin ME. A small-cell-predominant variant of primary Ki-1 (CD30)+ T-cell lymphoma. Am J Surg Pathol. 1993;17(9):859–68.
19. Brugieres L, Deley MC, Pacquement H, Meguerian-Bedoyan Z, Terrier-Lacombe MJ, Robert A, et al. CD30(+) anaplastic large-cell lymphoma in children: analysis of 82 patients enrolled in two consecutive studies of the French Society of Pediatric Oncology. Blood. 1998;92(10):3591–8.
20. Damm-Welk C, Busch K, Burkhardt B, Schieferstein J, Viehmann S, Oschlies I, et al. Prognostic significance of circulating tumor cells in bone marrow or peripheral blood as detected by qualitative and quantitative PCR in pediatric NPM-ALK-positive anaplastic large-cell lymphoma. Blood. 2007;110(2):670–7.
21. Benharroch D, Meguerian-Bedoyan Z, Lamant L, Amin C, Brugieres L, Terrier-Lacombe MJ, et al. ALK-positive lymphoma: a single disease with a broad spectrum of morphology. Blood. 1998;91(6):2076–84.
22. Lamant L, de Reynies A, Duplantier MM, Rickman DS, Sabourdy F, Giuriato S, et al. Gene-expression profiling of systemic anaplastic large-cell lymphoma reveals differences based on ALK status and two distinct morphologic ALK+ subtypes. Blood. 2007;109(5):2156–64.
23. Iwahara T, Fujimoto J, Wen D, Cupples R, Bucay N, Arakawa T, et al. Molecular characterization of ALK, a receptor tyrosine

kinase expressed specifically in the nervous system. Oncogene. 1997;14(4):439–49.
24. Falini B, Pileri S, Zinzani PL, Carbone A, Zagonel V, Wolf-Peeters C, et al. ALK+ lymphoma: clinico-pathological findings and outcome. Blood. 1999;93(8):2697–706.
25. Perkins SL, Pickering D, Lowe EJ, Zwick D, Abromowitch M, Davenport G, et al. Childhood anaplastic large cell lymphoma has a high incidence of ALK gene rearrangement as determined by immunohistochemical staining and fluorescent in situ hybridisation: a genetic and pathological correlation. Br J Haematol. 2005;131(5):624–7.
26. Kalinova M, Krskova L, Brizova H, Kabickova E, Kepak T, Kodet R. Quantitative PCR detection of NPM/ALK fusion gene and CD30 gene expression in patients with anaplastic large cell lymphoma--residual disease monitoring and a correlation with the disease status. Leuk Res. 2008;32(1):25–32.
27. Shiota M, Nakamura S, Ichinohasama R, Abe M, Akagi T, Takeshita M, et al. Anaplastic large cell lymphomas expressing the novel chimeric protein p80NPM/ALK: a distinct clinicopathologic entity. Blood. 1995;86(5):1954–60.
28. Gascoyne RD, Aoun P, Wu D, Chhanabhai M, Skinnider BF, Greiner TC, et al. Prognostic significance of anaplastic lymphoma kinase (ALK) protein expression in adults with anaplastic large cell lymphoma. Blood. 1999;93(11):3913–21.
29. Suzuki R, Kagami Y, Takeuchi K, Kami M, Okamoto M, Ichinohasama R, et al. Prognostic significance of CD56 expression for ALK-positive and ALK-negative anaplastic large-cell lymphoma of T/null cell phenotype. Blood. 2000;96(9):2993–3000.
30. Savage KJ, Harris NL, Vose JM, Ullrich F, Jaffe ES, Connors JM, et al. ALK- anaplastic large-cell lymphoma is clinically and immunophenotypically different from both ALK+ ALCL and peripheral T-cell lymphoma, not otherwise specified: report from the International Peripheral T-Cell Lymphoma Project. Blood. 2008;111(12):5496–504.
31. Hassler MR, Pulverer W, Lakshminarasimhan R, Redl E, Hacker J, Garland GD, et al. Insights into the pathogenesis of anaplastic large-cell lymphoma through genome-wide DNA methylation profiling. Cell Rep. 2016;17(2):596–608.
32. Hapgood G, Savage KJ. The biology and management of systemic anaplastic large cell lymphoma. Blood. 2015;126(1):17–25.
33. Mereu E, Pellegrino E, Scarfo I, Inghirami G, Piva R. The heterogeneous landscape of ALK negative ALCL. Oncotarget. 2017;8(11):18525–36.
34. Vasmatzis G, Johnson SH, Knudson RA, Ketterling RP, Braggio E, Fonseca R, et al. Genome-wide analysis reveals recurrent structural abnormalities of TP63 and other p53-related genes in peripheral T-cell lymphomas. Blood. 2012;120(11):2280–9.
35. Parrilla Castellar ER, Jaffe ES, Said JW, Swerdlow SH, Ketterling RP, Knudson RA, et al. ALK-negative anaplastic large cell lymphoma is a genetically heterogeneous disease with widely disparate clinical outcomes. Blood. 2014;124(9):1473–80.
36. Pedersen MB, Hamilton-Dutoit SJ, Bendix K, Ketterling RP, Bedroske PP, Luoma IM, et al. DUSP22 and TP63 rearrangements predict outcome of ALK-negative anaplastic large cell lymphoma: a Danish cohort study. Blood. 2017;130(4):554–7.
37. Pasqualini C, Minard-Colin V, Saada V, Lamant L, Delsol G, Patte C, et al. Clinical analysis and prognostic significance of haemophagocytic lymphohistiocytosis-associated anaplastic large cell lymphoma in children. Br J Haematol. 2014;165:117.
38. d'Amore ES, Menin A, Bonoldi E, Bevilacqua P, Cazzavillan S, Donofrio V, et al. Anaplastic large cell lymphomas: a study of 75 pediatric patients. Pediatr Dev Pathol. 2007;10(3):181–91.
39. Abramov D, Oschlies I, Zimmermann M, Konovalov D, Damm-Welk C, Wossmann W, et al. Expression of CD8 is associated with non-common type morphology and outcome in pediatric anaplastic lymphoma kinase-positive anaplastic large cell lymphoma. Haematologica. 2013;98(10):1547–53.
40. Nasr MR, Laver JH, Chang M, Hutchison RE. Expression of anaplastic lymphoma kinase, tyrosine-phosphorylated STAT3, and associated factors in pediatric anaplastic large cell lymphoma: a report from the children's oncology group. Am J Clin Pathol. 2007;127(5):770–8.
41. Gritsko T, Williams A, Turkson J, Kaneko S, Bowman T, Huang M, et al. Persistent activation of stat3 signaling induces survivin gene expression and confers resistance to apoptosis in human breast cancer cells. Clin Cancer Res. 2006;12(1):11–9.
42. Schlette EJ, Medeiros LJ, Goy A, Lai R, Rassidakis GZ. Survivin expression predicts poorer prognosis in anaplastic large-cell lymphoma. J Clin Oncol. 2004;22(9):1682–8.
43. Le Deley MC, Reiter A, Williams D, Delsol G, Oschlies I, McCarthy K, et al. Prognostic factors in childhood anaplastic large cell lymphoma: results of a large European intergroup study. Blood. 2008;111(3):1560–6.
44. Gisselbrecht C, Gaulard P, Lepage E, Coiffier B, Briere J, Haioun C, et al. Prognostic significance of T-cell phenotype in aggressive non-Hodgkin's lymphomas. Groupe d'Etudes des Lymphomes de l'Adulte (GELA). Blood. 1998;92(1):76–82.
45. Carbone PP, Kaplan HS, Musshoff K, Smithers DW, Tubiana M. Report of the committee on Hodgkin's disease staging classification. Cancer Res. 1971;31(11):1860–1.
46. Lister TA, Crowther D, Sutcliffe SB, Glatstein E, Canellos GP, Young RC, et al. Report of a committee convened to discuss the evaluation and staging of patients with Hodgkin's disease: Cotswolds meeting. J Clin Oncol. 1989;7(11):1630–6.
47. Murphy SB. Classification, staging and end results of treatment of childhood non-Hodgkin's lymphomas: dissimilarities from lymphomas in adults. Semin Oncol. 1980;7(3):332–9.
48. Williams DM, Hobson R, Imeson J, Gerrard M, McCarthy K, Pinkerton CR, et al. Anaplastic large cell lymphoma in childhood: analysis of 72 patients treated on The United Kingdom Children's Cancer Study Group chemotherapy regimens. Br J Haematol. 2002;117(4):812–20.
49. Fraga M, Brousset P, Schlaifer D, Payen C, Robert A, Rubie H, et al. Bone marrow involvement in anaplastic large cell lymphoma. Immunohistochemical detection of minimal disease and its prognostic significance. Am J Clin Pathol. 1995;103(1):82–9.
50. Mussolin L, Pillon M, d'Amore ES, Santoro N, Lombardi A, Fagioli F, et al. Prevalence and clinical implications of bone marrow involvement in pediatric anaplastic large cell lymphoma. Leukemia. 2005;19(9):1643–7.
51. Damm-Welk C, Schieferstein J, Schwalm S, Reiter A, Woessmann W. Flow cytometric detection of circulating tumour cells in nucleophosmin/anaplastic lymphoma kinase-positive anaplastic large cell lymphoma: comparison with quantitative polymerase chain reaction. Br J Haematol. 2007;138(4):459–66.
52. Mussolin L, Damm-Welk C, Pillon M, Zimmermann M, Franceschetto G, Pulford K, et al. Use of minimal disseminated disease and immunity to NPM-ALK antigen to stratify ALK-positive ALCL patients with different prognosis. Leukemia. 2013;27(2):416–22.
53. Sibon D, Fournier M, Briere J, Lamant L, Haioun C, Coiffier B, et al. Long-term outcome of adults with systemic anaplastic large-cell lymphoma treated within the Groupe d'Etude des Lymphomes de l'Adulte trials. J Clin Oncol. 2012;30(32):3939–46.
54. Guru Murthy GS, Hamadani M, Bhatt VR, Dhakal I, Mehta P. Systemic anaplastic lymphoma kinase-positive anaplastic large cell lymphoma: a population-based analysis of incidence and survival. Clin Lymphoma Myeloma Leuk. 2017;17(4):201–6.
55. Pulford K, Falini B, Banham AH, Codrington D, Roberton H, Hatton C, et al. Immune response to the ALK oncogenic tyrosine kinase in patients with anaplastic large-cell lymphoma. Blood. 2000;96(4):1605–7.
56. Passoni L, Scardino A, Bertazzoli C, Gallo B, Coluccia AM, Lemonnier FA, et al. ALK as a novel lymphoma-associated tumor

antigen: identification of 2 HLA-A2.1-restricted CD8+ T-cell epitopes. Blood. 2002;99(6):2100–6.
57. Singh VK, Werner S, Hackstein H, Lennerz V, Reiter A, Wolfel T, et al. Analysis of nucleophosmin-anaplastic lymphoma kinase (NPM-ALK)-reactive CD8+ T cell responses in children wiht NPM-ALK+ anaplastic large cell lymphoma. Clin Exp Immunol. 2016;186(1):96–105.
58. Knorr F, Damm-Welk C, Ruf S, Singh VK, Zimmermann M, Reiter A, et al. Blood cytokine concentrations of pediatric anaplastic lymphoma kinase-positive anaplastic large cell lymphoma patients. Haematologica. 2018;103(3):477–85.
59. Ait-Tahar K, Damm-Welk C, Burkhardt B, Zimmermann M, Klapper W, Reiter A, et al. Correlation of the autoantibody response to the ALK oncoantigen in pediatric anaplastic lymphoma kinase-positive anaplastic large cell lymphoma with tumor dissemination and relapse risk. Blood. 2010;115(16):3314–9.
60. Mussolin L, Pillon M, Zimmermann M, Carraro E, Basso G, Knoerr F, et al. Course of anti-ALK antibody titres during chemotherapy in children with anaplastic large cell lymphoma. Br J Haematol. 2018;182(5):733–5.
61. Josimovic-Alasevic O, Durkop H, Schwarting R, Backe E, Stein H, Diamantstein T. Ki-1 (CD30) antigen is released by Ki-1-positive tumor cells in vitro and in vivo. I. Partial characterization of soluble Ki-1 antigen and detection of the antigen in cell culture supernatants and in serum by an enzyme-linked immunosorbent assay. Eur J Immunol. 1989;19(1):157–62.
62. Pizzolo G, Vinante F, Chilosi M, Dallenbach F, Josimovic-Alasevic O, Diamantstein T, et al. Serum levels of soluble CD30 molecule (Ki-1 antigen) in Hodgkin's disease: relationship with disease activity and clinical stage. Br J Haematol. 1990;75(2):282–4.
63. Nadali G, Vinante F, Stein H, Todeschini G, Tecchio C, Morosato L, et al. Serum levels of the soluble form of CD30 molecule as a tumor marker in CD30+ anaplastic large-cell lymphoma. J Clin Oncol. 1995;13(6):1355–60.
64. Zinzani PL, Pileri S, Bendandi M, Buzzi M, Sabattini E, Ascani S, et al. Clinical implications of serum levels of soluble CD30 in 70 adult anaplastic large-cell lymphoma patients. J Clin Oncol. 1998;16(4):1532–7.
65. Cortelazzo S, Ponzoni M, Ferreri AJ, Hoelzer D. Lymphoblastic lymphoma. Crit Rev Oncol Hematol. 2011;79(3):330–43.
66. Sekimizu M, Sunami S, Nakazawa A, Hayashi Y, Okimoto Y, Saito AM, et al. Chromosome abnormalities in advanced stage T-cell lymphoblastic lymphoma of children and adolescents: a report from Japanese Paediatric Leukaemia/Lymphoma Study Group (JPLSG) and review of the literature. Br J Haematol. 2011;154(5):612–7.
67. van Dongen JJ, Wolvers-Tettero IL. Analysis of immunoglobulin and T cell receptor genes. Part II: possibilities and limitations in the diagnosis and management of lymphoproliferative diseases and related disorders. Clin Chim Acta. 1991;198(1–2):93–174.
68. Raetz EA, Perkins SL, Bhojwani D, Smock K, Philip M, Carroll WL, et al. Gene expression profiling reveals intrinsic differences between T-cell acute lymphoblastic leukemia and T-cell lymphoblastic lymphoma. Pediatr Blood Cancer. 2006;47(2):130–40.
69. Uyttebroeck A, Vanhentenrijk V, Hagemeijer A, Boeckx N, Renard M, Wlodarska I, et al. Is there a difference in childhood T-cell acute lymphoblastic leukaemia and T-cell lymphoblastic lymphoma? Leuk Lymphoma. 2007;48(9):1745–54.
70. Basso K, Mussolin L, Lettieri A, Brahmachary M, Lim WK, Califano A, et al. T-cell lymphoblastic lymphoma shows differences and similarities with T-cell acute lymphoblastic leukemia by genomic and gene expression analyses. Genes Chromosomes Cancer. 2011;50(12):1063–75.
71. Bonn BR, Huge A, Rohde M, Oschlies I, Klapper W, Voss R, et al. Whole exome sequencing hints at a unique mutational profile of paediatric T-cell lymphoblastic lymphoma. Br J Haematol. 2015;168(2):308–13.
72. Bonn BR, Rohde M, Zimmermann M, Krieger D, Oschlies I, Niggli F, et al. Incidence and prognostic relevance of genetic variations in T-cell lymphoblastic lymphoma in childhood and adolescence. Blood. 2013;121(16):3153–60.
73. Weng AP, Ferrando AA, Lee W, Morris JP, Silverman LB, Sanchez-Irizarry C, et al. Activating mutations of NOTCH1 in human T cell acute lymphoblastic leukemia. Science. 2004;306(5694):269–71.
74. Matsuoka S, Oike Y, Onoyama I, Iwama A, Arai F, Takubo K, et al. Fbxw7 acts as a critical fail-safe against premature loss of hematopoietic stem cells and development of T-ALL. Genes Dev. 2008;22(8):986–91.
75. Callens C, Baleydier F, Lengline E, Ben Abdelali R, Petit A, Villarese P, et al. Clinical impact of NOTCH1 and/or FBXW7 mutations, FLASH deletion, and TCR status in pediatric T-cell lymphoblastic lymphoma. J Clin Oncol. 2012;30(16):1966–73.
76. Burkhardt B, Bruch J, Zimmermann M, Strauch K, Parwaresch R, Ludwig WD, et al. Loss of heterozygosity on chromosome 6q14-q24 is associated with poor outcome in children and adolescents with T-cell lymphoblastic lymphoma. Leukemia. 2006;20(8):1422–9.
77. Lepretre S, Touzart A, Vermeulin T, Picquenot JM, Tanguy-Schmidt A, Salles G, et al. Pediatric-like acute lymphoblastic leukemia therapy in adults with lymphoblastic lymphoma: the GRAALL-LYSA LL03 study. J Clin Oncol. 2016;34(6):572–80.
78. Balbach ST, Makarova O, Bonn BR, Zimmermann M, Rohde M, Oschlies I, et al. Proposal of a genetic classifier for risk group stratification in pediatric T-cell lymphoblastic lymphoma reveals differences from adult T-cell lymphoblastic leukemia. Leukemia. 2016;30(4):970–3.
79. Baleydier F, Decouvelaere AV, Bergeron J, Gaulard P, Canioni D, Bertrand Y, et al. T cell receptor genotyping and HOXA/TLX1 expression define three T lymphoblastic lymphoma subsets which might affect clinical outcome. Clin Cancer Res. 2008;14(3):692–700.
80. Reiter A, Schrappe M, Ludwig WD, Tiemann M, Parwaresch R, Zimmermann M, et al. Intensive ALL-type therapy without local radiotherapy provides a 90% event-free survival for children with T-cell lymphoblastic lymphoma: a BFM group report. Blood. 2000;95(2):416–21.
81. Mora J, Filippa DA, Qin J, Wollner N. Lymphoblastic lymphoma of childhood and the LSA2-L2 protocol: the 30-year experience at Memorial-Sloan-Kettering Cancer Center. Cancer. 2003;98(6):1283–91.
82. Jabbour E, Koscielny S, Sebban C, Peslin N, Patte C, Gargi T, et al. High survival rate with the LMT-89 regimen in lymphoblastic lymphoma (LL), but not in T-cell acute lymphoblastic leukemia (T-ALL). Leukemia. 2006;20(5):814–9.
83. Shepherd SF, A'Hern RP, Pinkerton CR. Childhood T-cell lymphoblastic lymphoma—does early resolution of mediastinal mass predict for final outcome? The United Kingdom Children's Cancer Study Group (UKCCSG). Br J Cancer. 1995;72(3):752–6.
84. Attarbaschi A, Mann G, Dworzak M, Wiesbauer P, Schrappe M, Gadner H. Mediastinal mass in childhood T-cell acute lymphoblastic leukemia: significance and therapy response. Med Pediatr Oncol. 2002;39(6):558–65.
85. Stark B, Avigad S, Luria D, Manor S, Reshef-Ronen T, Avrahami G, et al. Bone marrow minimal disseminated disease (MDD) and minimal residual disease (MRD) in childhood T-cell lymphoblastic lymphoma stage III, detected by flow cytometry (FC) and real-time quantitative polymerase chain reaction (RQ-PCR). Pediatr Blood Cancer. 2009;52(1):20–5.
86. Coustan-Smith E, Sandlund JT, Perkins SL, Chen H, Chang M, Abromowitch M, et al. Minimal disseminated disease in childhood T-cell lymphoblastic lymphoma: a report from the children's oncology group. J Clin Oncol. 2009;27(21):3533–9.

87. Mussolin L, Buldini B, Lovisa F, Carraro E, Disaro S, Lo Nigro L, et al. Detection and role of minimal disseminated disease in children with lymphoblastic lymphoma: the AIEOP experience. Pediatr Blood Cancer. 2015;62(11):1906–13.
88. Seidemann K, Tiemann M, Lauterbach I, Mann G, Simonitsch I, Stankewitz K, et al. Primary mediastinal large B-cell lymphoma with sclerosis in pediatric and adolescent patients: treatment and results from three therapeutic studies of the Berlin-Frankfurt-Munster Group. J Clin Oncol. 2003;21(9):1782–9.
89. Burkhardt B, Zimmermann M, Oschlies I, Niggli F, Mann G, Parwaresch R, et al. The impact of age and gender on biology, clinical features and treatment outcome of non-Hodgkin lymphoma in childhood and adolescence. Br J Haematol. 2005;131(1):39–49.
90. Reiter A, Klapper W. Recent advances in the understanding and management of diffuse large B-cell lymphoma in children. Br J Haematol. 2008;142(3):329–47.
91. Jemal A, Clegg LX, Ward E, Ries LA, Wu X, Jamison PM, et al. Annual report to the nation on the status of cancer, 1975-2001, with a special feature regarding survival. Cancer. 2004;101(1):3–27.
92. Raetz E, Perkins S, Davenport V, Cairo MS. B large-cell lymphoma in children and adolescents. Cancer Treat Rev. 2003;29(2):91–8.
93. Minard-Colin V, Brugieres L, Reiter A, Cairo MS, Gross TG, Woessmann W, et al. Non-Hodgkin lymphoma in children and adolescents: progress through effective collaboration, current knowledge, and challenges ahead. J Clin Oncol. 2015;33(27):2963–74.
94. Pillon M, Mussolin L, Carraro E, Conter V, Arico M, Vinti L, et al. Detection of prognostic factors in children and adolescents with Burkitt and Diffuse Large B-Cell Lymphoma treated with the AIEOP LNH-97 protocol. Br J Haematol. 2016;175(3):467–75.
95. Sandlund JT, Martin MG. Non-Hodgkin lymphoma across the pediatric and adolescent and young adult age spectrum. Hematology Am Soc Hematol Educ Program. 2016;2016(1):589–97.
96. Cairo MS, Gerrard M, Sposto R, Auperin A, Pinkerton CR, Michon J, et al. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. Blood. 2007;109(7):2736–43.
97. Patte C, Auperin A, Gerrard M, Michon J, Pinkerton R, Sposto R, et al. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. Blood. 2007;109(7):2773–80.
98. Patte C, Auperin A, Michon J, Behrendt H, Leverger G, Frappaz D, et al. The Societe Francaise d'Oncologie Pediatrique LMB89 protocol: highly effective multiagent chemotherapy tailored to the tumor burden and initial response in 561 unselected children with B-cell lymphomas and L3 leukemia. Blood. 2001;97(11):3370–9.
99. Reiter A, Schrappe M, Tiemann M, Ludwig WD, Yakisan E, Zimmermann M, et al. Improved treatment results in childhood B-cell neoplasms with tailored intensification of therapy: a report of the Berlin-Frankfurt-Munster Group Trial NHL-BFM 90. Blood. 1999;94(10):3294–306.
100. Woessmann W, Seidemann K, Mann G, Zimmermann M, Burkhardt B, Oschlies I, et al. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. Blood. 2005;105(3):948–58.
101. Miles RR, Cairo MS, Satwani P, Zwick DL, Lones MA, Sposto R, et al. Immunophenotypic identification of possible therapeutic targets in paediatric non-Hodgkin lymphomas: a children's oncology group report. Br J Haematol. 2007;138(4):506–12.
102. Mussolin L, Basso K, Pillon M, D'Amore ES, Lombardi A, Luzzatto L, et al. Prospective analysis of minimal bone marrow infiltration in pediatric Burkitt's lymphomas by long-distance polymerase chain reaction for t(8;14)(q24;q32). Leukemia. 2003;17(3):585–9.
103. Lovisa F, Mussolin L, Corral L, Pillon M, Cazzaniga G, Biondi A, et al. IGH and IGK gene rearrangements as PCR targets for pediatric Burkitt's lymphoma and mature B-ALL MRD analysis. Lab Investig. 2009;89(10):1182–6.
104. Mussolin L, Pillon M, Conter V, Piglione M, Lo Nigro L, Pierani P, et al. Prognostic role of minimal residual disease in mature B-cell acute lymphoblastic leukemia of childhood. J Clin Oncol. 2007;25(33):5254–61.
105. Mussolin L, Pillon M, d'Amore ES, Conter V, Piglione M, Lo Nigro L, et al. Minimal disseminated disease in high-risk Burkitt's lymphoma identifies patients with different prognosis. J Clin Oncol. 2011;29(13):1779–84.
106. Rosolen A, Perkins SL, Pinkerton CR, Guillerman RP, Sandlund JT, Patte C, et al. Revised international pediatric non-Hodgkin lymphoma staging system. J Clin Oncol. 2015;33(18):2112–8.
107. Shiramizu B, Goldman S, Smith L, Agsalda-Garcia M, Galardy P, Perkins SL, et al. Impact of persistent minimal residual disease post-consolidation therapy in children and adolescents with advanced Burkitt leukaemia: a Children's Oncology Group Pilot Study Report. Br J Haematol. 2015;170(3):367–71.
108. Attarbaschi A, Carraro E, Abla O, Barzilai-Birenboim S, Bomken S, Brugieres L, et al. Non-Hodgkin lymphoma and pre-existing conditions: spectrum, clinical characteristics and outcome in 213 children and adolescents. Haematologica. 2016;101(12):1581–91.
109. Miles RR, Raphael M, McCarthy K, Wotherspoon A, Lones MA, Terrier-Lacombe MJ, et al. Pediatric diffuse large B-cell lymphoma demonstrates a high proliferation index, frequent c-Myc protein expression, and a high incidence of germinal center subtype: report of the French-American-British (FAB) international study group. Pediatr Blood Cancer. 2008;51(3):369–74.
110. Oschlies I, Klapper W, Zimmermann M, Krams M, Wacker HH, Burkhardt B, et al. Diffuse large B-cell lymphoma in pediatric patients belongs predominantly to the germinal-center type B-cell lymphomas: a clinicopathologic analysis of cases included in the German BFM (Berlin-Frankfurt-Munster) Multicenter Trial. Blood. 2006;107(10):4047–52.
111. Szczepanowski M, Lange J, Kohler CW, Masque-Soler N, Zimmermann M, Aukema SM, et al. Cell-of-origin classification by gene expression and MYC-rearrangements in diffuse large B-cell lymphoma of children and adolescents. Br J Haematol. 2017;179(1):116–9.
112. Poirel HA, Cairo MS, Heerema NA, Swansbury J, Auperin A, Launay E, et al. Specific cytogenetic abnormalities are associated with a significantly inferior outcome in children and adolescents with mature B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study. Leukemia. 2009;23(2):323–31.
113. Seidemann K, Zimmermann M, Book M, Meyer U, Burkhardt B, Welte K, et al. Tumor necrosis factor and lymphotoxin alfa genetic polymorphisms and outcome in pediatric patients with non-Hodgkin's lymphoma: results from Berlin-Frankfurt-Munster Trial NHL-BFM 95. J Clin Oncol. 2005;23(33):8414–21.
114. Cairo MS, Sposto R, Gerrard M, Auperin A, Goldman SC, Harrison L, et al. Advanced stage, increased lactate dehydrogenase, and primary site, but not adolescent age (>/= 15 years), are associated with an increased risk of treatment failure in children and adolescents with mature B-cell non-Hodgkin's lymphoma: results of the FAB LMB 96 study. J Clin Oncol. 2012;30(4):387–93.
115. Salzburg J, Burkhardt B, Zimmermann M, Wachowski O, Woessmann W, Oschlies I, et al. Prevalence, clinical pattern, and outcome of CNS involvement in childhood and adolescent non-Hodgkin's lymphoma differ by non-Hodgkin's lymphoma subtype: a Berlin-Frankfurt-Munster Group Report. J Clin Oncol. 2007;25(25):3915–22.
116. Pillon M, Carraro E, Mussolin L, Conter V, Tondo A, Arico M, et al. Primary mediastinal large B-cell lymphoma: outcome of a series of pediatric patients treated with high-dose methotrexate and cytarabine plus anti-CD20. Pediatr Blood Cancer. 2018;65(2)
117. Pillon M, Arico M, Mussolin L, Mainardi C, Giraldi E, Garaventa A, et al. Mediastinal Burkitt lymphoma in childhood. Pediatr Blood Cancer. 2014;61(11):2127–8.

Part IV

Lymphoblastic Lymphoma

Lymphoblastic Lymphoma

12

Birgit Burkhardt and Birte Wistinghausen

Introduction

Great strides have been made in the treatment of lymphoblastic lymphoma (LBL) with event-free and overall survivals exceeding 80% [2, 3]. The vast majority is of T-lymphoblastic origin (70–80%) with only 20–25% arising from B-lymphoblasts and mixed myeloid/lymphoblastic phenotypes being very rare [4, 5]. Current research efforts aim to increase cure rates, to identify high-risk patients in need of more intensive or novel therapies, and to avoid unnecessary exposure to excessive toxicity, especially of low-risk patients. Essential steps in achieving these aims are a better understanding of the molecular drivers of the disease, the development of robust risk stratifications and the identification of novel therapies that retrieve relapsed patients, can be brought to front line therapy quickly and reduce toxicity.

Epidemiology and Clinical Presentation

The median age of diagnosis is around 9 years of age and not significantly different between B-LBL (8 years) and T-LBL (8.8 years) [4]. T-LBL affects males 2.5 times more often than girls, but there are no differences in gender distribution for B-LBL [4].

Adolescent and young adult (AYA) T-LBL patients most commonly present with an anterior mediastinal mass arising from the thymus that can cause airway compression or superior vena cava (SVC) syndrome and is frequently accompanied with pleural or pericardial effusions. T-LBL can arise in any lymph node of the body, but the majority of patients have involvement of mediastinum (52%) and cervical nodes (31%) [6]. Symptoms of airway include shortness of breath, cough, stridor, dyspnea, and acute respiratory distress. Edema of the neck and face and jugular venous distension should raise the suspicion of SVC syndrome. Most patients with T-LBL present with disseminated disease (Murphy stage III). About 15–20% of patients exhibit bone marrow infiltration. Less than 5% show central nervous system (CNS) involvement.

B-LBL patients are more likely to present with limited stage (Murphy stage I and II) compared to T-LBL patients; however, they have a higher incidence of bone marrow involvement of about 30–40% [4, 7]. The most frequent sites of involvement in B-LBL are in the head and neck area and include the bone, skin, lymph nodes, and soft tissue [7, 8]. CNS involvement was detected in about 5%. Depending on the site of manifestation, clinical presentations vary. Bulky lymphadenopathy and respiratory and systemic symptoms are uncommon in B-LBL.

Pathology

Histologically, LBL shows an infiltrate of small, round blue cells. Further evaluation either by flow cytometry of malignant effusions and fresh tissue or immunohistochemical analysis of paraffin embedded biopsies is needed to confirm the diagnosis. The European Group established guidelines for Immunophenotyping of Leukemias (EGIL) and the World Health Organization (WHO). EGIL published criteria for flow cytometric diagnosis of acute lymphoblastic leukemia (ALL) in 1995 which were updated by the European LeukemiaNet in 2011 [9, 10]. The WHO revised the fourth edition of the classification of tumors of the hematopoietic and lymphoid lineage in 2016 which incorporates new

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information, especially genetic and molecular findings [11]. The International BFM-FLOW network published AIEOP-BFM consensus guidelines for immunophenotyping by flow cytometry in 2016 designed to fulfill EGIL and WHO 2008/2016 requirements for ALL subtyping including early-T-cell (ETP) and mixed phenotype acute leukemia (MPAL) [12]. The recurrent genetic and molecular changes seen in ALL have been very well characterized for B-ALL and are important prognostic predictors [13]. There is an ongoing debate whether ALL and LBL are distinct entities or a spectrum of the same disease, but they share similar molecular alterations [14–16].

Immunophenotypic and Immunohistochemical Analysis

The AIEOP-BFM consensus antibody panel includes mandatory markers to satisfy WHO, EGIL, and ETP classifications and recommendations for optional markers (Table 12.1) [12].

Lymphoblastic lymphomas of precursor B-cell lineage express a combination of B-cell markers including CD19, cCD79a, and cCD22 which are not specific by themselves but in combination strongly support the diagnosis (Fig. 12.1) [11]. In addition, PAX5, surface CD22, CD24, CD10, and TdT are expressed in most cases. Expression of CD20, a mature B-cell marker, and/or CD34, a stem cell marker, can be less commonly seen. Co-expression of the myeloid-associated antigens CD13 and 33 can occur but does not indicate mixed lineage. The degree of maturation can vary. In early precursor B-ALL/B-LBL, expression of CD19, cCD79A, cCD22, and nuclear TdT is seen. During the common ALL phenotypic stage, expression of CD10 is found. In more mature pre-B- or early B-cell lymphomas with L1/2 morphology, expression of cytoplasmic μ chain with occasional expression of surface heavy chain is seen but without any light chain expression.

Table 12.1 The AIEOP-BFM consensus antibody panel for pediatric ALL

Recommendation	Marker
Mandatory	Mandatory and optional markers (each combined with CD45)
Intracellular[a,b]	iCD3, iCD22, iCD79a, iIgM (μ-chain), iLysozyme, iMPO
Surface[a]	CD2[c], CD3, CD5, CD7; CD10, CD19, CD20; CD11c, CD11b, CD13, CD14, CD15, CD33, CD64, CD65[d], CD117; CD34, (CD45), CD56, HLA-DR If T-ALL: CD1a, CD4, CD8, TCR[a,b], TCR[d,e] If B-IV suspected: λ-chain, Κ-chain (surface staining after pre-washing or intracellular)
Optional/ recommended	All cases: NG2[f], CD371[c,g] If BCP-ALL: CD11ac, CD22, CD24,CD38, CD44, CD58, CD66[c], CD123[c], CRLF2[c,e] If T-ALL: CD99, iTdT If BAL according to general panel: CD24, iTdT

[a]Mandatory markers for WHO, EGIL, and ETP classifications
[b]Prefix "i" stands for intracellular staining
[c]Phycoerythrin conjugate (PE) recommended
[d]Available only labelled with fluorescein isothiocyanate (FITC)
[e]Clone 1D3
[f]Clone 7.1
[g]Clone 50C1

T-LBL express cytoplasmic or membrane bound CD3, which is lineage specific (Fig. 12.2). In addition, they are mostly positive for TdT and variably positive for CD1a, CD2, CD4, CD5, CD7, and CD8. They are further subclassified by the differentiation stage of T-lymphoblasts on their passage through the thymus [17]. In addition to TdT, the most specific markers to indicate the precursor nature of T-lymphoblasts are CD99, CD34, and CD1a. Oschlies and colleagues published an easily followed algorithm for the diagnosis of lymphoblastic lymphomas using immunohistochemical staining based on the analysis of 188 patients [17]. TdT expression has been identified as the best marker for determining the precursor cell nature of a lymphoma. In TdT-negative lymphoma with typical lymphoblastic morphology, either expression of CD1a or CD34, co-expression of CD79a and CD3, or co-expression of CD4 and CD8 can be used to determine the precursor cell nature of lymphoma.

The Children's Oncology Group (COG) reported immunophenotypic characteristics of T-lymphoblastic lymphoma in 180 children and adolescents enrolled on the COG trial A5971, which showed that the immunoprofile was similar to T-ALL but with a higher incidence of CD4/CD8 double positivity pointing to a more mature phenotype [6]. Diagnostically useful immunophenotypic features of T-LBL were identified as well as distinct immunophenotypic subgroups, but none were statistically related to event-free or overall survival. As previously published by Oschlies et al., the majority of cases demonstrated subcapsular or cortical thymocyte phenotypes [17]. Smock et al. reported that a majority of LBL samples expressed MIB1 (59%) and cMyc (77%) in greater than 50% of analyzed cells by immunohistochemistry [18]. It is unknown if the c-Myc overexpression is due to NOTCH-signaling perturbation or if other NOTCH-independent mechanisms are involved.

The St. Jude's group in collaboration with the Italian AIEOP group first recognized a T-ALL phenotype of very early differentiation that occurs in 10–15% of patients with T-ALL, termed early T-precursor acute lymphoblastic leukemia (ETP-ALL) and defined by the expression of CD7 and low level CD5 (and occasionally cytoplasmic CD3) but lacking expression of CD1a, CD4, and CD8 [19, 20]. Co-expression of at least one of the stem cell markers CD34 or CD117 or myeloid-related antigens such as CD33 or

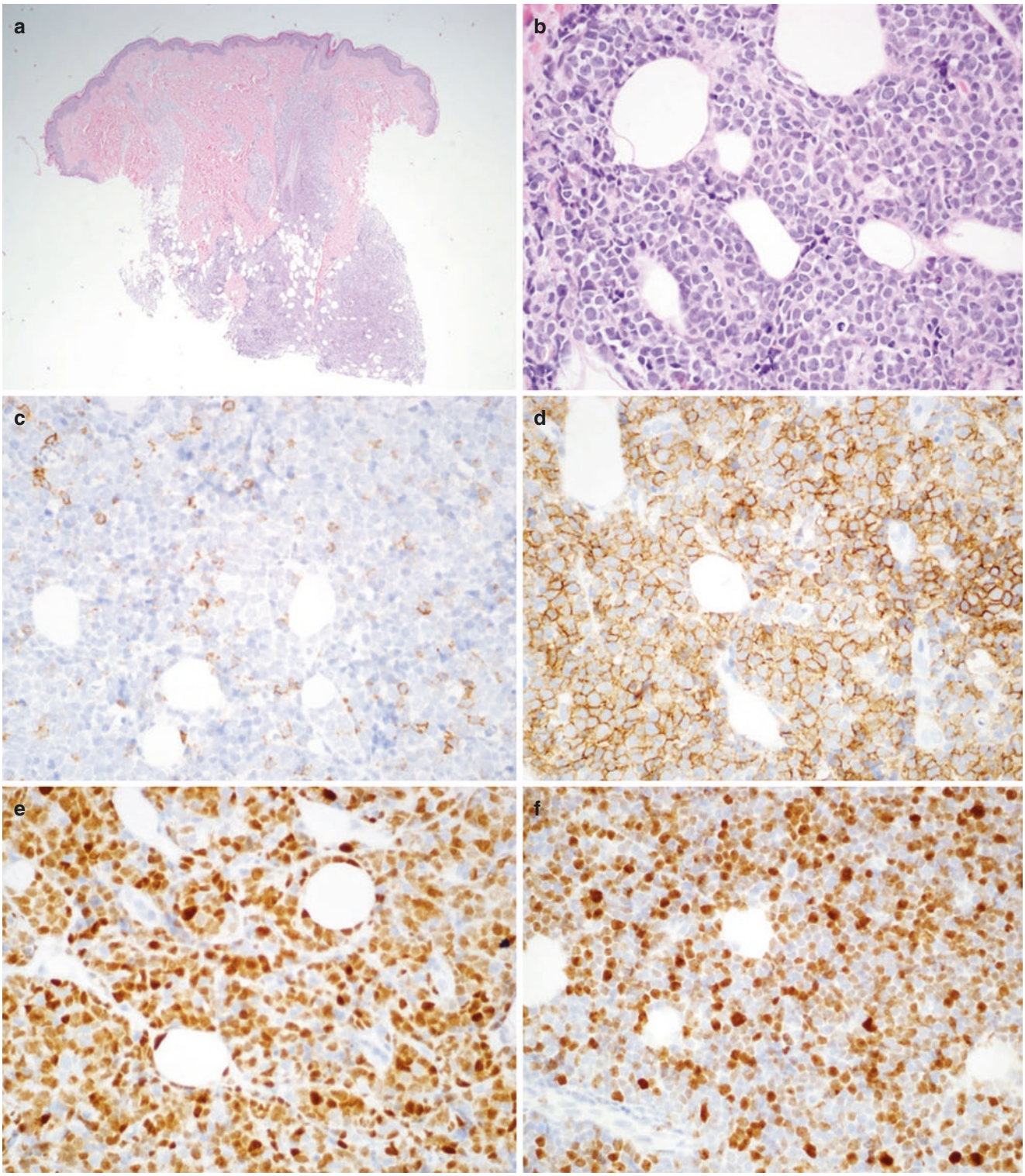

Fig. 12.1 B-lymphoblastic lymphoma of the skin. Infiltration of the dermis and subcutis by blasts (**a**, **h**, and **e**) showing a monoform population of small round blue cells (**b**, **h**, and **e**) that is negative for CD20 expression (**c**) but positive for CD19 on the surface and TdT in the nucleus (**e**) and a high proliferative index (MIB1 expression, **f**); courtesy of Ilske Oschlies and Wolfram Klapper

CD13 is present [19, 21]. In concordance with the frequently found aberrant expression of myeloid- or stem cell-related antigens such as CD13, CD33, CD34, or CD117, ETP-ALL is associated with increased incidence of AML-type mutations rather than T-ALL-/T-LBL-associated *NOTCH* mutations [22]. ETP-ALL was originally described being

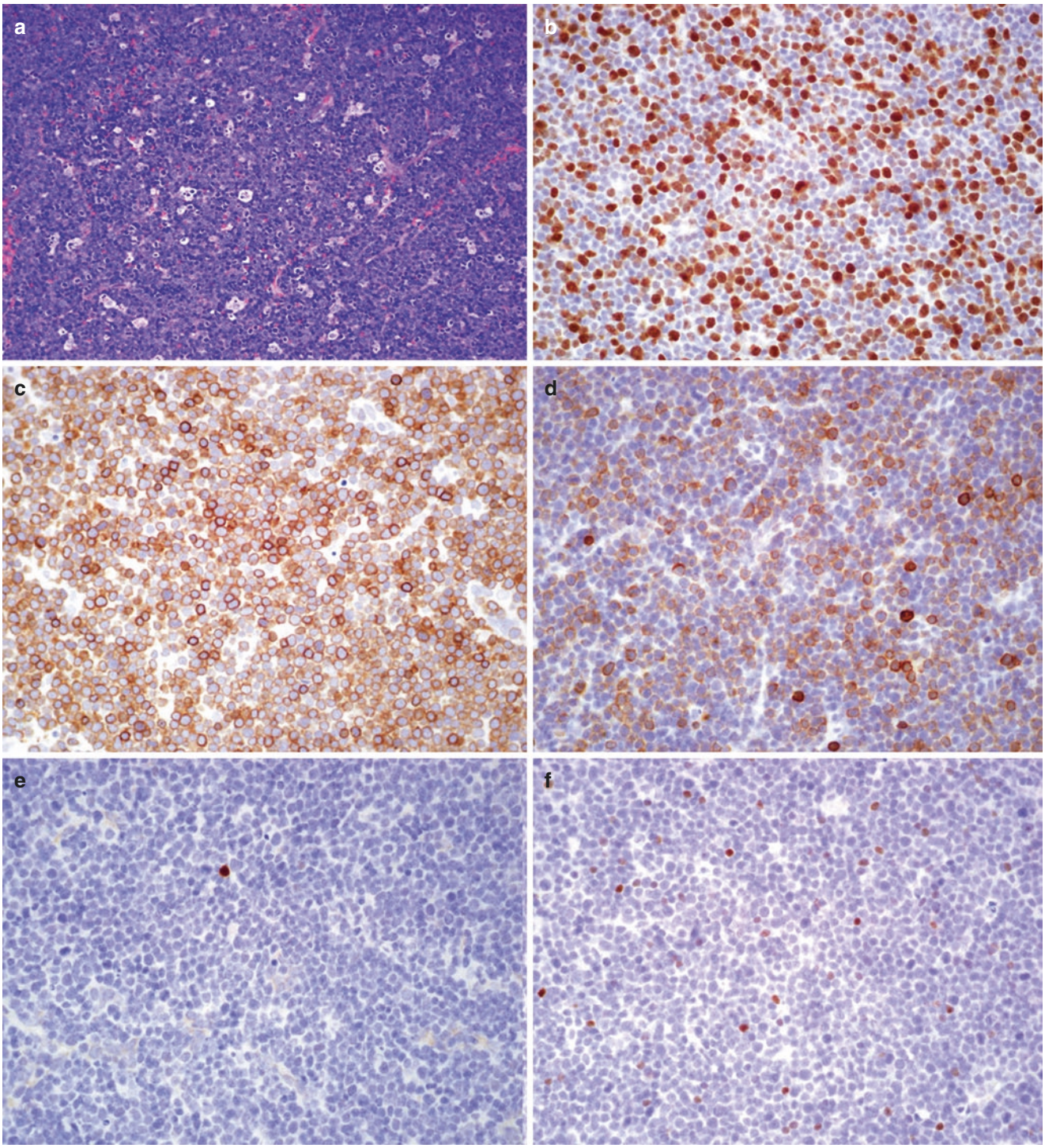

Fig. 12.2 Nodal T-lymphoblastic lymphoma. Complete infiltration and replacement of normal lymph node architecture by dense blast infiltration (**a**, Giemsa) with high proliferative index (MIB1 expression, **b**). Blasts are positive for the T-cell marker CD3 (**c**) with aberrant expression of CD79a (**d**) but negative for other B-cell markers (Pax5, **e**) and partial expression of TdT (**f**); courtesy of Ilske Oschlies and Wolfram Klapper

associated with increased rate of induction failures, but with current treatment protocols there is no significant difference in prognosis [23]. The ETP phenotype appears to occur in T-LBL with similar frequency with 14% of T-LBL patients enrolled on the COG study 5971 classified as the ETP phenotype [6].

Mixed phenotype acute leukemia (MPAL) phenotype has been reported in LBL with or without limited bone

marrow involvement [24]. Of 188 LBL patients enrolled on EURO-LB 02, 7% of the cases were classified as MPAL by central review of pathology [17]. There are no current treatment recommendations for this rare entity, but there is some evidence that outcomes are better with ALL-type therapy [25].

Recurrent Genetic and Molecular Alterations

The recurrent molecular alterations have been well characterized and associated with prognosis for B-ALL [13], but much less is known about the molecular makeup of LBL due to the lack of available fresh tissue for molecular typing.

The poor-risk cytogenetic markers BCR-ABL1 and KMT2A rearrangements have been described in B-LBL, but the incidence and prognostic significance are unknown [26, 27]. The most commonly described cytogenetic abnormalities are alterations in chromosome 21 (trisomy or duplications of 21q22) [28, 29]. More recently, Meyer et al. compared copy number alterations in cohorts of B-ALL versus B-LBL using formalin-fixed tissue and found some significant differences [30]. While the incidence of CDKN2A/B, IKZF1, and PAX5 deletions were similar, ETV6 and EBF1 deletions were less common in B-LBL. In addition, hyperdiploidy is common in both B-ALL and B-LBL, but the classic triple trisomy of chromosomes 4, 10, and 17 associated with favorable prognosis in B-ALL was not found in B-LBL. Interestingly, all cases of hyperdiploidy presented in localized stages.

Due to the recombination processes of T-cell receptor rearrangement, the T-cell receptor (TCR) genes are predisposed to recombination with oncogenes or genes involved in thymocyte development through chromosomal translocations. These recurrent translocations are found in 50% of pediatric T-ALLs. The prevalence of these translocations in pediatric T-LBL is not exactly known. The current literature shows that most cytogenetic abnormalities reported in T-LBL are also seen in T-ALL [15, 16, 31, 32].

While there are no characteristic translocations in T-ALL/T-LBL correlating with prognosis as in B-ALL, recent molecular studies have identified candidate genes of prognostic relevance for T-LBL including NOTCH1 and FBXW7. Mutations in *NOTCH1* and/or *FBXW7* at recurrent hotspots within the genes are observed in approximately 50% of pediatric T-ALL patients and have been associated with an improved treatment response or outcome [33, 34]. Concerning pediatric T-LBL patients, five studies are published reporting the results of *NOTCH1* and/or *FBXW7* mutation analyses in 116, 54, 14, 11, and 9 cases, respectively [15, 35–38]. In the larger series, *NOTCH1* and/or *FBXW7* mutations are associated with a favorable response to treatment and/or outcome.

In descriptive retrospective analyses of pediatric T-LBL patients, loss of heterozygosity at chromosomal region 6q14-24 (LOH6q) is shown to be highly significantly associated with adverse outcome and increased risk of relapse [31, 35, 39]. Within a total of 217 analyzed patients, pEFS at 5 years is 86 ± 3% for LOH6q negative patients compared to 27 ± 9% in LOH6q positive patients ($p < 0.0001$).

Mutations in the tumor suppressor gene *PTEN* have been reported in different types of solid and hematological malignancies and were associated with unfavorable outcome of patients. A report of the NHL-BFM study group identified a significant association of *PTEN* mutations with adverse outcome of analyzed T-LBL patients [40]. *PTEN* mutations were detected in 15% of 114 pediatric T-LBL patients and were associated with a poor pEFS at 5 years of 59 ± 12% compared to 82 ± 4 for *PTEN* non-mutated cases ($p = 0.014$). Although biological data suggest that any *PTEN* mutation leads to hyperactivated PI3K-AKT signaling, the prognostic impact is weaker when other PI3K-AKT pathway mutations are included in the analysis, indicating that the negative prognostic impact mostly depended on *PTEN* mutations. It is hence hypothesized that *PTEN* controls resistance to therapy by PI3K-AKT-independent signaling. Outcomes of patients with heterozygous or homozygous/biallelic *PTEN* mutations are similar, suggesting that *PTEN* acts as haploinsufficient tumor suppressor in pediatric T-LBL. It is reported that the expression of *PTEN* was transcriptionally repressed by active NOTCH1 in T-ALL cell lines, as well as normal mouse thymocytes [41, 42]. This suggests a synergistic effect of both mutations in *NOTCH1* and *PTEN*, but investigation of the prognostic impact of a combination of both genetic markers in the analyzed cohort of patients treated according to NHL-BFM regimens revealed the opposite: the unfavorable prognostic effect of *PTEN* mutations seems to be abrogated by the favorable prognostic impact of *NOTCH1* mutations, as this group of patients presented with a pEFS of 91 ± 9% [40]. Similar associations and interactions of *NOTCH1* and *PTEN* mutations with outcome have recently been described in pediatric T-ALL treated with BFM-type regimens [43]. An analysis of 271 pediatric patients treated on AIEOP-BFM protocols confirmed the negative prognostic significance of PTEN showing the worst prognosis in patients with PTEN mutations combined with unmutated NOTCH1 [44]. In contrast to these findings, pediatric T-ALL patients treated according to DCOG and COALL regimens without *NOTCH1* and *PTEN* mutations showed a significantly lower cumulative incidence of relapse at 5 years compared with the rest of the cohort [45].

Absence of biallelic T-cell receptor gene gamma (TRG) locus deletion (ABD), which is characteristic for early thymocyte precursors before V(D)J recombination, correlates statistically significantly with the failure to induction chemotherapy of pediatric T-ALL patients but also poorer outcomes

[46, 47]. For pediatric T-LBL, ABD is observed in a small subgroup of 4 out of 54 patients (7%) treated according to NHL-BFM 95 or EURO-LB 02 regimen [36]. All four patients had mutations in *NOTCH1* and/or *FBXW7*. ABD was in this cohort associated with a poor pEFS of 0% compared to 80 ± 6% for non-ABD patients ($p = 0.01$).

SIK1, an anti-metastatic protein, is a direct target of *miR-223* and consequently is significantly reduced in *miR-223*-overexpressing tumor cells. Overexpression of *miR-223* promotes tumor T-LBL cell growth, migration, and invasion in vitro. To evaluate the clinical relevance in T-LBL, the expression levels of *miR-223* was measured in tumor biopsies from 67 T-LBL pediatric patients and correlated with clinical data. Multivariate analysis confirmed that only a high level of *miR-223* was an independent factor for worse prognosis [48].

Risk Classification and Prognostic Factors

Prognostic predictors of therapy are needed for pediatric LBL patients to prevent overtreatment and subsequent acute and long-term toxicities such as osteonecrosis in low-risk patients but also to identify patients at highest risk of often fatal relapse and thus in need of novel therapies [49]. The prognostic relevance of clinical characteristics, such as age, sex, stage, presence of mediastinal mass, and level of serum lactate dehydrogenase, has been described [49]. Tubergen et al. reported an unfavorable prognosis for children older than 14 years based on the results of the CCG502 trial [50]. Analysis of the outcome of T-LBL patients treated within different NHL-BFM trials revealed a lower pEFS in adolescent females compared to males with comparable clinical characteristics [51]. Analysis of the EORTC 58881 trial led to the identification of response to therapy as a prognostic factor [52]. Non-response after 7 days of prephase with prednisolone and one intrathecal injection with MTX was associated with very poor outcome. But these data have not been confirmed uniformly across cooperative groups.

Traditionally, patients were stratified according to stage. In the NHL-BFM 90, NHL-BFM 95, and EURO-LB 02 trials, patients with limited stage I/II disease did not receive re-induction treatment protocol II. Outcome analysis of the trial EURO-LB 02 supported stage of disease as stratification criterion for pB-LBL resulting in favorable pEFS for pB-LBL with limited disease, representing almost half of pB-LBL patients [3]. However, for pB-LBL patients with advanced stage III/IV disease, pEFS and cumulative incidence of relapse were poor even with intensified treatment including protocol II [3]. In the COG protocol A5971, patients with localized stages (Murphy stage I and II) received similar therapy to patients with disseminated disease but were not included in the randomization to intensify therapy. Patients with localized disease had a 5-year EFS of 90% compared to patients with disseminated stage who achieved a 5-year EFS of 82% [2, 53]. However, since 74% of patients with localized stage had a B-LBL phenotype and 86% of patients with disseminated disease had T-LBL, no analysis by phenotype was possible. In T-LBL, it is well known that the number of patients with limited stage I/II disease is very low. Therefore, stage of disease is an insufficient parameter to identify low-risk T-LBL patients potentially available for treatment de-escalation. In the EURO-LB 02 trial, only 8 out of 233 T-LBL patients (3%) were diagnosed with stage I/II disease [3]. Importantly, there was no relevant difference in pEFS for T-LBL stage III compared to stage IV disease. On A5971, T-LBL patients with CNS involvement had a significantly poorer outcome with 5-year EFS of 62% compared to 82% for patients with disseminated T-LBL but no CNS disease [2].

Minimal Disseminated (MDD) and Minimal Residual Disease (MRD)

Risk stratification in ALL is primarily based on the detection of MRD in the peripheral blood and bone marrow using either quantitative polymerase chain reaction (PCR)-based patient-specific TCR gene rearrangements or flow cytometric analysis. Both methods have also been used for detection of MDD in the peripheral blood and bone marrow at diagnosis [54, 55]. Coustan-Smith et al. showed that flow cytometric analysis of peripheral blood samples can be used to detect evidence of disseminated T-LBL, rendering it a valuable method to monitor blast clearance during therapy [55]. Out of 99 pediatric T-LBL patients, 72% had detectable levels (>0.01%) of T-lymphoblasts in their bone marrow. The level of detectable disease correlated with outcome with a 2-year EFS of 68% for patients with ≥1% lymphoblasts in the bone marrow versus 91% for patients with <1% involvement ($p = 0.031$) [55]. In a more recent study conducted by the Italian AIEOP study group, the prognostic value of MDD analyzed by multiparametric flow cytometry (FCM) in bone marrow and peripheral blood samples was evaluated in a cohort of 65 children with T- and B-lineage lymphoblastic lymphoma. MDD was detected in 49% (32/65) of BM samples, whereas only 21% (14/65) were positive at standard morphological evaluation. Using an MDD cutoff level of 3% by FCM, the 5-year EFS is 60% for patients with MDD >3% LBL cells versus 83% for the remaining patients ($p = 0.04$) [56]. Additional prospective trials are needed to determine the level of significant MDD and the role of bone marrow MRD in pediatric T-LBL as prognostic markers.

Staging

The work-up of newly diagnosed patients consists of a complete medical history, physical examination including testicular examination in boys, and basic laboratory tests including a complete blood count with differential, electrolytes, lactate dehydrogenase (LDH), uric acid, and assessment of kidney (creatinine and blood urea nitrogen (BUN)) and liver function. Staging and extent of disease are evaluated by imaging of the neck, chest, abdomen and pelvis, lumbar puncture with cerebrospinal fluid (CSF) cytology, bone marrow aspirate, and possible bone marrow biopsy. Imaging modalities vary between centers and countries but at a minimum need to include ultrasound of the neck, abdomen, pelvis, and lymph nodes, but more commonly patients are staged with computerized tomography (CT), magnetic resonance imaging (MRI) and fluoro-deoxy-glucose (FDG) positron emission tomography (PET) CT/MRI scanning, although published data for the use of PET scans in pediatric patients is limited [57–61]. The role of PET imaging to document response as a prognostic marker is also still under investigation. Magnetic resonance imaging (MRI) can be used for imaging of the neck, head, abdomen, and pelvis but is not as good for the chest because of motion artefacts due to breathing. The modified Murphy or St. Jude staging classification is used in lymphoblastic lymphomas [62]. Recently, a revised classification system, the International Paediatric Non-Hodgkin Lymphoma Staging System (IPNHLSS), that allows more precise documentation of extranodal dissemination and advanced diagnostic and imaging methods has been introduced [63].

Treatment

Frontline Therapy

Using ALL-type treatment regimens, event-free survival rates of 75–90% have been achieved as summarized in Table 12.2. Current protocols are mostly derived from either the LSA2L2 regimen that was established in the USA (Memorial Sloan-Kettering Cancer Center) or the NHL-BFM protocol based on the ALL-BFM strategy that achieved the first durable cure rates in pediatric patients with LBL [64–67]. Almost all subsequently developed treatment regimens in Western Europe and the USA use the backbone of one of these pioneer protocols. There is limited published data about Asian studies on pediatric LBL [68–70]. Results of a small retrospective Chinese cohort study on pediatric T-LBL patients treated with one of three treatment protocols revealed outcomes in the range of 64% for all patients [71]. Recently, data on 136 analyzed Japanese pediatric LBL patients with advanced disease were published [72] confirming by univariate analysis results of a previous Japanese report that showed an inferior outcome of T-LBL patients presenting with stage III compared to stage IV [69].

In contrast to ALL treatment regimens, stage at diagnosis is the only parameter for risk group stratification of patients with LBL classifying patients into limited stage (stage I and II) versus advanced stage (stage III and IV). Published EFS for patients with limited stage disease ranges from 73 ± 8% (LMT81) to 100% (LNH92). These results may be achievable with relevant dose reductions in anthracyclines and alkylators, as indicated by the NHL-BFM 90 trial, which administered no re-induction for patients with limited disease [67]. Treatment durations for patients with limited disease range from 12 to 24 months.

To improve CNS directed treatment, several protocol modifications of methotrexate (MTX) administration are evaluated. The French LMT81 trial modified the LSA2L2 protocol by the addition of ten courses of high-dose MTX (HD-MTX) with a resultant EFS of 75% [73]. The US trial POG 9404 analyzed the effectiveness of a Dana-Farber backbone therapy with or without addition of HD-MTX in T-ALL and T-LBL patients. In T-LBL patients, in contrast to T-ALL patients, there were no significant differences in EFS in the two arms [74]. The subsequent COG trial A5971 tested a COG BFM-type regimen with different schedules of CNS directed treatment where HD-MTX without additional intrathecal MTX in interim maintenance was randomized against an intensified intrathecal MTX (IT-MTX) treatment arm without HD-MTX for CNS prophylaxis. Each treatment arm was randomized with or without early intensification. There were no significant differences in EFS, and the authors concluded that either IT-MTX or HD-MTX effectively prevented CNS relapse [2]. A recent report from the I-BFM group showed that escalating methotrexate (Capizzi MTX) in combination with the BFM backbone had an EFS of 90.8% in 58 LBL patients. CNS-directed treatment was mainly based on frequent intrathecal injections without routine cranial radiation [75]. Results from the COG protocol AALL0434 were recently released [76, 77]. The protocol enrolled pediatric and young adults patients with T-ALL and T-LBL and had a double randomization between Capizzi and HD MTX and the addition of nelarabine, a nucleoside analogue, respectively. T-LBL patients did not participate in the methotrexate randomization, and only high-risk (HR) T-LBL patients, defined as having more than 1% T-lymphoblasts in the bone marrow detected by MMD flow cytometry, were eligible for the nelarabine randomization. Most patients received prophylactic cranial radiation. The Capizzi methotrexate arm showed significantly improved outcome in T-ALL patients with an estimated 4-year disease-free survival (DFS) of 92% versus 85% in the HD MTX arm (p = 0.005). More importantly, the addition of nelarabine improved outcome in T-ALL patients with a 4-year DFS of

Table 12.2 Summary of treatment results of recent clinical trials for children and adolescents with lymphobastic lymphoma

Trial	Age	Stage	Treatment	No. of pts	pEFS	Reference
LMT81	9 years (0.9–16)	I–IV	mod. LSA2-L2	84	75 ± 3%	Patte et al. [73]
CCG502	9 years (0.5–19)	I–IV	mod. LSA2-L2 vs ADCOMP	143138	74% 64%	Tubergen et al. [50]
	10 years [5–15]	III/IV	L-Asp − vs L-Asp +	83 84	64 ± 6% 78 ± 5%	Amylon et al. [86]
NHL-BFM90	9 years [1–16]	I–IV	ALL-BFM	105	90%	Reiter et al. [67]
NHL-BFM95	8 years (0.2–19)	III/IV	ALL-BFM	169	78 ± 3%	Burkhardt et al. [87]
EORTC58881	8 years (0–16)	I–IV	ALL-BFM	119	78 ± 3%	Uyttebroeck et al. [52]
COG Pilot	n.d.	III/IV	mod. LSA2-L2	85	78 ± 5%	Abromowitch et al. [88]
COG A5971	10 years	III/IV	NHL-BFM95 MTXw/o HD-MTXintensification w/o intensification	257 (all)	85 ± 4%83 ± 4%83 ± 4%83 ± 4%	Abromowitch et al. (Abstract) [89]
LNH92	8 years (0–<16)	I–IV	mod. LSA2-L2	55	69 ± 6%	Pillon et al. [90]
St. Jude 13	n.d.	III/IV	T-ALL	41	83%	Sandlund et al. [91]
POG 9404	50% < 10 years	III/IV	mod. DFCI ALL with HDMTX w/o HDMTX	1376671	82 ± 5% 88 ± 4%	Asselin et al. [74]
A 5971	>12 months	I–II	CCG-BFM	56	90%	Termuhlen et al. [53]
EURO-LB 02	0–<21 years	I–IV	NHL/ALL-BFM 90dexa (10 mg/m^2) vs pred (60 mg/m^2)	319 (all)98 88	81 ± 2%84 ± 4 (dexa)84 ± 4% (pred)	Landmann et al. [3]
EORTC58951	n.d.		mod. BFM 90dexa (6 mg/m^2) vs. pred (60 mg/m^2)	3737	85%89 ± 5% (pred)81 ± 6% (dexa)	Uyttebroeck et al. (Abstract) [92]
SFOP LMT96	10.5 years		mod. BFM	79	85%	Bergeron et al. [93]
AALL0434		II–IV, *>1% MMD in BM*	COG-BFM ± Nelarabine	118	87% 85.0 ± 5.6% (standard arm) 89.0 ± 4.7% (nelarabine)	Dunsmore et al. [77]

90% versus 83% (p = 0.0332). One hundred eighteen HR T-LBL were enrolled, and nelarabine did not improve their outcome with a 4-year DFS 85.0 ± 5.6% versus 89.0 ± 4.7% for nelarabine (N = 60) versus no nelarabine (N = 58), p = 0.2788. The ongoing COG protocol AALL1231 is randomizing patients to the addition of bortezomib, a proteasome inhibitor on a COG BFM backbone.

Treatment Strategies at Relapse

Relapsed lymphoblastic lymphoma (LBL) still has a dismal outcome, with survival rates of 10–30%. In a Japanese cohort, the incidence of relapse/progression was 18%, with 48 cases among 260 LBL patients diagnosed between 1996 and 2004 [78]. Among 19 patients who underwent allogeneic hematopoietic stem cell transplant (HSCT), 6 suffered relapse, and 3 died of treatment-related mortality (TRM), while 10 survived without further progression. Among the six patients who had undergone autologous HSCT, four suffered relapse and died, while two survived. The Center for International Blood and Marrow Transplant Research of North America (CIBMTR) summarized 53 pediatric LBL patients who received HSCT between 1990 and 2005. The EFS for 39 patients treated with allo-HSCT was 40% compared with 4% in the 14 patients who underwent autologous HSCT [79]. The EORTC focused on LBL of B-cell phenotype and reported a 15% relapse/progression rate of 8 patients out of 53 diagnosed between 1989 and 2008. All these eight patients died, seven after allogeneic HSCT, five patients of disease progression, and three of TRM [7]. The NHL-BFM group reported a relapse rate of 10%, with 34 LBL relapses among 324 LBL patients, diagnosed between 1990 and 2003. Among 13 patients who received allo-HSCT, 5 survived, 6 suffered relapse, and 2 died of TRM. Two patients underwent autologous HSCT and both died of disease progression [80]. Overall, available data in relapsed LBL show that patients without high-dose treatment followed by autologous (auto) or allogeneic HSCT have almost no chance of cure. Concerning the ongoing discussion whether auto or allo-HSCT is superior, the available data indicate a trend for higher TRM but also higher probability of disease-free survival after allogeneic HSCT compared to autologous SCT. However the absolute numbers of cases in the literature is too small to draw definite conclusions.

Novel and Targeted Therapies

Because of the poor retrieval rates at relapse, new targeted and less toxic drugs are needed. Nelarabine was FDA approved based on two phase II trials in pediatric and adult patients with relapsed or refractory T-ALL or T-LBL [81]. Complete remission was achieved in 5/39 pediatric patients.

Further identification and implementation of additional new drugs for high-risk, refractory, or relapsed pediatric T-LBL are needed. A current review on pediatric T-ALL summarizes several potential drugs of interest targeting pathways like Notch, PI3K-Akt-mTOR, JAK-STAT, and MAPK pathways, the cell cycle regulation, the proteasome, or epigenetic targets or using approaches derived from the immunotherapy [82]. Despite the long list, only a limited number of substances are under investigation in T-ALL and none in T-LBL. Delgado-Martin et al. published preclinical data of JAK-STAT pathway inhibition with ruxolitinib, which can overcome intrinsic glucocorticoid resistance in T-ALL [83]. Preclinical data in a panel of patient-derived T-ALL xenograft showed robust expression of CD38 even after chemotherapy and striking efficacy of daratumumab, a CD38 antibody in T-ALL [84]. Recently, it was reported that PIM1, a serine/threonine kinase involved in cell cycle progression, transcription, and apoptosis, has oncogenic activity in T-cell lymphoblastic neoplasms and might act as an attractive molecular target. PIM1 is activated in a significant fraction of human T-ALL and T-LBL patient samples, by rare TCR driven translocations or aberrant activation of the JAK-STAT signaling pathway. Therefore second-generation pan-PIM inhibitors represent a new class of substances that might be evaluated in clinical trials for PIM1 expressing T-LBL patients [85].

Conclusions

Cure rates for pediatric patients with lymphoblastic lymphoma have dramatically improved. However, important challenges remain including identification of prognostic factors and a robust risk classification, increasing the survival of high-risk groups, reduction of acute and long-term toxicity, and developing more targeted treatment approaches with less toxicities.

References

1. Schmidt E, Burkhardt B. Lymphoblastic lymphoma in childhood and adolescence. Pediatr Hematol Oncol. 2013;30(6):484–508.
2. Termuhlen AM, Smith LM, Perkins SL, Lones M, Finlay JL, Weinstein H, et al. Disseminated lymphoblastic lymphoma in children and adolescents: results of the COG A5971 trial: a report from the Children's Oncology Group. Br J Haematol. 2013;162(6):792–801.
3. Landmann E, Burkhardt B, Zimmermann M, Meyer U, Woessmann W, Klapper W, et al. Results and conclusions of the European Intergroup EURO-LB02 trial in children and adolescents with lymphoblastic lymphoma. Haematologica. 2017;102(12):2086–96.

4. Burkhardt B, Zimmermann M, Oschlies I, Niggli F, Mann G, Parwaresch R, et al. The impact of age and gender on biology, clinical features and treatment outcome of non-Hodgkin lymphoma in childhood and adolescence. Br J Haematol. 2005;131(1):39–49.
5. Oschlies I, Burkhardt B, Salaverria I, Rosenwald A, d'Amore ES, Szczepanowski M, et al. Clinical, pathological and genetic features of primary mediastinal large B-cell lymphomas and mediastinal gray zone lymphomas in children. Haematologica. 2011;96(2):262–8.
6. Patel JL, Smith LM, Anderson J, Abromowitch M, Campana D, Jacobsen J, et al. The immunophenotype of T-lymphoblastic lymphoma in children and adolescents: a Children's Oncology Group report. Br J Haematol. 2012;159(4):454–61.
7. Ducassou S, Ferlay C, Bergeron C, Girard S, Laureys G, Pacquement H, et al. Clinical presentation, evolution, and prognosis of precursor B-cell lymphoblastic lymphoma in trials LMT96, EORTC 58881, and EORTC 58951. Br J Haematol. 2011;152(4):441–51.
8. Neth O, Seidemann K, Jansen P, Mann G, Tiemann M, Ludwig WD, et al. Precursor B-cell lymphoblastic lymphoma in childhood and adolescence: clinical features, treatment, and results in trials NHL-BFM 86 and 90. Med Pediatr Oncol. 2000;35(1):20–7.
9. Bene MC, Castoldi G, Knapp W, Ludwig WD, Matutes E, Orfao A, et al. Proposals for the immunological classification of acute leukemias. European Group for the Immunological Characterization of Leukemias (EGIL). Leukemia. 1995;9(10):1783–6.
10. Bene MC, Nebe T, Bettelheim P, Buldini B, Bumbea H, Kern W, et al. Immunophenotyping of acute leukemia and lymphoproliferative disorders: a consensus proposal of the European LeukemiaNet Work Package 10. Leukemia. 2011;25(4):567–74.
11. Borowitz MJC, J KC, Downing JR, Le Beau MM, Arber DA. Precursor lymphoid neoplasms. In: Swerdlow SH, Campo E, Harris NL, Jaffe ES, Plieri SA, Stein H, Thiele J, editors. WHO classification of tumours of haematopoietic and lymphoid tissue. Lyon: International Agency for Research on Cancer (IARC); 2017. p. 200–13.
12. Dworzak MN, Buldini B, Gaipa G, Ratei R, Hrusak O, Luria D, et al. AIEOP-BFM consensus guidelines 2016 for flow cytometric immunophenotyping of Pediatric acute lymphoblastic leukemia. Cytometry B Clin Cytom. 2018;94(1):82–93.
13. Roberts KG, Mullighan CG. Genomics in acute lymphoblastic leukaemia: insights and treatment implications. Nat Rev Clin Oncol. 2015;12(6):344–57.
14. Burkhardt B. Paediatric lymphoblastic T-cell leukaemia and lymphoma: one or two diseases? Br J Haematol. 2010;149(5):653–68.
15. Basso K, Mussolin L, Lettieri A, Brahmachary M, Lim WK, Califano A, et al. T-cell lymphoblastic lymphoma shows differences and similarities with T-cell acute lymphoblastic leukemia by genomic and gene expression analyses. Genes Chromosomes Cancer. 2011;50(12):1063–75.
16. Uyttebroeck A, Vanhentenrijk V, Hagemeijer A, Boeckx N, Renard M, Wlodarska I, et al. Is there a difference in childhood T-cell acute lymphoblastic leukaemia and T-cell lymphoblastic lymphoma? Leuk Lymphoma. 2007;48(9):1745–54.
17. Oschlies I, Burkhardt B, Chassagne-Clement C, d'Amore ES, Hansson U, Hebeda K, et al. Diagnosis and immunophenotype of 188 pediatric lymphoblastic lymphomas treated within a randomized prospective trial: experiences and preliminary recommendations from the European childhood lymphoma pathology panel. Am J Surg Pathol. 2011;35(6):836–44.
18. Smock KJ, Nelson M, Tripp SR, Sanger WG, Abromowitch M, Cairo MS, et al. Characterization of childhood precursor T-lymphoblastic lymphoma by immunophenotyping and fluorescent in situ hybridization: a report from the Children's Oncology Group. Pediatr Blood Cancer. 2008;51(4):489–94.
19. Coustan-Smith E, Mullighan CG, Onciu M, Behm FG, Raimondi SC, Pei D, et al. Early T-cell precursor leukaemia: a subtype of very high-risk acute lymphoblastic leukaemia. Lancet Oncol. 2009;10(2):147–56.
20. Zhang J, Ding L, Holmfeldt L, Wu G, Heatley SL, Payne-Turner D, et al. The genetic basis of early T-cell precursor acute lymphoblastic leukaemia. Nature. 2012;481(7380):157–63.
21. You MJ, Medeiros LJ, Hsi ED. T-lymphoblastic leukemia/lymphoma. Am J Clin Pathol. 2015;144(3):411–22.
22. Haydu JE, Ferrando AA. Early T-cell precursor acute lymphoblastic leukaemia. Curr Opin Hematol. 2013;20(4):369–73.
23. Patrick K, Wade R, Goulden N, Mitchell C, Moorman AV, Rowntree C, et al. Outcome for children and young people with Early T-cell precursor acute lymphoblastic leukaemia treated on a contemporary protocol, UKALL 2003. Br J Haematol. 2014;166(3):421–4.
24. Swerdlow SH, Campo E, Harris NL, Jaffe E, Pileri S, Stein H, et al., editors. WHO classification of tumours of haematopoietic and lymphoid tissue. Lyon: IARC; 2008.
25. Matutes E, Pickl WF, Van't Veer M, Morilla R, Swansbury J, Strobl H, et al. Mixed-phenotype acute leukemia: clinical and laboratory features and outcome in 100 patients defined according to the WHO 2008 classification. Blood. 2011;117(11):3163–71.
26. Boddu P, Yin CC, Kanagal-Shamanna R, Tang G, Thakral B, Kadia T, et al. An Unsuspected Finding of t(9;22): a rare case of Philadelphia chromosome-positive b-lymphoblastic lymphoma. Case Rep Hematol. 2017;2017:2413587.
27. Ahlmann M, Meyer C, Marschalek R, Burkhardt B, Koehler G, Klapper W, et al. Complex MLL rearrangement in non-infiltrated bone marrow in an infant with stage II precursor B-lymphoblastic lymphoma. Eur J Haematol. 2014;93(4):349–53.
28. Maitra A, McKenna RW, Weinberg AG, Schneider NR, Kroft SH. Precursor B-cell lymphoblastic lymphoma. A study of nine cases lacking blood and bone marrow involvement and review of the literature. Am J Clin Pathol. 2001;115(6):868–75.
29. Geethakumari PR, Hoffmann MS, Pemmaraju N, Hu S, Jorgensen JL, O'Brien S, et al. Extramedullary B lymphoblastic leukemia/lymphoma (B-ALL/B-LBL): a diagnostic challenge. Clin Lymphoma Myeloma Leuk. 2014;14(4):e115–8.
30. Meyer JA, Zhou D, Mason CC, Downie JM, Rodic V, Abromowitch M, et al. Genomic characterization of pediatric B-lymphoblastic lymphoma and B-lymphoblastic leukemia using formalin-fixed tissues. Pediatr Blood Cancer. 2017;64(7) https://doi.org/10.1002/pbc.26363.
31. Burkhardt B, Bruch J, Zimmermann M, Strauch K, Parwaresch R, Ludwig WD, et al. Loss of heterozygosity on chromosome 6q14-q24 is associated with poor outcome in children and adolescents with T-cell lymphoblastic lymphoma. Leukemia. 2006;20(8):1422–9.
32. Lones MA, Heerema NA, Le Beau MM, Sposto R, Perkins SL, Kadin ME, et al. Chromosome abnormalities in advanced stage lymphoblastic lymphoma of children and adolescents: a report from CCG-E08. Cancer GenetCytogenet. 2007;172(1):1–11.
33. Breit S, Stanulla M, Flohr T, Schrappe M, Ludwig WD, Tolle G, et al. Activating NOTCH1 mutations predict favorable early treatment response and long term outcome in child-hood precursor T-cell lymphoblastic leukemia. Blood. 2006;108(4):1151–7.
34. Kox C, Zimmermann M, Stanulla M, Leible S, Schrappe M, Ludwig WD, et al. The favorable effect of activating NOTCH1 receptor mutations on long-term outcome in T-ALL patients treated on the ALL-BFM 2000 protocol can be separated from FBXW7 loss of function. Leukemia. 2010;24(12):2005–13.
35. Bonn BR, Rohde M, Zimmermann M, Krieger D, Oschlies I, Niggli F, et al. Incidence and prognostic relevance of genetic variations in T-cell lymphoblastic lymphoma in childhood and adolescence. Blood. 2013;121(16):3153–60.
36. Callens C, Baleydier F, Lengline E, Ben Abdelali R, Petit A, Villarese P, et al. Clinical impact of NOTCH1 and/or FBXW7 mutations, FLASH deletion, and TCR status in pediatric T-cell lymphoblastic lymphoma. J Clin Oncol. 2012;30(16):1966–73.

37. Park MJ, Taki T, Oda M, Watanabe T, Yumura-Yagi K, Kobayashi R, et al. FBXW7 and NOTCH1 mutations in childhood T cell acute lymphoblastic leukaemia and T cell non-Hodgkin lymphoma. Br J Haematol. 2009;145(2):198–206.
38. Baleydier F, Decouvelaere AV, Bergeron J, Gaulard P, Canioni D, Bertrand Y, et al. T cell receptor genotyping and HOXA/TLX1 expression define three T lymphoblastic lymphoma subsets which might affect clinical outcome. Clin Cancer Res. 2008;14(3):692–700.
39. Burkhardt B, Moericke A, Klapper W, Greene F, Salzburg J, Damm-Welk C, et al. Pediatric precursor T lymphoblastic leukemia and lymphoblastic lymphoma: differences in the common regions with loss of heterozygosity at chromosome 6q and their prognostic impact. LeukLymphoma. 2008;49(3):451–61.
40. Balbach ST, Makarova O, Bonn BR, Zimmermann M, Rohde M, Oschlies I, et al. Proposal of a genetic classifier for risk group stratification in pediatric T-cell lymphoblastic lymphoma reveals differences from adult T-cell lymphoblastic leukemia. Leukemia. 2016;30(4):970–3.
41. Song MS, Salmena L, Pandolfi PP. The functions and regulation of the PTEN tumour suppressor. Nat Rev Mol Cell Biol. 2012;13(5):283–96.
42. Hales EC, Taub JW, Matherly LH. New insights into Notch1 regulation of the PI3K-AKT-mTOR1 signaling axis: targeted therapy of gamma-secretase inhibitor resistant T-cell acute lymphoblastic leukemia. Cell Signal. 2014;26(1):149–61.
43. Bandapalli OR, Zimmermann M, Kox C, Stanulla M, Schrappe M, Ludwig WD, et al. NOTCH1 activation clinically antagonizes the unfavorable effect of PTEN inactivation in BFM-treated children with precursor T-cell acute lymphoblastic leukemia. Haematologica. 2013;98(6):928–36.
44. Paganin M, Grillo MF, Silvestri D, Scapinello G, Buldini B, Cazzaniga G, et al. The presence of mutated and deleted PTEN is associated with an increased risk of relapse in childhood T cell acute lymphoblastic leukaemia treated with AIEOP-BFM ALL protocols. Br J Haematol. 2018;182(5):705–11.
45. Zuurbier L, Homminga I, Calvert V, te Winkel ML, Buijs-Gladdines JG, Kooi C, et al. NOTCH1 and/or FBXW7 mutations predict for initial good prednisone response but not for improved outcome in pediatric T-cell acute lymphoblastic leukemia patients treated on DCOG or COALL protocols. Leukemia. 2010;24(12):2014–22.
46. Gutierrez A, Dahlberg SE, Neuberg DS, Zhang J, Grebliunaite R, Sanda T, et al. Absence of biallelic TCRgamma deletion predicts early treatment failure in pediatric T-cell acute lymphoblastic leukemia. J Clin Oncol. 2010;28(24):3816–23.
47. Yang YL, Hsiao CC, Chen HY, Lin KH, Jou ST, Chen JS, et al. Absence of biallelic TCRgamma deletion predicts induction failure and poorer outcomes in childhood T-cell acute lymphoblastic leukemia. Pediatr Blood Cancer. 2012;58(6):846–51.
48. Pomari E, Lovisa F, Carraro E, Primerano S, D'Amore ESG, Bonvini P, et al. Clinical impact of miR-223 expression in pediatric T-Cell lymphoblastic lymphoma. Oncotarget. 2017;8(64):107886–98.
49. Burkhardt B, Mueller S, Khanam T, Perkins SL. Current status and future directions of T-lymphoblastic lymphoma in children and adolescents. Br J Haematol. 2016;173(4):545–59.
50. Tubergen DG, Krailo MD, Meadows AT, Rosenstock J, Kadin M, Morse M, et al. Comparison of treatment regimens for pediatric lymphoblastic non-Hodgkin's lymphoma: a Childrens Cancer Group study. J Clin Oncol. 1995;13(6):1368–76.
51. Burkhardt B, Oschlies I, Klapper W, Zimmermann M, Woessmann W, Meinhardt A, et al. Non-Hodgkin's lymphoma in adolescents: experiences in 378 adolescent NHL patients treated according to pediatric NHL-BFM protocols. Leukemia. 2011;25(1):153–60.
52. Uyttebroeck A, Suciu S, Laureys G, Robert A, Pacquement H, Ferster A, et al. Treatment of childhood T-cell lymphoblastic lymphoma according to the strategy for acute lymphoblastic leukaemia, without radiotherapy: long term results of the EORTC CLG 58881 trial. Eur J Cancer. 2008;44(6):840–6.
53. Termuhlen AM, Smith LM, Perkins SL, Lones M, Finlay JL, Weinstein H, et al. Outcome of newly diagnosed children and adolescents with localized lymphoblastic lymphoma treated on Children's Oncology Group trial A5971: a report from the Children's Oncology Group. Pediatr Blood Cancer. 2012;59(7):1229–33.
54. Stark B, Avigad S, Luria D, Manor S, Reshef-Ronen T, Avrahami G, et al. Bone marrow minimal disseminated disease (MDD) and minimal residual disease (MRD) in childhood T-cell lymphoblastic lymphoma stage III, detected by flow cytometry (FC) and real-time quantitative polymerase chain reaction (RQ-PCR). Pediatr Blood Cancer. 2009;52(1):20–5.
55. Coustan-Smith E, Sandlund JT, Perkins SL, Chen H, Chang M, Abromowitch M, et al. Minimal disseminated disease in childhood T-cell lymphoblastic lymphoma: a report from the children's oncology group. J Clin Oncol. 2009;27(21):3533–9.
56. Mussolin L, Buldini B, Lovisa F, Carraro E, Disaro S, Nigro LL, et al. Detection and role of minimal disseminated disease in children with lymphoblastic lymphoma: the AIEOP experience. Pediatr Blood Cancer. 2015;62(11):1906–13.
57. Bardi E, Csoka M, Garai I, Szegedi I, Muller J, Gyorke T, et al. Value of FDG-PET/CT examinations in different cancers of children, focusing on lymphomas. Pathol Oncol Res. 2014;20(1):139–43.
58. Nakatani K, Nakamoto Y, Watanabe K, Saga T, Higashi T, Togashi K. Roles and limitations of FDG PET in pediatric non-Hodgkin lymphoma. Clin Nucl Med. 2012;37(7):656–62.
59. Riad R, Omar W, Kotb M, Hafez M, Sidhom I, Zamzam M, et al. Role of PET/CT in malignant pediatric lymphoma. Eur J Nucl Med Mol Imaging. 2010;37(2):319–29.
60. Riad R, Omar W, Sidhom I, Zamzam M, Zaky I, Hafez M, et al. False-positive F-18 FDG uptake in PET/CT studies in pediatric patients with abdominal Burkitt's lymphoma. Nucl Med Commun. 2010;31(3):232–8.
61. Sioka C. The utility of FDG PET in diagnosis and follow-up of lymphoma in childhood. Eur J Pediatr. 2013;172(6):733–8.
62. Murphy SB. Classification, staging and end results of treatment of childhood non-Hodgkin's lymphomas: dissimilarities from lymphomas in adults. Semin Oncol. 1980;7(3):332–9.
63. Rosolen A, Perkins SL, Pinkerton CR, Guillerman RP, Sandlund JT, Patte C, et al. Revised International Pediatric Non-Hodgkin Lymphoma Staging System. J Clin Oncol. 2015;33(18):2112–8.
64. Wollner N, Burchenal JH, Lieberman PH, Exelby P, D'Angio G, Murphy ML. Non-Hodgkin's lymphoma in children. A comparative study of two modalities of therapy. Cancer. 1976;37(1):123–34.
65. Wollner N, Exelby PR, Lieberman PH. Non-Hodgkin's lymphoma in children: a progress report on the original patients treated with the LSA2-L2 protocol. Cancer. 1979;44(6):1990–9.
66. Reiter A, Schrappe M, Parwaresch R, Henze G, Muller-Weihrich S, Sauter S, et al. Non-Hodgkin's lymphomas of childhood and adolescence: results of a treatment stratified for biologic subtypes and stage--a report of the Berlin-Frankfurt-Munster Group. J Clin Oncol. 1995;13(2):359–72.
67. Reiter A, Schrappe M, Ludwig WD, Tiemann M, Parwaresch R, Zimmermann M, et al. Intensive ALL-type therapy without local radiotherapy provides a 90% event-free survival for children with T-cell lymphoblastic lymphoma: a BFM group report. Blood. 2000;95(2):416–21.
68. Jin L, Zhang R, Huang S, Yang J, Duan YL, Zhang YH. Clinical features and prognosis of children with lymphoblastic lymphoma. Zhonghua Zhong Liu Za Zhi. 2012;34(2):138–42.
69. Kobayashi R, Takimoto T, Nakazawa A, Fujita N, Akazai A, Yamato K, et al. Inferior outcomes of stage III T lymphoblastic lymphoma relative to stage IV lymphoma and T-acute lymphoblastic leukemia: long-term comparison of outcomes in the JACLS NHL T-98 and ALL T-97 protocols. Int J Hematol. 2014;99(6):743–9.

70. Sun XF, Xia ZJ, Zhen ZJ, Xiang XJ, Xia Y, Ling JY, et al. Intensive chemotherapy improved treatment outcome for Chinese children and adolescents with lymphoblastic lymphoma. Int J Clin Oncol. 2008;13(5):436–41.
71. Gao YJ, Pan C, Tang JY, Lu FJ, Chen J, Xue HL, et al. Clinical outcome of childhood lymphoblastic lymphoma in Shanghai China 2001-2010. Pediatr Blood Cancer. 2014;61(4):659–63.
72. Sunami S, Sekimizu M, Takimoto T, Mori T, Mitsui T, Fukano R, et al. Prognostic impact of intensified maintenance therapy on children with advanced lymphoblastic lymphoma: a report from the Japanese Pediatric Leukemia/Lymphoma Study Group ALB-NHL03 study. Pediatr Blood Cancer. 2016;63(3):451–7.
73. Patte C, Kalifa C, Flamant F, Hartmann O, Brugieres L, Valteau-Couanet D, et al. Results of the LMT81 protocol, a modified LSA2L2 protocol with high dose methotrexate, on 84 children with non-B-cell (lymphoblastic) lymphoma. Med Pediatr Oncol. 1992;20(2):105–13.
74. Asselin BL, Devidas M, Wang C, Pullen J, Borowitz MJ, Hutchison R, et al. Effectiveness of high-dose methotrexate in T-cell lymphoblastic leukemia and advanced-stage lymphoblastic lymphoma: a randomized study by the Children's Oncology Group (POG 9404). Blood. 2011;118(4):874–83.
75. Sterba J, Kovacs G, Matus M, Bubanska E, Kyr M, Krenova Z, et al. Capizzi methotrexate with BFM backbone without craniospinal irradiation is effective treatment for pediatric lymphoblastic lymphoma: results from 5 countries with I-BFM LL 09 protocol. Br J Haematol. 2015;171(Suppl. 1):33.
76. Winter SS, Dunsmore KP, Devidas M, Wood BL, Esiashvili N, Chen Z, et al. Improved survival for children and young adults with T-lineage acute lymphoblastic leukemia: results from the Children's Oncology Group AALL0434 methotrexate randomization. J Clin Oncol. 2018;36(29):2926–34. https://doi.org/10.1200/JCO.2018.77.7250.
77. Dunsmore KP, Winter S, Devidas M, Wood BL, Esiashvili N, Eisenberg N, Briegel N, Hayashi RJ, Gastier-Foster JM, Carroll AJ, Heerema N, Asselin B, Rabin KR, Zweidler-McKay P, Raetz RA, Loh ML, Winick NJ, Carroll WL, Hunger S. COG AALL0434: a randomized trial testing nelarabine in newly diagnosed t-cell malignancy. J Clin Oncol. 2018;36:10500.
78. Mitsui T, Mori T, Fujita N, Inada H, Horibe K, Tsurusawa M. Retrospective analysis of relapsed or primary refractory childhood lymphoblastic lymphoma in Japan. Pediatr Blood Cancer. 2009;52:591.
79. Gross TG, Hale GA, He W, Camitta BM, Sanders JE, Cairo MS, et al. Hematopoietic stem cell transplantation for refractory or recurrent non-Hodgkin lymphoma in children and adolescents. Biol Blood Marrow Transplant. 2009;16(2):223–30.
80. Burkhardt B, Zimmermann M, Oschlies I, Klapper W, Meinhardt A, Landmann E, et al., editors. Outcome of adolescents with non-Hodgkin lymphoma in the BFM studies: relevance of gender and histological subtype. In: 3rd international symposium on childhood, adolescent and young adult non-Hodgkin's Lymphoma, Frankfurt: Hematology meeting reports; June 2009.
81. Cohen MH, Johnson JR, Justice R, Pazdur R. FDA drug approval summary: nelarabine (Arranon) for the treatment of T-cell lymphoblastic leukemia/lymphoma. Oncologist. 2008;13(6):709–14.
82. Raetz EA, Teachey DT. T-cell acute lymphoblastic leukemia. Hematology Am Soc Hematol Educ Program. 2016;2016(1):580–8.
83. Delgado-Martin C, Meyer LK, Huang BJ, Shimano KA, Zinter MS, Nguyen JV, et al. JAK/STAT pathway inhibition overcomes IL7-induced glucocorticoid resistance in a subset of human T-cell acute lymphoblastic leukemias. Leukemia. 2017;31(12):2568–76.
84. Bride KL, Vincent TL, Im SY, Aplenc R, Barrett DM, Carroll WL, et al. Preclinical efficacy of daratumumab in T-cell acute lymphoblastic leukemia. Blood. 2018;131(9):995–9.
85. De Smedt R, Peirs S, Morscio J, Matthijssens F, Roels J, Reunes L, et al. Preclinical evaluation of second generation PIM inhibitors for the treatment of T-cell acute lymphoblastic leukemia and lymphoma. Haematologica. 2019;104(1):e17–20.
86. Amylon MD, Shuster J, Pullen J, Berard C, Link MP, Wharam M, et al. Intensive high-dose asparaginase consolidation improves survival for pediatric patients with T cell acute lymphoblastic leukemia and advanced stage lymphoblastic lymphoma: a Pediatric Oncology Group study. Leukemia. 1999;13(3):335–42.
87. Burkhardt B, Woessmann W, Zimmermann M, Kontny U, Vormoor J, Doerffel W, et al. Impact of cranial radiotherapy on central nervous system prophylaxis in children and adolescents with central nervous system-negative stage III or IV lymphoblastic lymphoma. J Clin Oncol. 2006;24(3):491–9.
88. Abromowitch M, Sposto R, Perkins S, Zwick D, Siegel S, Finlay J, et al. Shortened intensified multi-agent chemotherapy and non-cross resistant maintenance therapy for advanced lymphoblastic lymphoma in children and adolescents: report from the Children's Oncology Group. Br J Haematol. 2008;143(2):261–7.
89. Abromowitch M, Termuhlen A, Chang M, Perkins S, Gross T, Weinstein H, et al. High-dose methotrexate and early intensification of therapy do not improve 3 year EFS in children and adolescents with disseminated lymphoblastic lymphoma. Results of the randomized arms of COG A5971. Blood. 2008;12:Abstract No. 3610.
90. Pillon M, Piglione M, Garaventa A, Conter V, Giuliano M, Arcamone G, et al. Long-term results of AIEOP LNH-92 protocol for the treatment of pediatric lymphoblastic lymphoma: a report of the Italian Association of pediatric hematology and oncology. Pediatr Blood Cancer. 2009;53(6):953–9.
91. Sandlund JT, Pui CH, Zhou Y, Behm FG, Onciu M, Razzouk BI, et al. Effective treatment of advanced-stage childhood lymphoblastic lymphoma without prophylactic cranial irradiation: results of St Jude NHL13 study. Leukemia. 2009;23(6):1127–30.
92. Uyttebroeck A, Suciu S, Plat G, Laureys G, Costa V, Rohrlich P, et al. Dexamethasone (DEX) versus prednisone (PRED) in T-cell non Hodgkin lymphoma (T-NHL): results of the randomized phase III trial 58951 of the EORTC Children Leukemia Group. Br J Haematol. 2012;159(Suppl. 1):37.
93. Bergeron C, Coze C, Segura C, Pacquement H, Gandemer V, Ducassou S, et al. Treatment of childhood T-cell lymphoblastic lymphoma-long-term results of the SFOP LMT96 trial. Pediatr Blood Cancer. 2015;62(12):2150–6.

Part V

Mature B- Non-Hodgkin's Lymphomas

Burkitt Lymphoma and Diffuse Large B-Cell Lymphoma

13

Grace Egan, Sheila Weitzman, and Sarah Alexander

Introduction

Mature B-cell lymphomas account for 50–60% of pediatric NHL. This chapter will focus on the two most common subtypes, Burkitt lymphoma (BL) and diffuse large B-cell lymphoma (DLBCL). Rarer subtypes, including primary mediastinal B-cell lymphoma (PMBCL), will be discussed elsewhere.

The survival of children and adolescents with BL and DLBCL has improved dramatically over the past 60 years. Despite the differences in biology, childhood BL and DLBCL have been treated similarly [15]. Success has been driven by sequential clinical trials of multi-agent intensive chemotherapy based on disease risk features, as well as the incorporation of immunotherapy in recent trials.

This chapter will review epidemiology, biology, and clinical presentation of BL and DLBCL and discuss treatment strategies for these diseases.

Burkitt Lymphoma

Epidemiology

There are three subgroups of BL, namely, endemic, sporadic, and immunodeficiency-associated.

Endemic Burkitt Lymphoma

Denis Burkitt was the first to describe BL in equatorial Africa in the late 1950s in children presenting with jaw tumors and/or abdominal tumors [12]. BL accounts for approximately 30–50% of all pediatric cancers in equatorial Africa [50], and BL most commonly affects children ages 4–7 years, with a male-female ratio of 2:1.

Sporadic Burkitt Lymphoma

By contrast, sporadic BL accounts for less than 5% of all pediatric cancers in the developed countries, though it is the most common subtype of NHL. There is a striking male predominance with an incidence of 3.2 new cases per million in boys compared to 0.7 cases per million in girls less than 20 years of age [82], with 5–14-year-olds being the most common age group affected [82].

Immunodeficiency-Related Burkitt Lymphoma

In the early 1980s, patients with human immunodeficiency virus (HIV) infection were noted to have a propensity to develop NHL including BL [118]. This observation led to the inclusion of immunodeficiency-related BL in the subsequent WHO classification of hematological malignancies [106]. In adults, BL is commonly associated with immunodeficiency, particularly HIV related. Children with primary and secondary immunodeficiencies, including solid organ transplant-related immunosuppression, are also at increased risk of developing mature B-NHL [28, 40, 98].

Morphology

Endemic and sporadic BL are morphologically indistinguishable [53] with both forms exhibiting a diffuse pattern of infiltration by intermediate-sized undifferentiated homogenous cells with round to oval nuclei with multiple, variably prominent, basophilic nucleoli [85] (Table 13.1). On cytological preparations, the narrow rim of cytoplasm is

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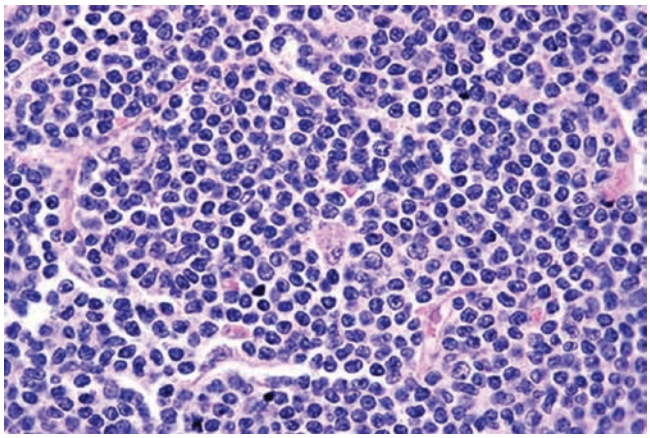
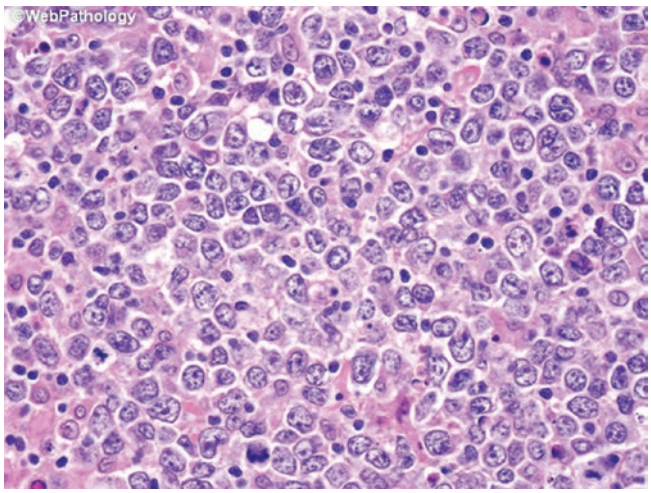
Table 13.1 Morphology, immunophenotype, and cytogenetic characteristics of pediatric mature B-NHL

Disease	Histology	Immunophenotype	Cytogenetics
Burkitt Lymphoma	Intermediate-sized homogenous cells with round nuclei. Multiple prominent basophilic nucleoli [106]. Basophilic cytoplasm with lipid vacuoles 	Usually positive for B-cell-associated antigens [85]: CD10, CD19, CD20, CD22, BCL6, CD38, CD43 Membrane IgM and light chain restriction Ki-67 or MIB-1 >90% Negative for TdT	cMYC translocations: t(8, 14) (q24;q32) in 80% Others: t(2, 8) (p11;q24) t(8, 22) (q24;q11)
DLBCL	Variable but with large lymphoid cells that in nodal tissue efface nodal architecture 	Usually positive for B-cell-associated antigens: CD19, CD20, CD22 Positive for surface immunoglobulin Ki-67 or MIB-1 <90% Variable for: CD10, BCL6, BCL2, CD30, MUM1 Negative for TdT	Can have complex karyotype with structural and numerical abnormalities

basophilic and appears vacuolated because of the presence of lipid droplets [85]. Given the tumors extremely high rate of proliferation and apoptosis, tissue sections often show a "starry sky" appearance that results from reactive macrophages which spread among the rapidly dividing clonal lymphoid cells and phagocytose cellular debris [83]. This appearance is not specific for BL and can be seen with other rapidly dividing tumors [83].

Immunophenotype and Immunohistochemistry

Immunophenotypic analysis of BL demonstrates an expression of B-cell-related antigens including CD19, CD20, CD22, CD79a, and PAX5 [37] as well as clonal immunoglobulin heavy and light chain antigens and the leukocyte antigens CD45 and CD43. Reflecting the germinal center subtype of BL, the surface antigen CD10 is usually expressed in BL, as is the protein product of the B-cell lymphoma 6 (*BCL6*) gene [59]. BL cells are negative for terminal deoxynucleotidyl transferase (TdT) [19].

The high mitotic rate (>90%) [19] makes staining with proliferative markers Ki-67 and/or MIB1 a useful tool in differentiation of BL from other intermediate- and high-grade mature B-NHL. The presence of antigens such as the multiple myeloma antigen, MUM1/IRF4, and BCL2 by immunostaining is occasionally seen in sporadic BL [8].

Pathogenesis of BL

EBV infection at a young age on a background of intense malaria infection has been suggested to be an important fac-

tor in the development of BL in equatorial Africa [50]. The correlation between malaria infection and risk of BL [90] has been supported by in vitro findings showing that malarial extracts cause proliferation of B cells and MYC immunoglobulin rearrangements, as well as indirect in vivo evidence showing a decrease in incidence of BL in areas where malaria is controlled.

Molecular studies of BL demonstrate clonal immunoglobulin gene rearrangements. BL was, in fact, the first human tumor in which a chromosomal translocation was found to be involved in its pathogenesis [20]. The immunoglobulin heavy chain gene, located at chromosome 14, band q32, is interrupted and involved in translocations involving the *cMYC* gene which is found on chromosome 8q24, t(8;14)(q24;q32). This translocation, present in approximately 80% of BL cases [2], results in aberrant overexpression of the oncoprotein cMYC in BL cells. Less commonly, BL may be associated with translocations involving the *cMYC* gene and either the kappa light-chain gene on chromosome 2p12, t(2;8)(p12;q24), or the lambda light-chain gene on chromosome 22q11, t(8;22)(q24;q11). The underlying translocation does not appear to affect the clinical presentation or the outcome.

Differences in the chromosome 8 and 14 breakpoint locations have been noted in endemic and sporadic BL [81]. Endemic tumors have breakpoints more than 100 kb upstream of the first coding exon of the *cMYC* oncogene, while the Ig breakpoint occurs in the VDJ region of the *IgH* gene [2]. Sporadic BL tends to have breakpoints between exon 1 and 2 of *cMYC* and within the *IgH* gene class switch region [2, 81].

Additional chromosomal alterations can occur in pediatric BL, including gains at 1q and 7q and losses at 13q [89]. Chromosome 22 abnormalities, independent of t(8;22), have also been found in pediatric BL [74]. These additional chromosomal abnormalities are more common in pediatric BL than adult BL [89]. For pediatric BL, complex karyotypes appear to be associated with an inferior outcome [74, 89].

More recently, molecular diagnostics has expanded our understanding of the biology of NHL with gene expression profiling (GEP) enabling more accurate classifications of mature B-NHL. A molecular signature for adult BL was first described using GEP and matrix comparative genome hybridization (CGH) in 2006 [21, 38]. This molecular signature was subsequently confirmed in pediatric BL [42]. BL is defined molecularly by the upregulation of *MYC* target genes and germinal center (GC) B-cell genes and decreased expression of nuclear factor *(NF)-κB*-associated genes and MHC class I genes and has been shown to cluster separately from DLBCL [21]. This molecular signature remains stable in both pediatric and adult BL and confirms what was noted in earlier studies; that BL is a more homogenous disease with respect to gene expression and genetic aberrations, compared to DLBCL. With regard to the distinct subtypes of BL, recent miRNA profiling data has demonstrated that the three subtypes of BL are also similar to each other and represent the same biologic entity, with only minor miRNA expression profile differences between sporadic and endemic BL [47].

In addition to *MYC* deregulation, high-throughput sequencing has enabled the discovery of a number of additional genetic mutations in BL. Pediatric BL appears to have an increased number of additional mutations compared to adult BL [30]. *TP53* appears to be a common alteration [30], along with *TCF3* and/or *ID3*, its negative regulator. Mutations in *ID3* have been found in 34–58% of BL cases and were associated with the proliferation of BL cells, indicating that *ID3* might function as a tumor suppressor gene [49, 102]. Mutations in subunits of *SWI/SNF*, a nucleosome-remodeling complex that has tumor suppressor functions, have also been discovered, primarily in *ARID1A* or *SMARCA4* [30, 49]. *CCND3*, a regulator of the G1-S phase transition, was found to be mutated in 38% of sporadic BL samples but uncommon in endemic BL tumors [102]. Interestingly, Giulino-Roth and colleagues found that EBV-negative pediatric tumors are more likely to have additional mutations than EBV-positive tumors [30].

Co-expression of BLC2 and cMYC (double-hit lymphoma) has not been associated with an adverse outcome in pediatric BL [54]; however when present in adults, this "double hit" appears to infer a worse prognosis. It has been speculated whether the more intensive treatment given in the pediatric protocols negates the prognostic implication of MYC-BCL2 co-expression [54].

Diffuse Large B-Cell Lymphoma

Epidemiology

DLBCL is less common in children accounting for 10–20% of pediatric NHL [10], in contrast to adults where it represents the most common form of NHL. Before age 4, DLBCL is rarely seen; thereafter its incidence increases steadily with age. Similar to BL, males have a higher incidence of disease (M/F = 1.4), although this is not as striking as the male preponderance in BL [82]. DLBCL is a heterogenous group of lymphoid neoplasms [93], particularly in adults, as opposed to the homogeneity exhibited with BL. The current WHO classification of DLBCL includes a group of clinically and biologically different subtypes, which is likely to increase in the future as molecular diagnostics evolve. Pediatric and adult DLBCL also show differences with respect to morphology, immunophenotype, and genetics, with DLBCL in children appearing to have a more homogenous genomic landscape based on recent studies [41].

Preexisting cancer predisposition syndromes are now recognized as a significant risk factor for developing NHL [6]. Defects in DNA repair, particularly in patients with ataxia-telangiectasia (AT), result in an increased risk of developing mature B-cell NHL, with DLBCL nearly three times more frequent than BL. Inherited immune deficiency may also be

more common in DLBCL compared to BL, with data from the BFM group indicating a rate of 6% of immune deficiency syndromes in those with DLBCL compared to 1% in patients with BL [93].

Morphology

DLBCL is composed of large cells with a variety of morphologic appearances that have vesicular nuclei, prominent nucleoli, basophilic cytoplasm, and a moderate-to-high proliferation fraction [19, 37]. The nuclei are larger than the size of a tissue histiocyte or twice the size of a small lymphocyte [83]. The cytoplasm is more abundant than in BL and varies from pale to plasmacytoid. The WHO guidelines classify DLBCL into morphological variants including centroblastic, immunoblastic, anaplastic, and plasmablastic variants [106] with the centroblastic morphology predominating in pediatric DLBCL [75].

Immunophenotype and Immunohistochemistry

Immunophenotypic analysis of DLBCL demonstrates a mature B-cell phenotype with expression of CD20 and CD79a in most cases [19]. In the few cases that are CD20/CD79a negative, expression of other B-cell markers, including CD19, PAX5, or monoclonal immunoglobulin chains, is detected. They do not express TdT. The majority of DLBCL tumors in pediatrics demonstrate a high proliferation rate as demonstrated by the high expression of Ki-67 or MIB-1 [57], although usually not as high as BL. Most pediatric DLBCL are of the GC subtype and also express CD10 and BCL6 with a minority expressing the post-GC marker MUM1 [75]. Immunohistochemical staining for cMYC protein can be positive [25]. BCL2 protein expression is more commonly seen in pediatric DLBCL than BL [25] but is rarely associated with the t(14:18) translocation [57].

Pathogenesis

Similar to BL, DLBCL involves the malignant transformation of mature B cells. In contrast to BL, there is no common genetic feature defining DLBCL. In adult DLBCL, a multitude of genetic lesions and molecular events have been described, but these results are generally not applicable to the pediatric population with DLBCL. Gene expression profiling studies have enabled the identification of two distinct DLBCL subtypes: germinal center B cell-like (GCB) or activated B cell-like (ABC) [96]. In adults there is a clear prognostic difference in favor of the GCB subtype. Pediatric DLBCL are for the most part GCB subtype, but in contrast to adults where t(14;18) is frequently seen in the GCB subtype, this translocation is rarely seen in childhood DLBCL [75]. The age at which the GCB subtype changes to the ABC subtype represents a continuous variable with the AYA population continuing to exhibit more GCB disease, similar to younger children [41]. Biomarkers that are prognostic in adult DLBCL, including expression of BCL2 and BCL6, do not seem to be prognostic in pediatric DLBCL, indicating DLBCL in childhood may represent a more homogenous, biologically unique subgroup [75].

Pediatric patients with DLBCL have more frequent *MYC* rearrangements and copy number gains compared to adults [22, 89]. Some data suggests that DLBCL when defined molecularly may be less common in the pediatric population than when classification is done by standard diagnostic techniques including morphology and immunohistochemistry [42]. One GEP study reclassified 31% of DLBCL as molecular BL [42]. Meanwhile, another GEP study had the opposite finding with 10/57 morphologically diagnosed BLs reclassified as molecular DLBCL and 2/13 morphologically diagnosed DLBCL reclassified as molecular BL. In molecular-classified DLBCL, frequent abnormalities of 8q24 were found, with *MYC* rearrangements in 31% of samples and gain or amplification affecting the *MYC* locus in 50% of non-rearranged cases [22].

Intermediate Lymphoma

The term "Burkitt-like lymphoma" was used in the past for cases that share features in common with BL but have atypical morphologic features [16]. This term was deemed obsolete by the most recent WHO guidelines, given the difficulty that even expert hematopathologists have in distinguishing these tumors [1]. "Atypical Burkitt lymphoma" is the current term used to indicate a high-grade B-cell lymphoma that is not readily classified as either BL or DLBCL [16].

Although there is evidence in adults to suggest that an "atypical Burkitt lymphoma" may represent a true intermediate subgroup of tumors that can have complex karyotypes, simultaneous t(8;14) and t(14;18) translocation, and aggressive behavior [21], this may not be the case for pediatric BL. Molecular intermediates in children do not appear to harbor a genetic "double hit" and have more BL qualities than adult molecular intermediates, suggesting that that the majority of molecular intermediates in children and young adults are, in fact, BL [42] and should be treated as BL.

Clinical Presentation

Burkitt Lymphoma

The most common signs and symptoms at the time of presentation of a patient with sporadic BL are related to disease in the abdomen. Symptoms may include abdominal pain,

distension, and symptoms related to bowel obstruction, intussusception, or perforation. The head and neck is involved in 15–20% of sporadic cases of BL [78]. Bone marrow disease occurs in approximately 20% of patients, and these children may present with signs and symptoms related to cytopenias including pallor, bleeding, and fevers. Children with CNS disease can present with symptoms of leptomeningeal disease including headache, cranial nerve palsies, or spinal cord compression.

Patients with endemic BL classically present with growing masses in the bones of the jaw or maxilla; less commonly the kidneys, gastrointestinal (GI) tract, and other extra-nodal sites are involved.

The rapid growth of these tumors often leads to children presenting unwell with evidence of organ dysfunction and with evidence of spontaneous tumor lysis.

Diffuse Large B-Cell Lymphoma

Compared to BL patients, children with DLBCL more frequently present with localized disease with focal lesions in the liver, spleen, lung, or mediastinum. Ascites or pleural effusions are not commonly seen [93]. CNS involvement in children is uncommon in DLBCL. The rare cases with DLBCL and CNS involvement tend to have parenchymal disease rather than meningeal disease [99]. Bone marrow involvement is less common than in BL. In terms of mediastinal disease, DLBCL involving the mediastinum usually affects mediastinal lymph nodes, in comparison to PBMCL, which is predominantly located within the thymic area of the mediastinum [93]. Whether the distinct DLBCL variants as defined by the WHO differ with respect to their clinical presentation in children cannot be determined due to the rarity of non-GCB disease in the pediatric population.

Diagnostic Evaluation

Evaluation of a child with suspected mature B-cell lymphoma requires a systematic approach and efficiency, given that many patients will present with rapidly evolving organ impairment from quickly growing tumor.

Obtaining an adequate pathologic sample is critical. If the disease is localized and complete excision is feasible, this should be done as the amount of chemotherapy required can be significantly decreased. If complete excision is not feasible, then a biopsy should be performed and more aggressive surgical options, which may delay chemotherapy, should be avoided. The diagnosis requires assessment of morphology, immunophenotypic features, and genetic features.

Pathologic staging includes bilateral bone marrow aspirates and biopsies and an evaluation of the CSF. Radiographic staging should include evaluation of the chest, abdomen, and pelvic region. In some countries this involves chest X-ray and abdominal ultrasound, whereas in higher-income countries, MRI and/or CT of the neck, chest, abdomen, and pelvis is usually performed with or without PET FDG scanning.

Evaluation for organ dysfunction and signs/symptoms of potential tumor lysis syndrome is critical.

Staging and Risk Group Allocation

Staging for children with NHL has traditionally been done according to the Murphy (St. Jude) staging system [61]. Newer pathologic and imaging modalities for disease detection including flow cytometry, molecular diagnostics, and functional imaging are not included in this system. The International Pediatric NHL Staging System published in 2015, is a revision of the prior system that maintains the general structure of the Murphy Staging System but offers more explicit descriptions with regard to site of disease and extra-nodal extension and incorporates recent diagnostic strategies including FISH analysis and PCR [97] (see Table 13.2).

Risk group allocation, which is critical for therapeutic decision-making, incorporates stage, disease site, surgical resectability, and LDH (see Table 13.3). The BFM protocols have incorporated both stage and LDH in risk group

Table 13.2 Revised international pediatric non-Hodgkin lymphoma staging system

Stage 1	Single tumor with exclusion of the mediastinum and abdomen
Stage 2	1. Single extra-nodal tumor with regional node involvement 2. ≥2 nodal areas on the same side of the diaphragm 3. Primary GI tract tumor ± involvement of associated mesenteric nodes that is completed resected.
Stage 3	1. ≥2 extra-nodal tumor above and/or below the diaphragm 2. ≥2 nodal areas above and below the diaphragm 3. Any intrathoracic tumor 4. Any intraabdominal or retroperitoneal disease (except for primary GI track meeting criteria for stage 2) 5. Any paraspinal or epidural tumor 6. Single bone lesion with concomitant extra-nodal and/or non-regional nodal site
Stage 4	Any of the above with involvement of the central nervous system[a] or bone marrow[b]

Adapted from Rosolen et al. [97]

CSFm CSF positive by morphology, *CSFi* CSF positive by immunophenotype method, *CSFc* CSF positive by cytogenetics, *CSFmol* CSF positive by PCR-based assay

[a]CNS positivity is defined as either (1) any CNS tumor mass identified by imaging, (2) cranial nerve palsy that cannot be explained by extradural lesions, or (3) blasts morphologically identified in CSF. The type of CNS involvement should be specified

[b]Bone marrow involvement (stage IV disease) is currently defined by morphologic evidence of ≥5% blasts or lymphoma cells by BM aspiration. BM positivity by flow cytometry, cytogenetic, or molecular techniques will be specified and the degree of BM involvement reported but will not change the assigned stage if BM morphology is <5% blasts

Table 13.3 Risk group definitions used by BFM and LMB-FAB recent trials

Group	Definition	% of patients
BFM-NHL-95 [114]		
R1	Stages I + II, resected	10%
R2	Stages I + II, not resected; stage III, LDH <500 U/L	46%
R3	Stage III, LDH 500-999 U/L; Stage IV and LDH <1000 U/L and CNS neg	16%
R4	Stage III and IV and LDH >1000 U/L and/or CNS pos	28%
LMB-FAB risks group and division of group B on the International B-NHL 2010 (study included only patients with group B, LDH > 2× *N* and group C) [13, 77, 58]		
A	Completely resected stage I/II	10%
B, LDH ≤ 2× *N*	Non-resected stage I-III	40%
B HR, LDH >2× *N*	Stage III/IV	25%
C	Leukemic (25% blasts) and/or CNS disease	25%

assignment. Patients are divided into four risk groups (R1–R4) with approximately 50% of patients falling into the intermediate-risk R2 group [114].

The FAB-LMB protocols allocated patients to risk groups based on stage as well as resectability (Table 13.3). In the most recent international trial, LDH was incorporated in the definition of higher-risk patients [58].

Though the general principles are the same, the difference in the details of the risk group stratification between the two major cooperative groups (BFM and LMB-COG) makes it problematic to directly compare outcomes across the studies. Giulino-Roth and colleagues cite a good example of the problem with the following example: "a child with BL and 10% bone marrow histological involvement (Stage IV disease) with LDH >1000 would have been classified as Group B (intermediate risk) in the FAB96 trial and receive intermediate risk therapy while the same patient would be classified as R4 based on BFM criteria and receive the most aggressive therapy" [29].

Prognostic Factors

Age and Gender

Historically, it was thought that BL in adolescence was an independent risk factor for poorer event-free survival (EFS). In the LMB89 study, there was an increased risk of recurrence in children over 15 years with mature B-NHL, compared with children less than 15 years [78]. The BFM95 study also found that female sex and age over 10 years were associated with an increased risk of relapse in high-risk disease (R3/R4) [113]. However, the more recent FAB96 study demonstrated that with more intensive therapy, age over 15 years and gender were not associated with a worse prognosis [15].

Stage of Disease

While patients with advanced-stage disease due to bone marrow involvement alone did well in the FAB-96 study (90% survival), CNS involvement in patients with BL was associated with a particularly poor outcome. Combined BM-positive and CNS disease confers the worst prognosis [15]. Elevated LDH at diagnosis continues to remain an independent risk factor for inferior event-free survival [94] in patients treated with intermediate-risk protocols, but the effect is negated by intensification of therapy [13, 114].

Poor Response to Treatment

Within the context of LMB therapy, high-risk (group C) patients found to be poor responders to the initial COP prephase treatment (<20% reduction of measurable disease by day 6 of therapy) did very poorly with a 4-year EFS of 30% on the FAB96 trial [13]. Failure to achieve a complete remission (CR) after the consolidation phases also predicts a poor prognosis, and intensification of therapy (from Group B to group C and from Group C to autologous stem cell transplant) is recommended for patients with pathologically proven residual disease at this time point [78]. Whether negative PET FDG uptake in a residual mass shown on CT scan will negate the need for surgical excision of a residual mass remains to be proven.

Minimal Disseminated Disease and Minimal Residual Disease

The Italian group studied bone marrow minimal disseminated disease (MDD) at diagnosis among high-risk patients with morphologically negative bone marrows treated on a BFM protocol using a long-distance PCR assay that detects the t(8;14)(q24;q32). Patients who were MDD-positive at diagnosis (31% of patients) had an inferior outcome with a relative risk of relapse of 4.7, compared to those without MDD at diagnosis [63]. In contrast, MDD assessed by IgIGHV primer pools did not predict relapse in the Children's Oncology Group pilot trial of rituximab added to intensive therapy for children with group B disease [104].

Persistence of disease in the bone marrow, measured as minimal residual disease (MRD), was also found to be prognostic by Mussolin and colleagues. The 3-year relapse-free survival in patients with continued MRD positivity after cycle 1 was 38% vs. 84% in patients who were MRD-negative [62]. In contrast, in an evaluation of MRD in patients enrolled on the Children's Oncology Group Pilot trial that incorporated rituximab, MRD at the end of induction did not predict relapse [105]. It is possible that these conflicting results reflect the addition of rituximab to intensive therapy, differences in the sensitivity of the assays, and small patient and event numbers [29]. Further research is needed on this topic.

Cancer Predisposition Syndromes

Outcomes for patients with certain cancer predisposition syndromes associated with lymphoma (AT, Nijmegen breakage syndrome, and constitutional mismatch repair disease) were poor in one study with a 5-year EFS of 40% [6]. Approximately 50% of deaths were due to therapy-related toxicity. In contrast, patients with X-linked lymphoproliferative syndrome and Wiskott-Aldrich syndrome with mature B-cell NHL had a comparatively good outcome with 70–90% of the patients remaining in first complete remission [6].

Treatment for Mature B-Cell Lymphoma

History

Prior to the 1970s, children with NHL faced a dismal prognosis.

The initial experience in the use of chemotherapy in treating BL was described by Denis Burkitt, who treated patients in Africa with single-agent therapy, initially with cyclophosphamide given at a dose of 40 mg/kg, orally or intravenously [11]. After single-agent therapy, approximately 20% of patients achieved long-term remissions [60].

Over the next decade, researchers in Uganda, Kenya, and Nigeria investigated the use of available drugs including cyclophosphamide, methotrexate, nitrogen mustard, and melphalan [66, 69, 70]. Although dosing, routes of administration, and schedules differed within and between studies, there was clear evidence of tumor responsiveness.

In the late 1960s, the National Cancer Institute (NCI) and the Uganda Cancer Institute of Makerere University described the importance of using multiple agents that were non-cross-resistance [120].

Subsequent studies confirmed the favorable results with combination chemotherapy [72, 73, 119] and demonstrated the importance of intrathecal chemotherapy as CNS-directed treatment and prophylaxis [51, 67, 71].

The cyclophosphamide, vincristine, and methotrexate (COM) regimen, initially developed in Africa, was subsequently adopted by the NCI as a backbone for further therapy [117] and was the foundation for future studies in North America.

Separation from Leukemia Protocols

In the early 1980s, the Children's Cancer Study Group (CCG-551) performed a randomized trial designed to study the relative effectiveness of two therapy programs for the treatment of childhood and adolescent NHL. They compared a COMP regimen (cyclophosphamide, vincristine, methotrexate, and prednisone) to a ten-drug regimen (modified LSA2-L2) [4]. Patients were divided into lymphoblastic lymphoma versus non-lymphoblastic lymphoma. The four-drug regimen was more effective than the ten-drug program in those with advanced non-lymphoblastic disease (57% versus 28% 2-year disease-free survival) [3]. Shorter, more intensive therapy also improved survival in extensive B-NHL patients in BFM studies, when compared to a protocol designed for acute lymphoblastic leukemia [26].

Role of Surgery

Initial studies in Africa suggested that the extent of surgical resection impacted outcome, with a retrospective review demonstrating improved EFS >90%, with complete tumor resection, whereas partial resection resulted in similar outcomes to no resection at all [52]. To determine the appropriate role of surgical intervention in NHL primaries in the abdomen, the CCG performed a multivariate analysis on 84 patients with abdominal lymphoma and found that only the extent of disease, but not complete surgical resection, was an independent predictor of outcome [46]. The current guidelines suggest that a complete resection should be done if feasible as adjuvant chemotherapy can be minimized, but that attempts at incomplete resection simply delay the start of chemotherapy.

Role of Radiation

The role of radiotherapy in treating mature B-NHL had been evaluated in early trials in Africa where craniospinal radiation proved to have no benefit when administered prophylactically [71]. A randomized study conducted by the St. Jude research group demonstrated no benefit but increased toxicity when 30–35 Gy of involved field radiotherapy was added

to combination chemotherapy for patients with advanced stage III–IV NHL [61]. Additionally it was demonstrated that the addition of involved field radiation in children receiving cyclophosphamide, vincristine, prednisone, and doxorubicin followed by a 24-week maintenance had no impact on disease-free survival [48].

Radiotherapy for the treatment of CNS disease has been replaced by intensive systemic multi-agent therapy and IT chemotherapy. The FAB96 study found that by intensifying IT administration and incorporating an additional dose of systemic high-dose methotrexate, similar outcomes in patients with CNS disease could be achieved without the use of radiation [13], compared to earlier trials where radiation was incorporated [78]. This is not surprising since BL remains at least four times as common as DLBCL, and early trials in Africa showed that radiation therapy given in single fractions or even two fractions per day is ineffective against the very rapidly cycling BL cells [68, 88].

Current Treatment Approach

The substantial progress in improving the outcomes of children with mature B-NHL has evolved through sequential cooperative groups with international collaborations.

General therapeutic strategies have included matching intensity of the regimen with disease risk and the use of non-cross-resistant agents with fractionated administration schedules. These regimens are associated with substantial acute toxicities including infection and mucositis but with limited risks of long-term side effects of infertility and cardiotoxicity, as well as low treatment-related mortality in countries with good supportive care.

Therapy for Low-Risk Disease

Low-risk disease as defined by both the FAB and BFM as resected stage I or completely resected abdominal stage II disease. Both groups have adopted similar therapeutic approaches, and with contemporary treatment, the outcomes for these patients are excellent with minimal long-term sequelae. Over the years, therapy has gradually been decreased to two cycles of multi-agent chemotherapy for both cooperative groups: the main difference being that the BFM continues to administer IT chemotherapy to low-risk patients, while the FAB have excluded IT chemotherapy in this group. In the FAB LMB 96 study, low-risk patients had a 4-year EFS of 98.3% and OS of 99.2%, after two cycles of COPAD (fractioned cyclophosphamide, doxorubicin, vincristine, prednisone) [27]. The BFM95 study reported a 3-year EFS of 94% with two cycles of chemotherapy which included dexamethasone, ifosfamide, methotrexate, cytarabine, etoposide (cycle 1) and dexamethasone, cyclophosphamide, methotrexate, doxorubicin (cycle 2), and IT therapy [79, 114] (see Table 13.4).

Therapy for Intermediate-Risk B-NHL

The intermediate-risk group is the largest and most heterogenous and is defined by the FAB and BFM slightly differently. FAB group B includes all patients that neither meet criteria for group A (stage 1 or completed resected abdominal stage 2) or group C (any CNS involvement or >25% blast cells in the bone marrow). Group B encompasses approximately 70% of the children and adolescents with B-NHL [77].

Attempts have been made by both the FAB and BFM groups to decrease intensity while maintaining excellent outcomes. The FAB group successfully decreased therapy for good responders (defined as good response to COP and a CR after the first consolidation cycle) by reducing the dose of cyclophosphamide in COPADM2 by 50% and removing a maintenance cycle, thus establishing a new standard of care for this group consisting of COP reduction and four cycles of chemotherapy [77].

Importantly, a delay between the two induction courses in this intermediate group of patients was prognostic, with an interval longer than 21 days between COPADM1 and COPADM2 associated with an 8% lower EFS [79].

Contemporaneously with the LMB-FAB96 study, the BFM group decreased therapy from six to four cycles in BFM90 in those with non-resected stage I/II and stage III with LDH <500 IU/L [94] and decreased methotrexate dosing (from 5 gm/m^2 to 1 gm/m^2) and infusion time (from 24 h to 4 h) in BFM95 [114], maintaining excellent survival rates (94% vs 96%) with significantly decreased toxicity.

Therapy for High-Risk B-NHL

The highest-risk groups are defined differently by BFM and FAB. The BFM R3 group initially included all patients with LDH >500 U/L who have had either no resection or an incomplete resection of abdominal lymphoma, along with all patients with BM involvement and/or CNS disease, and/or multifocal bone involvement [94] (see Table 13.3). Based on previous results indicating an especially poor prognosis for patients with LDH ≥1000 IU/L, the NHL BFM95 study subdivided this group into an even higher-risk group, R4, which includes patients with stages III and IV disease and LDH ≥1000IU/L with/without CNS disease [113]. The updated FAB high-risk classification now includes a subset of group B patients with LDH greater than twice the normal value (LDH >2N) [58]. Group C patients who have >25% blast

Table 13.4 Treatment stratification and results in contemporary cooperative group trials of pediatric mature B-cell neoplasms

Group	Definition	*N*	EFS,% (year)	Chemotherapy courses (randomized study questions)	Comments
FAB-LMB 96 Trial [13, 77, 27]					
A	Stage I, resected; stage II, abdominal, resected	132	98 (4 years)	COPAD-COPAD	2 courses of therapy and no intrathecal treatment is curative in patients with resected localized disease
B	Stage I+II not resected; Stage III Stage IV with bone marrow blasts < 25%	657	90 (4 years)	COP-COPADM1-COPADM2-CYM-CYM-M1 (randomization for complete responders after COPADM1 to therapy reduction versus standard)	Therapy reductions of 50% dose of cyclophosphamide and omission of M1 was equally efficacious than the standard regimen
C	>25% BM blasts and/or CNS+ Nonresponder to COP of group B	235	79 (4 years)	COP-COPADM1-COPADM2-CYVE1-CYVE2-M1-M2-M3-M4 (randomization for complete responders after COPADM1 + 2)	Therapy reductions of reduced-intensity "mini-CYVE" and omission of M2, M3, M4 resulted in inferior outcome
International B-NHL 2010 [58]					
B, high LDH and C	Stage III with LDH level >2*N*, stage IV, and B-AL	310	94 (with Rituxan) vs 82% (without) (1 year)	Group B: COP-COPADM1-COPADM2-CYM-CYM-M1 ± rituximab × 6 courses Group C: COP-COPADM1-COPADM2-CYVE1-CYVE2-M1-M2 ± rituximab (with additional IT and MTX for CNS-positive patients	Study terminated early based on superiority of rituximab arm
BFM-NHL-95 [113]					
R1	Stages I + II, resected	48	94 (3 years)	A-B (randomization: MTX IV over 24 h vs. 4 h)	MTX over 4 h less toxic, equally efficacious
R2	Stages I + II, not resected; stage III, LDH < 500/U/L	233	94 (3 years)	p-A-B A-B (randomization: MTX IV over 24 h vs. 4 h)	MTX over 4 h less toxic, equally efficacious
R3	Stage III, LDH 500–999 U/L; Stage IV and LDH <1000U/L and CNS neg	82	85 (3 years)	p-AA-BB-CC-AA- BB (randomization: MTX in AA, BB IV over 24 h vs. 4 h)	MTX over 4 h less toxic BUT in this group less effective
R4	Stage III + IV and LDH ≥ 1000 U/L and/or CNS+	142	81 (3 years)	p-AA-BB-CC-AA-BB-CC (randomization: MTX in AA, BB IV over 24 h vs. 4 h)	MTX over 4 h less toxic BUT in this group less effective

cells in the bone marrow are considered C1, while C3 represents patients with the highest risk (i.e., leptomeningeal CNS involvement) [58].

Both the FAB and BFM groups realized the importance of intensifying therapy for high-risk patients while simultaneously trying to avoid excessive toxicity. In the BFM 90 study, patients with high-risk disease received two cycles of intensive therapy and then had assessment of response. Patients in remission received another four cycles of intensive therapy, while those with incomplete responses who achieved a CR had even further treatment intensification with four cycles of therapy, including high-dose ARAC or autologous transplant for those with persistent viable tumor [94]. Six-year EFS for this patient subgroup increased to 78% [94] and also improved to 65% for those with CNS involvement. The LMB group in their LMB81 study recognized the poor survival in CNS-positive patients [80] and succeeded in improving the 5-year EFS to 79% in this group in LMB89 by giving eight cycles of therapy which included high-dose methotrexate, high-dose ARAC, etoposide, and cranial irradiation [78].

Toxicity remained a significant problem on both treatment protocols, so attempts were made by both study groups to decrease either the dose of chemotherapy or the length of administration. The BFM 95 study tested a reduction of the high-dose methotrexate infusion time (5 g/m^2 over 24 h vs. 4 h) ([113]. An interim analysis revealed that treatment failure was five times higher in the shorter methotrexate infusion arm resulting in discontinuation of the randomization in the high-risk groups. The 3-year EFS for the 40 CNS-positive patients in this study was 69% [114].

Similarly, the FAB96 study randomized high-risk (Group C) patients to a reduced-intensity consolidation (33% reduction in ARAC dosing and 50% reduction of etoposide) and also removed three maintenance cycles [13]. Although toxicity was reduced, there was a 10% decrease in EFS; standard FAB/LMB therapy resulted in a 90% 4-year EFS, while reduced therapy resulted in an EFS of 80% [13]. Both study groups therefore demonstrated that reduced therapy was feasible and effective in the intermediate but not in the highest-risk group patients. Another important finding of the FAB-96

study was that a poor response to the COP prephase therapy was the poorest prognosticator, followed by leptomeningeal involvement. CNS disease defined by cranial nerve palsy or spinal cord compression did not imply the same poor outcome. This has led to the LMB group further intensifying the methotrexate dosing by increasing the infusion time of the 8 gm/m^2 MTX from 4 h to 24 h, a strategy that was adopted for CSF-positive patients in the international rituximab trial.

Rituximab in the Therapy of Pediatric Mature B-NHL

CD20 is expressed on 100% of BL and 98% of DLBCL in childhood [84]. Rituximab is a chimeric antibody directed at CD20 that is a component of standard therapy combined with CHOP for adults with DLBCL [86]. Evidence to inform the safety and efficacy of rituximab for the treatment of children with B-NHL has been established over the last decade. A BFM phase II study that administered rituximab as monotherapy in newly diagnosed pediatric B-NHL before chemotherapy was started, reported response rates of 41% [56]. A single-arm pilot COG study demonstrated the safety of addition of six doses of rituximab to LMB therapy for children with group B and C disease [33, 34].

Based on the results of this trial, the intergroup B-NHL Ritux 2010 trial was conducted. This was an international prospective phase III randomized trial of LMB chemotherapy with and without rituximab for patients with group B disease with high LDH and those with group C disease. The study was closed to accrual after approximately 50% of the planned patients were enrolled because of superiority of the rituximab arm. In this study, including only higher-risk patients, the event-free survival at 1 year was 94% in those patients randomized to receive rituximab versus 86% in the standard arm [58].

Ongoing studies evaluating the role of rituximab in children with mature B-NHL include the BFM and NOPHO trial in which higher-risk patients are randomized to one versus seven doses of rituximab, in addition to standard intensive BFM therapy. For lower-risk patients, the study addresses whether excellent outcomes can be preserved with rituximab replacing anthracycline. Similarly in the United States, REBOOT (reduced burden of oncologic therapy in advanced B-cell lymphoma) is a multicenter, nonrandomized trial investigating whether the addition of rituximab can maintain excellent outcomes with reduced anthracycline dosing in selected lower-risk patients [32].

Summary of BFM/FAB-LMB trials

Summarizing the results of the study group trials, it was found that in patients with localized and completely excised disease, chemotherapy could be significantly reduced while maintaining excellent outcomes. For intermediate-risk patients as defined by the BFM and FAB study groups, the length and intensity of chemotherapy could be safely reduced without compromising results. In the highest-risk groups, attempts at reduction of doses and length of conventional chemotherapy were not successful. Addition of rituximab to higher-risk patients has now been shown to be effective in improving the outcome in this group.

Ongoing trials described above are studying whether with the addition of rituximab, further reductions in therapy will be safe and feasible.

Surveillance Post Completion of Therapy

Of the patients treated with contemporary therapy, the majority of relapses will occur within 1 year of diagnosis [77, 78, 114]. Relapse tends to occur earlier in BL than DLBCL (5 months in BL vs 27 months in DLBCL) [39]. Surveillance imaging for disease recurrence is generally not recommended for children with mature B-NHL, given that the risk of recurrence is small [39]. Additionally, the use of PET/CT is not recommended based on lack of evidence of utility along with the risk of false-positive results and unnecessary radiation exposure [9].

Relapsed and Refractory Disease

Excellent outcomes have now been achieved with intensive, risk-adapted therapy. For the small percentage of patients with refractory or relapsed disease, the outcomes remain very poor. The 5-year OS for all patients with relapsed/refractory mature B-NHL is approximately 30% [39].

Ifosfamide, carboplatin, and etoposide (ICE) have been commonly employed for salvage therapy for pediatric refractory/relapsed lymphoma [14, 44]. A COG single-arm study evaluated the toxicity and efficacy of ICE combined with rituximab in children with recurrent or refractory mature B-NHL [35]. Of 20 evaluable patients, there were 12 responders and 6 proceeded to HSCT. Patients that did not respond to R-ICE had a very short survival [35]. It should be noted, however, that none of these patients would have received rituximab as frontline therapy. Patients with BL who relapse after the current rituximab-containing intensive protocols are likely to continue to have very poor salvage rates, and alternative strategies need to be sought.

Hematopoietic Stem Cell Transplant

For patients with chemotherapy responsive disease, evidence suggests that proceeding to HSCT is an essential component of curative therapy. Given that many of the earlier studies describing the role of transplant in patients with relapsed disease included patients treated with less intensive upfront

regimens, there needs to be caution when interpreting the findings described below. Most contemporary patients will have received more intensive primary regimens [23]. The SFOP reported on 27 patients with relapsed mature B-NHL treated as per LMB84. Twelve patients received conventional chemotherapy without HSCT and no patient survived. Overall survival for the 15 patients receiving HSCT was 27%. All survivors were in complete remission prior to transplant [87], emphasizing the importance of chemo-sensitive disease. The European Lymphoma Bone Marrow Transplantation Registry reported 5-year EFS of 40% for 89 pediatric patients with refractory/recurrent mature B-NHL who survived to get to autologous HSCT from 1979 to 1991. Again, no patient with chemoresistant disease survived [45].

The choice of autologous versus allogeneic HSCT remains controversial for relapsed mature B-NHL. The Center for International Blood and Marrow Transplant Research assessed their results retrospectively and found that 5-year EFS were similar after allogeneic and autologous HSCT for DLBCL (50% vs. 52%) and BL (31% vs. 27%). Patients who received autologous transplant died mostly of recurrent disease, while death from transplant-related complications was more common after allogeneic HSCT. This suggests some graft vs. lymphoma effect in those receiving allogeneic transplant, albeit with an increased risk of mortality [36].

In patients with NHL who have failed an autologous HSCT, a second allogeneic transplant has been attempted [100]. Sequential transplants have been examined in a small cohort of patients. In this setting patients undergo myeloablative regimens prior to autologous HSCT as a debulking therapy. This is then followed by reduced-intensity conditioning regimen (busulfan/cytarabine) and allogeneic HSCT, which can result in engraftment of donor cells to achieve a graft vs. lymphoma effect with less toxicity compared to an upfront myeloablative regimen and allogeneic HSCT [101]. In a prospective multicenter study in children with poor-risk refractory/recurrent HL and NHL, this approach appeared tolerable. The study included 14 patients with NHL; 8 had B-cell NHL. Three of these patients (37%) relapsed after the autologous HSCT; however the other five had long-term CR (1.9–8.8 years) [101].

Novel Therapies

There are numerous agents that show promise, most of which to date have not been studied in children with mature B-NHL. Categories of novel therapies include monoclonal antibodies, cellular therapy, and small-molecule inhibitors.

Monoclonal Antibodies

New generations of anti-CD20 monoclonal antibodies (mAbs) have been developed with increased antibody-dependent cellular cytotoxicity (ADCC) and increased Fc binding affinity for the FcγRIIIa receptor (CD16) on immune effector cells, resulting in enhanced tumor killing [95]. Obinutuzumab is a third-generation humanized type II antibody. A preclinical study comparing the in vitro and in vivo efficacy of obinutuzumab to the chimeric antibody rituximab, in rituximab-sensitive and rituximab-resistant BL, found significantly increased cell death with obinutuzumab [7]. However, in a randomized trial of adult patients with newly diagnosed DLBCL, obinutuzumab was not superior to rituximab when combined with CHOP chemotherapy [110].

Polatuzumab vedotin (PV) is an antibody-drug conjugate containing an anti-CD79B monoclonal antibody conjugated to monomethyl auristatin E, a microtubule-disrupting agent. It is well tolerated in adults with refractory NHL with 14 of 25 patients with DLBCL (56%) responding to the recommended phase II dose [76]. Preclinical studies have indicated that PV combined with obinutuzumab significantly enhances cell death in both rituximab-sensitive and rituximab-resistant CD79B/CD20 BL lines compared to either therapy alone, indicating a possible synergistic effect [107].

Blinatumomab is a bispecific single-chain antibody construct of the BiTE (bispecific T-cell engager) class. It binds with one arm to CD19 on both benign and malignant B cells, while the other arm binds CD3 present on T cells. This triggers the signaling cascade of the T-cell receptor complex and can redirect cytotoxic T cells to target tumor cells at very low concentrations of the BiTE antibody [64]. Blinatumomab has proven efficacy in both adult and pediatric patients with relapsed or refractory ALL [108, 111]. In a phase I study, 6 of 11 patients with refractory and/or relapsed DLBCL (55%) responded to blinatumomab monotherapy including 4 CRs (36%), with a median response duration of 404 days [31]. The phase II portion of this study resulted in responses in 43% of evaluable patients (9 of 21 patients were from the original phase I study) with a CR rate of 19% [109].

Cellular Therapy

Chimeric antigen receptor (CAR) T-cell therapy offers another cell-based therapy, which has proven highly successful in the treatment of children and young adults with refractory ALL [55] and is currently being investigated for its applicability to lymphomas. T cells expressing CARs directed at CD19 and CD20 offer potential benefits over rituximab including active trafficking to tumor sites, in vivo expansion, and long-term persistence [91]. In contrast to conventional T cells, which rely on their native TCRs for tumor antigen recognition, CAR-T cells recognize unprocessed antigen and therefore kill tumor cells independently of their expression of major histocompatibility complex (MHC) antigens; thus they can circumvent some of the major mechanisms by which tumors avoid MHC-restricted T-cell

recognition [91]. A recent clinical study demonstrated complete response rates of 43% (6/14 patients) with relapsed and/or refractory DLBCL in those that received CD19 CAR T cells, with sustained remissions lasting up to 3 years after a single dose [103]. A multicenter, phase II trial enrolled 111 adult patients with refractory B-cell lymphoma, including patients with DLBCL and PMBCL, and treated them with autologous anti-CD19 CARs with similarly promising results. Eighty-two percent of patients who were treated with the anti-CD19 CARs had an objective response and 54% had a complete response. The overall rate of survival at 18 months was 52% [65].

CAR T cells targeting alternative lymphoma-associated antigens are also being assessed in adult populations. Clinical studies of anti-CD20 CAR T cells have been conducted. Results of a phase II study of an anti-CD20 CAR in 11 patients with DLBCL or indolent NHL demonstrated a CR rate of 55%. Complete responses occurred in four of eight patients with DLBCL [116]. The immunoglobulin kappa (κ) light chain antigen also offers a novel target for CAR-T-cell therapy because complete B-cell aplasia could be avoided. In a phase I trial, seven patients with various NHL subtypes that expressed κ light chains, including DLBCL, were treated with anti-κ-light-chain CAR T cells. Overall, three of nine patients (33%) had an objective response; however these responses were in patients with either lymphoplasmacytic lymphoma or transformed follicular lymphoma and not in patients with DLBCL [92].

Immune Checkpoint Inhibitors

Immune checkpoint inhibitors have also been investigated in DLBCL, primarily in adults. By blocking the cytotoxic T lymphocyte-associated protein 4 (CTLA-4) and programmed death 1 (PD-1) pathways, which function as breaks on the immune system, immune checkpoint inhibitors enable restoration of T-cell effector function and enhanced tumor killing. Pidilizumab was the first humanized IgG1 monoclonal antibody directed against PD-1 used in adult patients with DLBCL, with at least partial disease response [5]. In a phase II study, pidilizumab given after autologous transplant was found to be well tolerated [5]. Pembrolizumab, an IgG4 antibody against PD-1, and ipilimumab, a fully humanized IgG1 monoclonal antibody targeting the CTLA-4 pathway, are currently being investigated in clinical trials in patients with B-NHL, either alone or in combination with rituximab (NCT02362997, NCT01729806).

An interesting combination therapy involves combining CAR T cells with immune checkpoint inhibitors. Following anti-CD19 CAR T-cell infusion, it has been noted that the CARs can acquire a more exhausted phenotype associated with increased expression of PD-1 [43]. In vitro mouse models have found that interruption of the PD-L1/PD-1 axis using anti-PD-1 antibodies can restore CAR-T-cell function and may be an effective way to improve the efficacy of CAR T-cell therapies [17]. In the clinical context, this was tested in one patient with refractory PMBCL that progressed through CAR T-cell infusion and had a subsequent response to pembrolizumab, a PD-1 blocking antibody [18]. Both the percentage of CAR T cells and IL6 increased after the initial pembrolizumab infusion and the patient had significant clinical improvement and was alive and well 1-year post initial infusion. Although pembrolizumab may have had activity independent of CART19 cells, these findings suggest the combination may induce more potent tumor killing. A phase I clinical trial is currently underway in which patients with refractory/relapsed lymphoma, including DLBCL, are treated with pembrolizumab following anti-CD19 CAR-T-cell infusion in an attempt to reactivate exhausted CAR T cells (NCT02650999). Another phase I/II study is evaluating the safety and efficacy of CD 19 CARs in combination with atezolizumab, a humanized monoclonal IgG1 antibody against PD-L1 (NCT02926833).

Molecular-Targeted Therapies

Novel therapies that target key cell-signaling pathways in B-cell development represent another area of potential targeted therapy.

Bruton's tyrosine kinase (BTK) is essential to the B-cell antigen receptor-signaling cascade. Ibrutinib is a small-molecule inhibitor of BTK that has demonstrated activity in relapsed/refractory DLBCL in adults [112]. When combined with R-CHOP, it was tolerable and efficacious in a phase Ib study of adult patients with DLBCL [115]. It should be noted however that higher responses are observed in patients with the ABC subtype of DLBCL, compared with the GCB subtype [112, 115], potentially decreasing the applicability of this agent in the pediatric population. This is because active BCR signaling appears to be more important for the ABC subtype of DLBCL than the GCB subtype [24]. Research indicates that BL appears to rely on tonic BCR signaling networks for lymphomagenesis; therefore targeting the PI3K/Akt/mTOR pathway may offer a more promising therapeutic target [102]. Pharmacologic inhibition of PI3K or treatment with the mTOR inhibitor rapamycin has been shown to kill BL cells, and thus, small-molecule inhibitors, including PI3K inhibitors, may be a useful targeted therapy for BL in the future [102].

Summary

Outcomes for children with mature B-NHL receiving optimal contemporary therapy are excellent with an overall sur-

vival over 90% for even those in the highest-risk groups. Short-term toxicity remains substantial for current intensive regimens and outcomes for those with relapsed disease remain poor. Highly effective therapy in resource-limited settings remains challenging. Better understanding of disease biology and genetics and the development and application of novel and targeted therapies will hopefully allow for the substitution of cytotoxic therapy by more specific and less toxic agents for all patients, as well as improvement of outcomes for those with refractory disease. Well-designed collaborative clinical trials will be critical in building on the successes made to date in the treatment of children with mature B-NHL.

References

1. A clinical evaluation of the International Lymphoma Study Group classification of non-Hodgkin's lymphoma. The Non-Hodgkin's Lymphoma Classification Project. *Blood*. 1997;89(11):3909–18.
2. Allday MJ. How does Epstein-Barr virus (EBV) complement the activation of Myc in the pathogenesis of Burkitt's lymphoma? *Semin Cancer Biol*. 2009;19(6):366–76.
3. Anderson JR, Jenkin RD, Wilson JF, Kjeldsberg CR, Sposto R, Chilcote RR, Coccia PF, Exelby PR, Siegel S, Meadows AT, et al. Long-term follow-up of patients treated with COMP or LSA2L2 therapy for childhood non-Hodgkin's lymphoma: a report of CCG-551 from the Childrens Cancer Group. *J Clin Oncol*. 1993;11(6):1024–32.
4. Anderson JR, Wilson JF, Jenkin DT, Meadows AT, Kersey J, Chilcote RR, Coccia P, Exelby P, Kushner J, Siegel S, Hammond D. Childhood non-Hodgkin's lymphoma. The results of a randomized therapeutic trial comparing a 4-drug regimen (COMP) with a 10-drug regimen (LSA2-L2). *N Engl J Med*. 1983;308(10):559–65.
5. Armand P, Nagler A, Weller EA, Devine SM, Avigan DE, Chen YB, Kaminski MS, Holland HK, Winter JN, Mason JR, Fay JW, Rizzieri DA, Hosing CM, Ball ED, Uberti JP, Lazarus HM, Mapara MY, Gregory SA, Timmerman JM, Andorsky D, Or R, Waller EK, Rotem-Yehudar R, Gordon LI. Disabling immune tolerance by programmed death-1 blockade with pidilizumab after autologous hematopoietic stem-cell transplantation for diffuse large B-cell lymphoma: results of an international phase II trial. *J Clin Oncol*. 2013;31(33):4199–206.
6. Attarbaschi A, Carraro E, Abla O, Barzilai-Birenboim S, Bomken S, Brugieres L, Bubanska E, Burkhardt B, Chiang AK, Csoka M, Fedorova A, Jazbec J, Kabickova E, Krenova Z, Lazic J, Loeffen J, Mann G, Niggli F, Miakova N, Osumi T, Ronceray L, Uyttebroeck A, Williams D, Woessmann W, Wrobel G, Pillon M. Non-Hodgkin lymphoma and pre-existing conditions: spectrum, clinical characteristics and outcome in 213 children and adolescents. *Haematologica*. 2016;101(12):1581–91.
7. Awasthi A, Ayello J, Van de Ven C, Elmacken M, Sabulski A, Barth MJ, Czuczman MS, Islam H, Klein C, Cairo MS. Obinutuzumab (GA101) compared to rituximab significantly enhances cell death and antibody-dependent cytotoxicity and improves overall survival against CD20+ rituximab-sensitive/-resistant Burkitt lymphoma (BL) and precursor B-acute lymphoblastic leukaemia (pre-B-ALL): potential targeted therapy in patients with poor risk CD20+BL and pre-B-ALL. *Br J Haematol*. 2015;171(5):763–75.
8. Ayers LW, Tumwine LK. Burkitt's lymphoma. New York: Springer; 2013.
9. Bhojwani D, McCarville MB, Choi JK, Sawyer J, Metzger ML, Inaba H, Davidoff AM, Gold R, Shulkin BL, Sandlund JT. The role of FDG-PET/CT in the evaluation of residual disease in paediatric non-Hodgkin lymphoma. *Br J Haematol*. 2015;168(6):845–53.
10. Burkhardt B, Zimmermann M, Oschlies I, Niggli F, Mann G, Parwaresch R, Riehm H, Schrappe M, Reiter A. The impact of age and gender on biology, clinical features and treatment outcome of non-Hodgkin lymphoma in childhood and adolescence. *Br J Haematol*. 2005;131(1):39–49.
11. Burkitt D. Long-term remissions following one and two-dose chemotherapy for African lymphoma. *Cancer*. 1967;20(5):756–9.
12. Burkitt DP. The discovery of Burkitt's lymphoma. *Cancer*. 1983;51(10):1777–86.
13. Cairo MS, Gerrard M, Sposto R, Auperin A, Pinkerton CR, Michon J, Weston C, Perkins SL, Raphael M, McCarthy K, Patte C, FAB LMB96 International Study Committee. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. *Blood*. 2007;109(7):2736–43.
14. Cairo MS, Shen V, Krailo MD, Bauer M, Miser JS, Sato JK, Blatt J, Blazar BR, Frierdich S, Liu-Mares W, Reaman GH. Prospective randomized trial between two doses of granulocyte colony-stimulating factor after ifosfamide, carboplatin, and etoposide in children with recurrent or refractory solid tumors: a children's cancer group report. *J Pediatr Hematol Oncol*. 2001;23(1):30–8.
15. Cairo MS, Sposto R, Gerrard M, Auperin A, Goldman SC, Harrison L, Pinkerton R, Raphael M, McCarthy K, Perkins SL, Patte C. Advanced stage, increased lactate dehydrogenase, and primary site, but not adolescent age (≥ 15 years), are associated with an increased risk of treatment failure in children and adolescents with mature B-cell non-Hodgkin's lymphoma: results of the FAB LMB 96 study. *J Clin Oncol*. 2012;30(4):387–93.
16. Campo E, Swerdlow SH, Harris NL, Pileri S, Stein H, Jaffe ES. The 2008 WHO classification of lymphoid neoplasms and beyond: evolving concepts and practical applications. *Blood*. 2011;117(19):5019–32.
17. Cherkassky L, Morello A, Villena-Vargas J, Feng Y, Dimitrov DS, Jones DR, Sadelain M, Adusumilli PS. Human CAR T cells with cell-intrinsic PD-1 checkpoint blockade resist tumor-mediated inhibition. *J Clin Invest*. 2016;126(8):3130–44.
18. Chong EA, Melenhorst JJ, Lacey SF, Ambrose DE, Gonzalez V, Levine BL, June CH, Schuster SJ. PD-1 blockade modulates chimeric antigen receptor (CAR)–modified T cells: refueling the CAR. *Blood*. 2017;129(8):1039–41.
19. Chu PG, Chang KL, Arber DA, Weiss LM. Practical applications of immunohistochemistry in hematolymphoid neoplasms. *Ann Diagn Pathol*. 1999;3(2):104–33.
20. Dalla-Favera R, Bregni M, Erikson J, Patterson D, Gallo RC, Croce CM. Human c-myc onc gene is located on the region of chromosome 8 that is translocated in Burkitt lymphoma cells. *Proc Natl Acad Sci U S A*. 1982;79(24):7824–7.
21. Dave SS, Fu K, Wright GW, Lam LT, Kluin P, Boerma EJ, Greiner TC, Weisenburger DD, Rosenwald A, Ott G, Muller-Hermelink HK, Gascoyne RD, Delabie J, Rimsza LM, Braziel RM, Grogan TM, Campo E, Jaffe ES, Dave BJ, Sanger W, Bast M, Vose JM, Armitage JO, Connors JM, Smeland EB, Kvaloy S, Holte H, Fisher RI, Miller TP, Montserrat E, Wilson WH, Bahl M, Zhao H, Yang L, Powell J, Simon R, Chan WC, Staudt LM. Molecular diagnosis of Burkitt's lymphoma. *N Engl J Med*. 2006;354(23):2431–42.
22. Deffenbacher KE, Iqbal J, Sanger W, Shen Y, Lachel C, Liu Z, Liu Y, Lim MS, Perkins SL, Fu K, Smith L, Lynch J, Staudt LM, Rimsza LM, Jaffe E, Rosenwald A, Ott GK, Delabie J, Campo E, Gascoyne RD, Cairo MS, Weisenburger DD, Greiner TC, Gross TG, Chan WC. Molecular distinctions between pediatric and

adult mature B-cell non-Hodgkin lymphomas identified through genomic profiling. *Blood*. 2012;119(16):3757–66.
23. El-Mallawany NK, Cairo MS. Advances in the diagnosis and treatment of childhood and adolescent B-cell non-Hodgkin lymphoma. *Clin Adv Hematol Oncol*. 2015;13(2):113–23.
24. Fowler N, Davis E. Targeting B-cell receptor signaling: changing the paradigm. *Hematology Am Soc Hematol Educ Program*. 2013;2013:553–60.
25. Frost M, Newell J, Lones MA, Tripp SR, Cairo MS, Perkins SL. Comparative immunohistochemical analysis of pediatric Burkitt lymphoma and diffuse large B-cell lymphoma. *Am J Clin Pathol*. 2004;121(3):384–92.
26. Gadner H, Muller-Weihrich S, Riehm H. Treatment strategies in malignant non-Hodgkin lymphomas in childhood. *Onkologie*. 1986;9(2):126–30.
27. Gerrard M, Cairo MS, Weston C, Auperin A, Pinkerton R, Lambilliote A, Sposto R, McCarthy K, Lacombe MJ, Perkins SL, Patte C. Excellent survival following two courses of COPAD chemotherapy in children and adolescents with resected localized B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study. *Br J Haematol*. 2008;141(6):840–7.
28. Gibson TM, Engels EA, Clarke CA, Lynch CF, Weisenburger DD, Morton LM. Risk of diffuse large B-cell lymphoma after solid organ transplantation in the United States. *Am J Hematol*. 2014;89(7):714–20.
29. Giulino-Roth L, Goldman S. Recent molecular and therapeutic advances in B-cell non-Hodgkin lymphoma in children. *Br J Haematol*. 2016;173(4):531–44.
30. Giulino-Roth L, Wang K, MacDonald TY, Mathew S, Tam Y, Cronin MT, Palmer G, Lucena-Silva N, Pedrosa F, Pedrosa M, Teruya-Feldstein J, Bhagat G, Alobeid B, Leoncini L, Bellan C, Rogena E, Pinkney KA, Rubin MA, Ribeiro RC, Yelensky R, Tam W, Stephens PJ, Cesarman E. Targeted genomic sequencing of pediatric Burkitt lymphoma identifies recurrent alterations in antiapoptotic and chromatin-remodeling genes. *Blood*. 2012;120(26):5181–4.
31. Goebeler M-E, Knop S, Viardot A, Kufer P, Topp MS, Einsele H, Noppeney R, Hess G, Kallert S, Mackensen A, Rupertus K, Kanz L, Libicher M, Nagorsen D, Zugmaier G, Klinger M, Wolf A, Dorsch B, Quednau BD, Schmidt M, Scheele J, Baeuerle PA, Leo E, Bargou RC. Bispecific T-cell engager (BiTE) antibody construct blinatumomab for the treatment of patients with relapsed/refractory non-Hodgkin lymphoma: final results from a phase I study. *J Clin Oncol*. 2016;34(10):1104–11.
32. Goldman S, Barth MJ, Oesterheld JE, Heym K, Harrison L, Nickerson B, El-Mallawany N, Hochberg J, Cairo MS. Preliminary results of a reduced burden of therapy trial by incorporation of rituximab and intrathecal liposomal cytarabine in children, adolescents and young adults with intermediate (FAB Group B) and high risk (FAB Group C) mature B-cell lymphoma/leukemia. *J Clin Oncol*. 2016;34(15_suppl):10534.
33. Goldman S, Smith L, Anderson JR, Perkins S, Harrison L, Geyer MB, Gross TG, Weinstein H, Bergeron S, Shiramizu B, Sanger W, Barth M, Zhi J, Cairo MS. Rituximab and FAB/LMB 96 chemotherapy in children with Stage III/IV B-cell non-Hodgkin lymphoma: a Children's Oncology Group report. *Leukemia*. 2013;27(5):1174–7.
34. Goldman S, Smith L, Galardy P, Perkins SL, Frazer JK, Sanger W, Anderson JR, Gross TG, Weinstein H, Harrison L, Shiramizu B, Barth M, Cairo MS. Rituximab with chemotherapy in children and adolescents with central nervous system and/or bone marrow-positive Burkitt lymphoma/leukaemia: a Children's Oncology Group Report. *Br J Haematol*. 2014;167(3):394–401.
35. Griffin TC, Weitzman S, Weinstein H, Chang M, Cairo M, Hutchison R, Shiramizu B, Wiley J, Woods D, Barnich M, Gross TG. A study of rituximab and ifosfamide, carboplatin, and etoposide chemotherapy in children with recurrent/refractory B-cell (CD20+) non-Hodgkin lymphoma and mature B-cell acute lymphoblastic leukemia: a report from the Children's Oncology Group. *Pediatr Blood Cancer*. 2009;52(2):177–81.
36. Gross TG, Hale GA, He W, Camitta BM, Sanders JE, Cairo MS, Hayashi RJ, Termuhlen AM, Zhang MJ, Davies SM, Eapen M. Hematopoietic stem cell transplantation for refractory or recurrent non-Hodgkin lymphoma in children and adolescents. *Biol Blood Marrow Transplant*. 2010;16(2):223–30.
37. Harris NL, Jaffe ES, Stein H, Banks PM, Chan JK, Cleary ML, Delsol G, De Wolf-Peeters C, Falini B, Gatter KC, et al. A revised European-American classification of lymphoid neoplasms: a proposal from the International Lymphoma Study Group. *Blood*. 1994;84(5):1361–92.
38. Hummel M, Bentink S, Berger H, Klapper W, Wessendorf S, Barth TF, Bernd HW, Cogliatti SB, Dierlamm J, Feller AC, Hansmann ML, Haralambieva E, Harder L, Hasenclever D, Kuhn M, Lenze D, Lichter P, Martin-Subero JI, Moller P, Muller-Hermelink HK, Ott G, Parwaresch RM, Pott C, Rosenwald A, Rosolowski M, Schwaenen C, Sturzenhofecker B, Szczepanowski M, Trautmann H, Wacker HH, Spang R, Loeffler M, Trumper L, Stein H, Siebert R. A biologic definition of Burkitt's lymphoma from transcriptional and genomic profiling. *N Engl J Med*. 2006;354(23):2419–30.
39. Jourdain A, Auperin A, Minard-Colin V, Aladjidi N, Zsiros J, Coze C, Gandemer V, Bertrand Y, Leverger G, Bergeron C, Michon J, Patte C. Outcome of and prognostic factors for relapse in children and adolescents with mature B-cell lymphoma and leukemia treated in three consecutive prospective "Lymphomes Malins B" protocols. A Societe Francaise des Cancers de l'Enfant study. *Haematologica*. 2015;100(6):810–7.
40. Kersey JH, Shapiro RS, Filipovich AH. Relationship of immunodeficiency to lymphoid malignancy. *Pediatr Infect Dis J*. 1988;7(5 Suppl):S10–2.
41. Klapper W, Kreuz M, Kohler CW, Burkhardt B, Szczepanowski M, Salaverria I, Hummel M, Loeffler M, Pellissery S, Woessmann W, Schwanen C, Trumper L, Wessendorf S, Spang R, Hasenclever D, Siebert R. Patient age at diagnosis is associated with the molecular characteristics of diffuse large B-cell lymphoma. *Blood*. 2012;119(8):1882–7.
42. Klapper W, Szczepanowski M, Burkhardt B, Berger H, Rosolowski M, Bentink S, Schwaenen C, Wessendorf S, Spang R, Moller P, Hansmann ML, Bernd HW, Ott G, Hummel M, Stein H, Loeffler M, Trumper L, Zimmermann M, Reiter A, Siebert R. Molecular profiling of pediatric mature B-cell lymphoma treated in population-based prospective clinical trials. *Blood*. 2008;112(4):1374–81.
43. Kochenderfer JN, Dudley ME, Kassim SH, Somerville RP, Carpenter RO, Stetler-Stevenson M, Yang JC, Phan GQ, Hughes MS, Sherry RM, Raffeld M, Feldman S, Lu L, Li YF, Ngo LT, Goy A, Feldman T, Spaner DE, Wang ML, Chen CC, Kranick SM, Nath A, Nathan DA, Morton KE, Toomey MA, Rosenberg SA. Chemotherapy-refractory diffuse large B-cell lymphoma and indolent B-cell malignancies can be effectively treated with autologous T cells expressing an anti-CD19 chimeric antigen receptor. *J Clin Oncol*. 2015;33(6):540–9.
44. Kung FH, Harris MB, Krischer JP. Ifosfamide/carboplatin/etoposide (ICE), an effective salvaging therapy for recurrent malignant non-Hodgkin lymphoma of childhood: a Pediatric Oncology Group phase II study. *Med Pediatr Oncol*. 1999;32(3):225–6.
45. Ladenstein R, Pearce R, Hartmann O, Patte C, Goldstone T, Philip T. High-dose chemotherapy with autologous bone marrow rescue in children with poor-risk Burkitt's lymphoma: a report from the European Lymphoma Bone Marrow Transplantation Registry. *Blood*. 1997;90(8):2921–30.
46. LaQuaglia MP, Stolar CJ, Krailo M, Exelby P, Siegel S, Meadows A, Hammond D. The role of surgery in abdominal non-Hodgkin's

lymphoma: experience from the Childrens Cancer Study Group. *J Pediatr Surg*. 1992;27(2):230–5.
47. Lenze D, Leoncini L, Hummel M, Volinia S, Liu CG, Amato T, De Falco G, Githanga J, Horn H, Nyagol J, Ott G, Palatini J, Pfreundschuh M, Rogena E, Rosenwald A, Siebert R, Croce CM, Stein H. The different epidemiologic subtypes of Burkitt lymphoma share a homogenous micro RNA profile distinct from diffuse large B-cell lymphoma. *Leukemia*. 2011;25(12):1869–76.
48. Link MP, Donaldson SS, Berard CW, Shuster JJ, Murphy SB. Results of treatment of childhood localized non-Hodgkin's lymphoma with combination chemotherapy with or without radiotherapy. *N Engl J Med*. 1990;322(17):1169–74.
49. Love C, Sun Z, Jima D, Li G, Zhang J, Miles R, Richards KL, Dunphy CH, Choi WW, Srivastava G, Lugar PL, Rizzieri DA, Lagoo AS, Bernal-Mizrachi L, Mann KP, Flowers CR, Naresh KN, Evens AM, Chadburn A, Gordon LI, Czader MB, Gill JI, Hsi ED, Greenough A, Moffitt AB, McKinney M, Banerjee A, Grubor V, Levy S, Dunson DB, Dave SS. The genetic landscape of mutations in Burkitt lymphoma. *Nat Genet*. 2012;44(12):1321–5.
50. Magrath I. Epidemiology: clues to the pathogenesis of Burkitt lymphoma. *Br J Haematol*. 2012a;156(6):744–56.
51. Magrath I. Towards curative therapy in Burkitt lymphoma: the role of early African studies in demonstrating the value of combination therapy and CNS prophylaxis. *Adv Hematol*. 2012b;2012:130680.
52. Magrath IT, Lwanga S, Carswell W, Harrison N. Surgical reduction of tumour bulk in management of abdominal Burkitt's lymphoma. *Br Med J*. 1974;2(5914):308–12.
53. Mann RB, Jaffe ES, Braylan RC, Nanba K, Frank MM, Ziegler JL, Berard CW. Non-endemic Burkitts's lymphoma. A B-cell tumor related to germinal centers. *N Engl J Med*. 1976;295(13):685–91.
54. Masque-Soler N, Szczepanowski M, Kohler CW, Aukema SM, Nagel I, Richter J, Siebert R, Spang R, Burkhardt B, Klapper W. Clinical and pathological features of Burkitt lymphoma showing expression of BCL2--an analysis including gene expression in formalin-fixed paraffin-embedded tissue. *Br J Haematol*. 2015;171(4):501–8.
55. Maude SL, Laetsch TW, Buechner J, Rives S, Boyer M, Bittencourt H, Bader P, Verneris MR, Stefanski HE, Myers GD, Qayed M, De Moerloose B, Hiramatsu H, Schlis K, Davis KL, Martin PL, Nemecek ER, Yanik GA, Peters C, Baruchel A, Boissel N, Mechinaud F, Balduzzi A, Krueger J, June CH, Levine BL, Wood P, Taran T, Leung M, Mueller KT, Zhang Y, Sen K, Lebwohl D, Pulsipher MA, Grupp SA. Tisagenlecleucel in children and young adults with B-cell lymphoblastic leukemia. *N Engl J Med*. 2018;378(5):439–48.
56. Meinhardt A, Burkhardt B, Zimmermann M, Borkhardt A, Kontny U, Klingebiel T, Berthold F, Janka-Schaub G, Klein C, Kabickova E, Klapper W, Attarbaschi A, Schrappe M, Reiter A. Phase II window study on rituximab in newly diagnosed pediatric mature B-cell non-Hodgkin's lymphoma and Burkitt leukemia. *J Clin Oncol*. 2010;28(19):3115–21.
57. Miles RR, Raphael M, McCarthy K, Wotherspoon A, Lones MA, Terrier-Lacombe MJ, Patte C, Gerrard M, Auperin A, Sposto R, Davenport V, Cairo MS, Perkins SL. Pediatric diffuse large B-cell lymphoma demonstrates a high proliferation index, frequent c-Myc protein expression, and a high incidence of germinal center subtype: report of the French-American-British (FAB) international study group. *Pediatr Blood Cancer*. 2008;51(3):369–74.
58. Minard-Colin V, Auperin A, Pillon M, Burke A, Anderson JR, Barkauskas DA, Wheatley K, Delgado R, Alexander S, Uyttebroeck A, Bollard C, Zsiros J, Csoka M, Goma G, Tulard A, Patte C, Gross TG. Results of the randomized Intergroup trial Inter-B-NHL Ritux 2010 for children and adolescents with high-risk B-cell non-Hodgkin lymphoma (B-NHL) and mature acute leukemia (B-AL): evaluation of rituximab (R) efficacy in addition to standard LMB chemotherapy (CT) regimen. *J Clin Oncol*. 2016;34(15_suppl):10507.
59. Molyneux EM, Rochford R, Griffin B, Newton R, Jackson G, Menon G, Harrison CJ, Israels T, Bailey S. Burkitt's lymphoma. *Lancet*. 2012;379(9822):1234–44.
60. Morrow RH, Pike MC, Kisuule A. Survival of Burkitt's lymphoma patients in Mulago Hospital, Uganda. *Br Med J*. 1967;4(5575):323–7.
61. Murphy SB, Hustu HO. A randomized trial of combined modality therapy of childhood non-Hodgkin's lymphoma. *Cancer*. 1980;45(4):630–7.
62. Mussolin L, Pillon M, Conter V, Piglione M, Lo Nigro L, Pierani P, Micalizzi C, Buffardi S, Basso G, Zanesco L, Rosolen A. Prognostic role of minimal residual disease in mature B-cell acute lymphoblastic leukemia of childhood. *J Clin Oncol*. 2007;25(33):5254–61.
63. Mussolin L, Pillon M, d'Amore ES, Conter V, Piglione M, Lo Nigro L, Garaventa A, Buffardi S, Arico M, Rosolen A. Minimal disseminated disease in high-risk Burkitt's lymphoma identifies patients with different prognosis. *J Clin Oncol*. 2011;29(13):1779–84.
64. Nagorsen D, Bargou R, Ruttinger D, Kufer P, Baeuerle PA, Zugmaier G. Immunotherapy of lymphoma and leukemia with T-cell engaging BiTE antibody blinatumomab. *Leuk Lymphoma*. 2009;50(6):886–91.
65. Neelapu SS, Locke FL, Bartlett NL, Lekakis LJ, Miklos DB, Jacobson CA, Braunschweig I, Oluwole OO, Siddiqi T, Lin Y, Timmerman JM, Stiff PJ, Friedberg JW, Flinn IW, Goy A, Hill BT, Smith MR, Deol A, Farooq U, McSweeney P, Munoz J, Avivi I, Castro JE, Westin JR, Chavez JC, Ghobadi A, Komanduri KV, Levy R, Jacobsen ED, Witzig TE, Reagan P, Bot A, Rossi J, Navale L, Jiang Y, Aycock J, Elias M, Chang D, Wiezorek J, Go WY. Axicabtagene ciloleucel CAR T-cell therapy in refractory large B-cell lymphoma. *N Engl J Med*. 2017;377(26):2531–44.
66. Ngu VA. Chemotherapy of Burkitt's tumor at the University of Ibadan, Nigeria. *JAMA*. 1972;222(9):1166.
67. Nkrumah FK, Perkins IV, Biggar RJ. Combination chemotherapy in abdominal Burkitt's lymphoma. *Cancer*. 1977;40(4):1410–6.
68. Norin T, Onyango J. Radiotherapy in Burkitt's lymphoma conventional or superfractionated regime—early results. *Int J Radiat Oncol Biol Phys*. 1977;2(5):399–406.
69. Oettgen HF, Burkitt D, Burchenal JH. Malignant lymphoma involving the jaw in African children: treatment with Methotrexate. *Cancer*. 1963a;16:616–23.
70. Oettgen HF, Clifford P, Burkitt D. Malignant lymphoma involving the jaw in African children: treatment with alkylating agents and actinomycin D. *Cancer Chemother Rep*. 1963b;28:25–34.
71. Olweny CL, Atine I, Kaddu-Mukasa A, Katongole-Mbidde E, Lwanga SK, Johansson B, Onyango J, Host H, Norin T, Willey B. Cerebrospinal irradiation of Burkitt's lymphoma. Failure in preventing central nervous system relapse. *Acta Radiol Ther Phys Biol*. 1977;16(3):225–31.
72. Olweny CL, Katongole-Mbidde E, Kaddu-Mukasa A, Atine I, Owor R, Lwanga S, Carswell W, Magrath IT. Treatment of Burkitt's lymphoma: randomized clinical trial of single-agent versus combination chemotherapy. *Int J Cancer*. 1976;17(4):436–40.
73. Olweny CL, Katongole-Mbidde E, Otim D, Lwanga SK, Magrath IT, Ziegler JL. Long-term experience with Burkitt's lymphoma in Uganda. *Int J Cancer*. 1980;26(3):261–6.
74. Onciu M, Schlette E, Zhou Y, Raimondi SC, Giles FJ, Kantarjian HM, Medeiros LJ, Ribeiro RC, Pui CH, Sandlund JT. Secondary chromosomal abnormalities predict outcome in pediatric and adult high-stage Burkitt lymphoma. *Cancer*. 2006;107(5):1084–92.
75. Oschlies I, Klapper W, Zimmermann M, Krams M, Wacker HH, Burkhardt B, Harder L, Siebert R, Reiter A, Parwaresch R. Diffuse large B-cell lymphoma in pediatric patients belongs predominantly to the germinal-center type B-cell lymphomas: a clinicopathologic

analysis of cases included in the German BFM (Berlin-Frankfurt-Munster) Multicenter Trial. *Blood*. 2006;107(10):4047–52.
76. Palanca-Wessels MCA, Czuczman M, Salles G, Assouline S, Sehn LH, Flinn I, Patel MR, Sangha R, Hagenbeek A, Advani R, Tilly H, Casasnovas O, Press OW, Yalamanchili S, Kahn R, Dere RC, Lu D, Jones S, Jones C, Chu Y-W, Morschhauser F. Safety and activity of the anti-CD79B antibody–drug conjugate polatuzumab vedotin in relapsed or refractory B-cell non-Hodgkin lymphoma and chronic lymphocytic leukaemia: a phase 1 study. *Lancet Oncol*. 2015;16(6):704–15.
77. Patte C, Auperin A, Gerrard M, Michon J, Pinkerton R, Sposto R, Weston C, Raphael M, Perkins SL, McCarthy K, Cairo MS. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. *Blood*. 2007;109(7):2773–80.
78. Patte C, Auperin A, Michon J, Behrendt H, Leverger G, Frappaz D, Lutz P, Coze C, Perel Y, Raphael M, Terrier-Lacombe MJ. The Societe Francaise d'Oncologie Pediatrique LMB89 protocol: highly effective multiagent chemotherapy tailored to the tumor burden and initial response in 561 unselected children with B-cell lymphomas and L3 leukemia. *Blood*. 2001;97(11):3370–9.
79. Patte C, Gerrard M., Auperin A, et al. (2003). Early treatment intensity has a major prognostic impact in the "intermediate risk" childhood and adolescent B-cell lymphoma: results of the international FAB LMB 96 trial. *Abstract of the American Society of Hematology*.
80. Patte C, Philip T, Rodary C, Bernard A, Zucker JM, Bernard JL, Robert A, Rialland X, Benz-Lemoine E, Demeocq F, et al. Improved survival rate in children with stage III and IV B cell non-Hodgkin's lymphoma and leukemia using multi-agent chemotherapy: results of a study of 114 children from the French Pediatric Oncology Society. *J Clin Oncol*. 1986;4(8):1219–26.
81. Pelicci PG, Knowles DM 2nd, Magrath I, Dalla-Favera R. Chromosomal breakpoints and structural alterations of the c-myc locus differ in endemic and sporadic forms of Burkitt lymphoma. *Proc Natl Acad Sci U S A*. 1986;83(9):2984–8.
82. Percy, C. L., M. A. Smith, M. Linet, L. A. Gloeckler Ries and D. L. Friedman (1999). Lymphomas and reticuloendothelial neoplasms. *Cancer incidence and survival among children and adolescents: United States SEER Program 1975-1995, National Cancer Institute, SEER Program* L Ries, MA Smith, JG Gurney et al. National Cancer Institute, SEER Program. NIH Pub. No. 99-4649. Bethesda, MD, 35–50.
83. Perkins SL. Work-up and diagnosis of pediatric non-Hodgkin's lymphomas. *Pediatr Dev Pathol*. 2000;3(4):374–90.
84. Perkins SL, Lones MA, Davenport V, Cairo MS. B-Cell non-Hodgkin's lymphoma in children and adolescents: surface antigen expression and clinical implications for future targeted bioimmune therapy: a children's cancer group report. *Clin Adv Hematol Oncol*. 2003;1(5):314–7.
85. Perkins SL, Morris SW. Biology and pathology of pediatric non-Hodgkin lymphoma *pediatric lymphomas*. H. J. H. Weinstein, M. M.; Link, M. P. Berlin Heidelberg: Springer; 2007.
86. Pfreundschuh M, Trumper L, Osterborg A, Pettengell R, Trneny M, Imrie K, Ma D, Gill D, Walewski J, Zinzani PL, Stahel R, Kvaloy S, Shpilberg O, Jaeger U, Hansen M, Lehtinen T, Lopez-Guillermo A, Corrado C, Scheliga A, Milpied N, Mendila M, Rashford M, Kuhnt E, Loeffler M. CHOP-like chemotherapy plus rituximab versus CHOP-like chemotherapy alone in young patients with good-prognosis diffuse large-B-cell lymphoma: a randomised controlled trial by the MabThera International Trial (MInT) Group. *Lancet Oncol*. 2006;7(5):379–91.
87. Philip T, Hartmann O, Pinkerton R, Zucker JM, Gentet JC, Lamagnere JP, Berhendt H, Perel Y, Otten J, Lutz P, et al. Curability of relapsed childhood B-cell non-Hodgkin's lymphoma after intensive first line therapy: a report from the Societe Francaise d'Oncologie Pediatrique. *Blood*. 1993;**81**(8):2003–6.
88. Pilecki B, Kopiec P, Zajusz A, Podworski H, Szelc S, Skladowski K, Maciejewski B. Hyperfractionated radiotherapy for Burkitt-type lymphoma. Radiobiological aspects. *Neoplasma*. 1991;38(6):609–15.
89. Poirel HA, Cairo MS, Heerema NA, Swansbury J, Auperin A, Launay E, Sanger WG, Talley P, Perkins SL, Raphael M, McCarthy K, Sposto R, Gerrard M, Bernheim A, Patte C. Specific cytogenetic abnormalities are associated with a significantly inferior outcome in children and adolescents with mature B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study. *Leukemia*. 2009;23(2):323–31.
90. Rainey JJ, Mwanda WO, Wairiumu P, Moormann AM, Wilson ML, Rochford R. Spatial distribution of Burkitt's lymphoma in Kenya and association with malaria risk. *Trop Med Int Health*. 2007;12(8):936–43.
91. Ramos CA, Heslop HE, Brenner MK. CAR-T cell therapy for lymphoma. *Annu Rev Med*. 2016a;67:165–83.
92. Ramos CA, Savoldo B, Torrano V, Ballard B, Zhang H, Dakhova O, Liu E, Carrum G, Kamble RT, Gee AP, Mei Z, Wu MF, Liu H, Grilley B, Rooney CM, Brenner MK, Heslop HE, Dotti G. Clinical responses with T lymphocytes targeting malignancy-associated kappa light chains. *J Clin Invest*. 2016b;126(7):2588–96.
93. Reiter A, Klapper W. Recent advances in the understanding and management of diffuse large B-cell lymphoma in children. *Br J Haematol*. 2008;142(3):329–47.
94. Reiter A, Schrappe M, Tiemann M, Ludwig WD, Yakisan E, Zimmermann M, Mann G, Chott A, Ebell W, Klingebiel T, Graf N, Kremens B, Muller-Weihrich S, Pluss HJ, Zintl F, Henze G, Riehm H. Improved treatment results in childhood B-cell neoplasms with tailored intensification of therapy: a report of the Berlin-Frankfurt-Munster Group Trial NHL-BFM 90. *Blood*. 1999;94(10):3294–306.
95. Robak T, Robak E. New anti-CD20 monoclonal antibodies for the treatment of B-cell lymphoid malignancies. *BioDrugs*. 2011;25(1):13–25.
96. Rosenwald A, Wright G, Chan WC, Connors JM, Campo E, Fisher RI, Gascoyne RD, Muller-Hermelink HK, Smeland EB, Giltnane JM, Hurt EM, Zhao H, Averett L, Yang L, Wilson WH, Jaffe ES, Simon R, Klausner RD, Powell J, Duffey PL, Longo DL, Greiner TC, Weisenburger DD, Sanger WG, Dave BJ, Lynch JC, Vose J, Armitage JO, Montserrat E, Lopez-Guillermo A, Grogan TM, Miller TP, LeBlanc M, Ott G, Kvaloy S, Delabie J, Holte H, Krajci P, Stokke T, Staudt LM. The use of molecular profiling to predict survival after chemotherapy for diffuse large-B-cell lymphoma. *N Engl J Med*. 2002;346(25):1937–47.
97. Rosolen A, Perkins SL, Pinkerton CR, Guillerman RP, Sandlund JT, Patte C, Reiter A, Cairo MS. Revised international pediatric non-Hodgkin lymphoma staging system. *J Clin Oncol*. 2015;33(18):2112–8.
98. Salavoura K, Kolialexi A, Tsangaris G, Mavrou A. Development of cancer in patients with primary immunodeficiencies. *Anticancer Res*. 2008;28(2b):1263–9.
99. Salzburg J, Burkhardt B, Zimmermann M, Wachowski O, Woessmann W, Oschlies I, Klapper W, Wacker HH, Ludwig WD, Niggli F, Mann G, Gadner H, Riehm H, Schrappe M, Reiter A. Prevalence, clinical pattern, and outcome of CNS involvement in childhood and adolescent non-Hodgkin's lymphoma differ by non-Hodgkin's lymphoma subtype: a Berlin-Frankfurt-Munster Group Report. *J Clin Oncol*. 2007;25(25):3915–22.
100. Sandlund JT, Bowman L, Heslop HE, Krance R, Mahmoud H, Pui C, Hale G, Benaim E. Intensive chemotherapy with hematopoietic stem-cell support for children with recurrent or refractory NHL. *Cytotherapy*. 2002;4(3):253–8.

101. Satwani P, Jin Z, Martin PL, Bhatia M, Garvin JH, George D, Chaudhury S, Talano J, Morris E, Harrison L, Sosna J, Peterson M, Militano O, Foley S, Kurtzberg J, Cairo MS. Sequential myeloablative autologous stem cell transplantation and reduced intensity allogeneic hematopoietic cell transplantation is safe and feasible in children, adolescents and young adults with poor-risk refractory or recurrent Hodgkin and non-Hodgkin lymphoma. *Leukemia*. 2015;29(2):448–55.
102. Schmitz R, Young RM, Ceribelli M, Jhavar S, Xiao W, Zhang M, Wright G, Shaffer AL, Hodson DJ, Buras E, Liu X, Powell J, Yang Y, Xu W, Zhao H, Kohlhammer H, Rosenwald A, Kluin P, Muller-Hermelink HK, Ott G, Gascoyne RD, Connors JM, Rimsza LM, Campo E, Jaffe ES, Delabie J, Smeland EB, Ogwang MD, Reynolds SJ, Fisher RI, Braziel RM, Tubbs RR, Cook JR, Weisenburger DD, Chan WC, Pittaluga S, Wilson W, Waldmann TA, Rowe M, Mbulaiteye SM, Rickinson AB, Staudt LM. Burkitt lymphoma pathogenesis and therapeutic targets from structural and functional genomics. *Nature*. 2012;490(7418):116–20.
103. Schuster SJ, Svoboda J, Chong EA, Nasta SD, Mato AR, Anak O, Brogdon JL, Pruteanu-Malinici I, Bhoj V, Landsburg D, Wasik M, Levine BL, Lacey SF, Melenhorst JJ, Porter DL, June CH. Chimeric antigen receptor T cells in refractory B-cell lymphomas. *N Engl J Med*. 2017;377(26):2545–54.
104. Shiramizu B, Goldman S, Kusao I, Agsalda M, Lynch J, Smith L, Harrison L, Morris E, Gross TG, Sanger W, Perkins S, Cairo MS. Minimal disease assessment in the treatment of children and adolescents with intermediate-risk (Stage III/IV) B-cell non-Hodgkin lymphoma: a children's oncology group report. *Br J Haematol*. 2011;153(6):758–63.
105. Shiramizu B, Goldman S, Smith L, Agsalda-Garcia M, Galardy P, Perkins SL, Frazer JK, Sanger W, Anderson JR, Gross TG, Weinstein H, Harrison L, Barth MJ, Mussolin L, Cairo MS. Impact of persistent minimal residual disease post-consolidation therapy in children and adolescents with advanced Burkitt leukaemia: a Children's Oncology Group Pilot Study Report. *Br J Haematol*. 2015;170(3):367–71.
106. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, Vardiman JW, editors. WHO classification of tumours of haematopoietic and lymphoid tissues. Lyon: WHO; 2008.
107. Tiwari A, Edani D, Ayello J, Klein C, Lee DA, Cairo MS. Polatuzumab vedotin alone or in combination with obinutuzumab synergistically enhances in-vitro cytotoxicity and cytokine release against CD20+/ CD79b+ Burkitt lymphoma (BL) / primary mediastinal large B cell lymphoma (PMBL). *Blood*. 2017;130(Suppl 1):1540.
108. Topp MS, Gokbuget N, Stein AS, Zugmaier G, O'Brien S, Bargou RC, Dombret H, Fielding AK, Heffner L, Larson RA, Neumann S, Foa R, Litzow M, Ribera JM, Rambaldi A, Schiller G, Bruggemann M, Horst HA, Holland C, Jia C, Maniar T, Huber B, Nagorsen D, Forman SJ, Kantarjian HM. Safety and activity of blinatumomab for adult patients with relapsed or refractory B-precursor acute lymphoblastic leukaemia: a multicentre, single-arm, phase 2 study. *Lancet Oncol*. 2015;16(1):57–66.
109. Viardot A, Goebeler ME, Hess G, Neumann S, Pfreundschuh M, Adrian N, Zettl F, Libicher M, Sayehli C, Stieglmaier J, Zhang A, Nagorsen D, Bargou RC. Phase 2 study of the bispecific T-cell engager (BiTE) antibody blinatumomab in relapsed/refractory diffuse large B-cell lymphoma. *Blood*. 2016;127(11):1410–6.
110. Vitolo U, Trneny M, Belada D, Burke JM, Carella AM, Chua N, Abrisqueta P, Demeter J, Flinn I, Hong X, Kim WS, Pinto A, Shi YK, Tatsumi Y, Oestergaard MZ, Wenger M, Fingerle-Rowson G, Catalani O, Nielsen T, Martelli M, Sehn LH. Obinutuzumab or rituximab plus cyclophosphamide, doxorubicin, vincristine, and prednisone in previously untreated diffuse large B-cell lymphoma. *J Clin Oncol*. 2017;35(31):3529–37.
111. von Stackelberg A, Locatelli F, Zugmaier G, Handgretinger R, Trippett TM, Rizzari C, Bader P, O'Brien MM, Brethon B, Bhojwani D, Schlegel PG, Borkhardt A, Rheingold SR, Cooper TM, Zwaan CM, Barnette P, Messina C, Michel G, DuBois SG, Hu K, Zhu M, Whitlock JA, Gore L. Phase I/Phase II study of blinatumomab in pediatric patients with relapsed/refractory acute lymphoblastic leukemia. *J Clin Oncol*. 2016;34(36):4381–9.
112. Wilson WH, Gerecitano JF, Goy A, de Vos S, Kenkre VP, Barr PM, Blum KA, Shustov AR, Advani RH, Lih J, Williams M, Schmitz R, Yang Y, Pittaluga S, Wright G, Kunkel LA, McGreivy J, Balasubramanian S, Cheng M, Moussa D, Buggy JJ, Staudt LM. The Bruton's tyrosine kinase (BTK) inhibitor, ibrutinib (PCI-32765), has preferential activity in the ABC subtype of relapsed/refractory de novo diffuse large B-cell lymphoma (DLBCL): interim results of a multicenter, open-label, Phase 2 study. *Blood*. 2012;120(21):686.
113. Woessmann W, Seidemann K, Mann G, Zimmermann M, Burkhardt B, Oschlies I, Ludwig WD, Klingebiel T, Graf N, Gruhn B, Juergens H, Niggli F, Parwaresch R, Gadner H, Riehm H, Schrappe M, Reiter A. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. *Blood*. 2005a;105(3):948–58.
114. Woessmann W, Seidemann K, Mann G, Zimmermann M, Burkhardt B, Oschlies I, Ludwig WD, Klingebiel T, Graf N, Gruhn B, Juergens H, Niggli F, Parwaresch R, Gadner H, Riehm H, Schrappe M, Reiter A, BFM Group. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. *Blood*. 2005b;105(3):948–58.
115. Younes A, Thieblemont C, Morschhauser F, Flinn I, Friedberg JW, Amorim S, Hivert B, Westin J, Vermeulen J, Bandyopadhyay N, de Vries R, Balasubramanian S, Hellemans P, Smit JW, Fourneau N, Oki Y. Combination of ibrutinib with rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone (R-CHOP) for treatment-naive patients with CD20-positive B-cell non-Hodgkin lymphoma: a non-randomised, phase 1b study. *Lancet Oncol*. 2014;15(9):1019–26.
116. Zhang WY, Wang Y, Guo YL, Dai HR, Yang QM, Zhang YJ, Zhang Y, Chen MX, Wang CM, Feng KC, Li SX, Liu Y, Shi FX, Luo C, Han WD. Treatment of CD20-directed Chimeric Antigen Receptor-modified T cells in patients with relapsed or refractory B-cell non-Hodgkin lymphoma: an early phase IIa trial report. *Signal Transduct Target Ther*. 2016;1:16002.
117. Ziegler JL. Treatment results of 54 American patients with Burkitt's lymphoma are similar to the African experience. *N Engl J Med*. 1977;297(2):75–80.
118. Ziegler JL, Beckstead JA, Volberding PA, Abrams DI, Levine AM, Lukes RJ, Gill PS, Burkes RL, Meyer PR, Metroka CE, et al. Non-Hodgkin's lymphoma in 90 homosexual men. Relation to generalized lymphadenopathy and the acquired immunodeficiency syndrome. *N Engl J Med*. 1984;311(9):565–70.
119. Ziegler JL, Bluming AZ, Magrath IT, Carbone PP. Intensive chemotherapy in patients with generalized Burkitt's lymphoma. *Int J Cancer*. 1972;10(2):254–61.
120. Ziegler JL, Morrow RH Jr, Fass L, Kyalwazi SK, Carbone PP. Treatment of Burkitt's tumor with cyclophosphamide. *Cancer*. 1970;26(2):474–84.

Primary Mediastinal and Gray Zone Lymphomas

14

Lisa Giulino-Roth and Kieron Dunleavy

Introduction

Primary mediastinal B-cell lymphomas (PMBCL) make up approximately 10% of all diffuse large B-cell lymphomas (DLBCL). Unlike DLBCL where the median age at diagnosis is over 60 years, PMBCL is almost exclusively seen in the adolescent and young adult (AYA) population. Although historically categorized as a subtype of DLBCL, PMBCL is demographically, clinically, and biologically very distinct from other subtypes of DLBCL. In fact, its clinical and biological features are much more closely related to those of nodular sclerosing Hodgkin lymphoma (NSHL). PMBCL and NSHL have similar clinical presentations and share approximately a third of their genes as well as common driver mutations, which have been recently identified [1, 2]. There is now a recognition that aggressive mediastinal B-cell lymphomas lie on a continuum of diseases with NSHL and PMBCL on opposite ends, and in between are mediastinal B-cell lymphomas with features intermediate between PMBCL and NSHL that have been termed mediastinal gray zone lymphomas (MGZL). These are rare and interestingly affect males more than females (in contrast to PMBCL), and studies that have focused on their biology suggest that MGZL is a unique molecular entity and distinct from the parent entities of PMBCL and NSHL [3].

The optimal therapy for PMBCL is controversial due to the rarity of this entity and the very recent recognition of it being a distinct entity [4]. In the past, most approaches included consolidation mediastinal radiation as early studies demonstrated that it was very effective when combined with chemotherapy. However, recently, the long-term toxicities of mediastinal radiation in a young population have been realized, and attempts have been made in this disease to develop approaches that obviate the need for radiation and eliminate the risk of its long-term side effects. Recently, increased dose intensity approaches, both in single-center and multicenter settings, have obviated the need for radiation while maintaining excellent cure rates. Novel insights into tumor biology have identified that mediastinal lymphomas harbor several unique targetable pathways, and ongoing studies are focused on targeting these molecular aberrations and critical pathways for lymphomagenesis.

Biology

In the WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues, PMBCL is recognized as a distinct entity based on its clinical presentation, immunophenotypic characteristics, and molecular profile that is markedly different from the germinal center B-cell (GCB) and activated B-cell (ABC) subtypes of DLBCL [5]. Mediastinal B-cell lymphomas, originating from a thymic B-cell, can be considered to lie on a pathobiological continuum of diseases with NSHL and PMBCL lying on either end and MGZL – with features intermediate and transitional between the PMBCL and NSHL – lying in between (Fig. 14.1) [6]. Recently, many insights into the biology of PMBCL have paved the way for investigating novel agents and new approaches in this disease. We now have improved understanding of genetic alterations and perturbations in the JAK-STAT and NFκB pathways and additionally recognition that mediastinal lymphomas are "immune privileged" with the ability to avoid immune destruction [7]. These recently elucidated genetic alterations underpin phenotypic characteristics of these lymphomas and are at play across the aforementioned pathobiological continuum, providing evidence that these entities are molecularly related and likely derived from a common thymic B-cell [2, 8].

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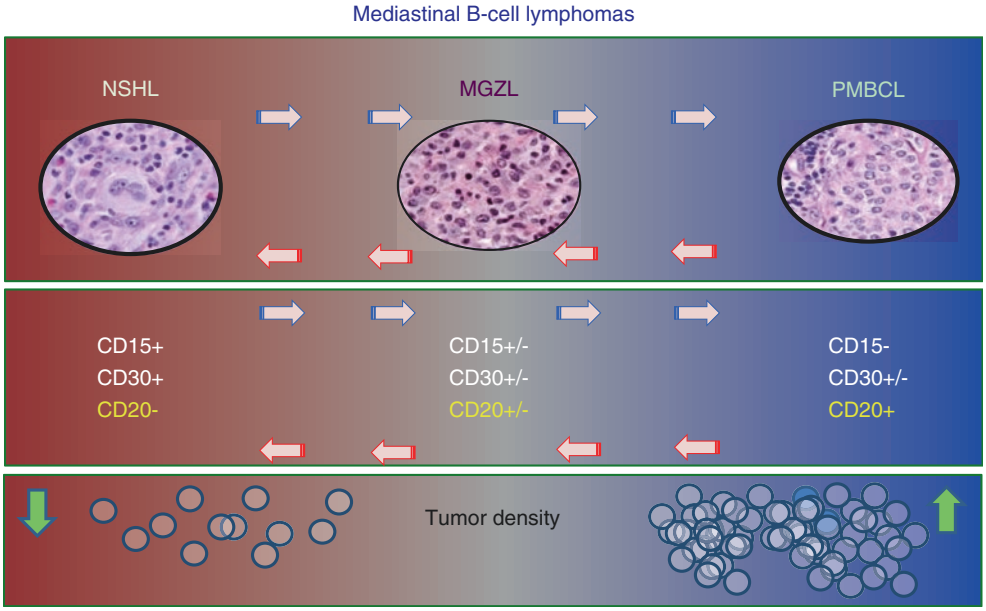

Fig. 14.1 Pathologic features of mediastinal B-cell lymphomas

JAK-STAT Pathway

Several functional genomic studies have identified that JAK-STAT signaling activation is a critical pathway in these two entities [9]. In PMBCL, JAK-STAT signaling likely depends on both IL13 receptor-mediated signaling and constitutive activation, resulting from various somatic gene mutations – among these are JAK2 amplifications, deletions or inactivating mutations of the negative regulators *SOCS1* and *PTPN1*, and mutations of *STAT6* [10]. More than half of PMBCL cases have genomic gains of chromosome 9p containing a locus for JAK2, and the minimally amplified region contains multiple genes including *JAK2* and the programmed death ligands *CD274* (*PDL1*), *PDCD1LG2* (*PDL2*), and *JMJD2C* [11] that contribute synergistically to the pathogenesis of PMBCL [7]. Somatic mutations of *SOCS1* have a similar frequency in PMBCL and are also found in a significant proportion of classical Hodgkin lymphoma (CHL) cases [12–14]. PTPN1 also negatively regulates JAK-STAT signaling, and mutations in this gene have been found in approximately 20% of PMBCL and CHL [2]. Point mutations in *STAT6* may transcriptionally contribute to the pathogenesis of PMBCL, and these have been reported in 36% of PMBCL cases [15]. NF-kappa B genes are typically activated in PMBCL with nuclear translocation of c-REL in most cases [16].

Tumor Microenvironment in PMBCL

The role of non-tumor cells in cross-talk and signaling to tumor cells is well recognized in the lymphoma microenvironment [17]. In PMBCL, this microenvironment can be highly variable, closely resembling NSHL (high cell diversity and sparse tumor cells) at one end of the continuum and DLBCL (high tumor cell content with sheeting out of these cells) at the other end [17]. "Immune privilege" in PMBCL likely results from downregulation of MHC class I and II molecules, as well as increased expression of programmed death ligands – this results in reduced immunogenicity and T-cell anergy [18, 19]. The genetic basis of these expression phenotypes has recently been partly elucidated, and many of these genetic features are also present in DLBCL arising in classic immune-privilege sites such as primary testicular DLBCL and primary central nervous system lymphoma – this suggests biologic overlap between PMBCL and classic IP-DLBCL.

PDL2 and PDL1 are both critical target genes of chromosome 9p gains and amplifications that are found in over 50% of PMBCL cases [20, 21]. Next-generation sequencing and FISH analysis demonstrated that 20% of PMBCL cases harbor recurrent genomic rearrangements involving the 9p locus, resulting in PDL1 and PDL2 gene fusions [21, 22]. Interestingly, PDL1/PDL2 expression is higher in the rearranged cases compared to cases with gains or amplifications [21]. In PMBCL, the co-amplification of JAK2 and the programmed death ligand locus on chromosome 9p24 suggest that JAK-STAT signaling and acquired immune privilege are synergistic in lymphomagenesis and strategies that combine JAK-STAT and immune checkpoint inhibition may be particularly effective.

Initial Chemotherapy

Primary Mediastinal B-Cell Lymphoma

Since PMBCL is a rare diagnosis, there are few prospective trials designed specifically for this NHL subtype. Rather,

pediatric patients with PMBCL have historically been treated on protocols designed for mature B-NHL which includes Burkitt lymphoma (BL), diffuse large B-cell lymphoma (DLBCL), and PMBCL (Table 14.1). Outcomes among patients with PMBCL have been inferior compared to patients with DLCBL and BL treated on the same protocols. For example, the Berlin-Frankfurt-Munster (BFM) Group reported outcomes among 30 pediatric patients with PMCBL treated on NHL-BFM trials between 1986 and 1999 [23]. Patients received 4–6 courses of multi-agent chemotherapy that included systemic dexamethasone, vincristine, ifosfamide, cyclophosphamide, methotrexate, cytarabine, etoposide, doxorubicin, and intrathecal methotrexate, cytarabine, and prednisolone. Patients did not receive rituximab or radiation therapy. PMBCL represented 1.8% of all patients treated on these trials. The 5-year event-free survival among patients with PMBCL was 70% (SE 8%) which was inferior to outcomes among other histologic subtypes (5-year EFS 84%, $p = 0.04$). Similar outcomes were reported in a subset analysis of the French-American-British/Lymphome Malins de Burkitt (FAB/LMB) 96 clinical trial. This trial enrolled patients from 1996 to 2001. Patients with stage III PMBCL were assigned to Group B treatment, consisting of 4–5 cycles of multi-agent chemotherapy including cyclophosphamide, vincristine, prednisone, high-dose methotrexate, doxorubicin, cytarabine, and intrathecal methotrexate, hydrocortisone, and cytarabine [24]. Patients with an inadequate response to treatment were changed to more intensive Group C therapy which included of additional cycles of therapy as well as high-dose cytarabine and etoposide [25]. Patients did not receive rituximab or RT. In a subset analysis, patients with stage III mediastinal large B-cell lymphoma ($n = 42$) were compared to patients with non-mediastinal stage III DLBCL ($n = 69$). The 5-year EFS in the mediastinal large B-cell lymphoma and non-mediastinal DLBCL groups were 66% (95% CI 49%–78%) and 85% (95% CI 71%–92%), respectively ($p < 0.001$) [26]. The largest report of pediatric patients with PMBCL is from a pooled database that merged the data from patients enrolled in the AIEOP trials 92 and 97 ($n = 24$) [27]; the BFM trials 86, 90, and 95 ($n = 40$) [28–30]; the SFOP LMB89 trial ($n = 8$) [31]; and the international FAB/LMB96 trial ($n = 42$) [26]. Among 114 pediatric patients with PMBCL, EFS and OS were 67% and 79%, respectively. Outcome did not differ based on national group trial, suggesting that the various regimens used in pediatrics result in similar outcomes [32].

Adult treatment for PMBCL has historically consisted of rituximab + an anthracycline-containing chemotherapy regimen with radiation. Common regimens include R-CHOP (rituximab, cyclophosphamide, doxorubicin, vincristine, prednisone), R-MACOP-B (rituximab, methotrexate, doxorubicin, cyclophosphamide, vincristine, prednisone, and bleomycin), and R-VACOP-B (rituximab, etoposide, doxorubicin, cyclophosphamide, vincristine, prednisone, and bleomycin). These regimens have not been prospectively compared to each other to determine the optimal frontline approach. Event-free survival using these regimens ranges from 62% to 84% in different reports (Table 14.2) [33–40]. More recently, efforts have been made to reduce the exposure to radiation in this young, predominantly female

Table 14.1 Selected studies of PMBCL and GZL in children and adolescents

Reference	Study type	Histologies	*n*	Treatment	EFS
Seidemann et al. [23]	Prospective, subgroup analysis pooled from three studies	PMBCL	28	NHL-BFM 86, 90, 95	70% (5 years)
Gerrard et al. [26]	Prospective, subgroup analysis	Mediastinal B-cell lymphoma	42	FAB/LMB 96	66% (5 years)
Patte et al. [32]	Prospective, pooled data from multiple trials	PMBCL	114	AIEOP 92, 97; BFM/GPOH 86, 90, 95; SFOP LMB89; FAB LMB 96	67%
Burke et al. [44]	Prospective	PMBCL	47	DA-EPOCH-R	72% (2 years)
Giulino-Roth et al. [42]	Retrospective	PMBCL	38	DA-EPOCH-R	81% (3 years)
Wössmann et al. [73]	Prospective	PMBCL	15	DA-EPOCH-R	92 ± 8% (2 years)

Table 14.2 Selected studies of PMCBL and GZL in adults

Reference	Study Type	Histologies	*n*	Treatment	EFS
Savage et al. [36]	Retrospective	PMBCL	153	(R)-CHOP, M/VACOP-B +/− RT	75% (5 years)
Zinzani et al. [39]	Retrospective	PMBCL	74	R-MACOP-B +/− RT	88% (10 years)
Gleeson et al. [34]	Prospective – subgroup analysis	PMBCL	50	R-CHOP 14 (n = 22) +/− RT R-CHOP 21 (n-28) +/− RT	84% (5 years) 80% (5 years)
Dunleavy et al. [41]	Prospective	PMBCL	51	DA-EPOCH-R	93% (3 years)
Giulino-Roth et al. [42]	Retrospective	PMBCL	118	DA-EPOCH-R	87% (3 years)
Wilson et al. [45]	Prospective	GZL	24	DA-EPOCH-R	62% (5 years)
Evens et al. [47]	Retrospective	GZL	112	Various regimens	40% (2 years)

population. One such approach is the infusional DA-EPOCH-R regimen (dose-adjusted etoposide, prednisone, vincristine, cyclophosphamide, doxorubicin, and rituximab), which is given for 6–8 cycles without consolidative radiation. A phase II trial of DA-EPOCH-R among patients at the NCI reported a 5-year EFS of 93% among 51 patients [41]. In the multicenter setting, outcomes with DA-EPOCH-R are not as high as reported in the single-center phase II trial but remain encouraging. In a retrospective study of 118 adults with PMCBL treated across 24 centers, the 3-year EFS was 87% [42]. Similar results have been reported in smaller retrospective series [41, 43].

Given the encouraging adult data with DA-EPOCH-R as well as the understanding that PMBCL is likely biologically similar in children and adults, the most recent international pediatric mature B-NHL trial studied the DA-EPOCH-R regimen for patients with PMBCL (NCT01516567). The preliminary results of this phase II trial were recently reported [44]. Among 47 patients enrolled on trial, the 2-year EFS was 72% (95% CI 57%–84%). This was disappointing compared with the single-center NCI adult data, however did not differ from the outcome among pediatric patients with PMBCL treated on historical trials (p = 0.71). The DA-EPOCH-R regimen has also been evaluated in pediatric patients with PMBCL in a multicenter retrospective study [42]. Among 38 patients under age 21 years, the 3-year EFS was 81% (95 CI 67–94%). Although the EFS in the retrospective series is higher than seen in the prospective trial, the confidence intervals are overlapping, suggesting that the true EFS may lie between the numbers observed in the two studies.

In summary, pediatric patients with PMBCL have been treated with mature B-NHL regimens as well as the DA-EPOCH-R regimen. These approaches include multiagent chemotherapy (+/− rituximab) without radiation. There is no evidence to suggest that a particular regimen is superior, and all are reasonable first-line approaches to PMBCL.

Gray Zone Lymphoma

Gray zone lymphoma is extremely rare in pediatrics, and patients are typically excluded from clinical trials. As a result there are no pediatric trials to guide initial therapy in gray zone lymphoma. Management decisions in GZL are based on data in pediatric mature B-NHL and a small number of studies of GZL in adults. Interestingly, gray zone lymphoma is much more common in males than females compared to the cases with PMBCL and NSHL, which are predominantly diseases of females.

The NCI evaluated the DA-EPOCH-R regimen in adolescents and adults with mediastinal gray zone lymphoma (MGZL) [45]. This prospective trial included 24 patients with a median age of 33 years (range 14–59). Patients received 6–8 cycles of DA-EPOCH-R without radiation therapy. At 59 months of median follow-up, the EFS was 62%. This was significantly lower than the EFS observed at the same center in PMBCL (EFS 93%, p = 0.0005). Similar outcomes were reported in a large retrospective series from the Lymphoma Study Association (LYSA) which included patients treated with a variety of initial regimens [46]. In this series 99 adult patients with MGZL received treatment including HL-like therapy (ABVD and escalated BEACOPP) or DLBCL-like therapy (R-CHOP and high-dose R-CHOP). The 3-year EFS in the entire cohort was 63%. Patients treated with more intensive approaches (escalated BEACOPP and high-dose R-CHOP) had superior outcomes compared to patients treated with less-intensive regimens (ABVD and R-CHOP) (p = 0.003). Another large retrospective series reported outcomes among adult patients with mediastinal and non-mediastinal GZL [47]. Common first-line regimens included CHOP +/− R (46%), ABVD +/−R (30%), and DA-EPOCH-R (10%). The 2-year PFS among the entire cohort was 40%. Outcome did not differ between patients with MGZL and non-mediastinal GZL. Patients treated with ABVD +/− R had a substantially inferior outcome compared to those treated with CHOP +/− R or DA-EPOCH-R (2-year PFS 22% vs. 52%, p = 0.03). Rituximab was associated with improved PFS on multivariate analysis (HR 0.35). MD Anderson Cancer Center has also reported disparities in outcome based on initial treatment regimen [48]. In this smaller series of 16 patients with GZL, the 2-year PFS for patients treated with ABVD, R-CHOP/R-HCAVD, and DA-EPOCH-R were 0%, 25%, and 100%, respectively. Cumulatively, these reports suggest that intensive therapy is required in GZL and that outcomes with ABVD are unacceptably low.

Consolidative Radiation and the Role of End-of-Therapy PET

Primary Mediastinal B-Cell Lymphoma

The role for radiation in PMBCL is not well defined. In pediatric trials, patients have not been treated with consolidative RT. In contrast, this has historically been considered the standard of care for adults with PMBCL. Recognition of the long-term sequelae of radiation, including secondary malignancy and cardiac disease, has driven more recent adult protocols to eliminate radiation or restrict its use to only those patients predicted to have an inferior outcome. One approach is to use a dose-intensive upfront regimen (such as DA-EPOCH-R or dose-dense R-CHOP followed by ICE) without radiation [41, 49, 50]. Another approach is to use a less-intensive upfront regimen (such as R-CHOP) and administer consolidative RT to a subset of patients based on end-of-therapy (EOT) FDG-PET scan.

End-of-therapy FDG-PET is predictive of outcome in PMBCL [41, 42, 51], however must be interpreted with caution. Inflammation in the mediastinum after therapy is common in PMCBL and can lead to false positives. The positive and negative predictive value of FDG-PET after DA-EPOCH-R have been evaluated in a prospective and retrospective series [42, 51]. In both studies EOT FDG-PET had a high negative predictive value (98% and 96%, respectively) but a low positive predictive value (20% and 42%, respectively). Both studies used a Deauville score of >3 to define a positive PET scan.

The question of whether FDG-PET can be used to identify patients for whom RT can be safely omitted has been evaluated in retrospective series. The BCCA reported their experience using R-CHOP followed by an EOT FDG-PET. RT was only administered to patients with a positive EOT PET-scan (not defined by Deauville score). The 5-year time-to-progression was 83%, which was not different than a historical control in the "pre-PET" era where all patients received R-CHOP followed by RT [52]. Similar outcomes were reported using this approach with a R-MACOP-B chemotherapy backbone [39]. The ongoing IELSG-37 trial will be evaluating the omission of RT among patients with a negative EOT-PET [53]. In this study patients with PMCBL treated with a rituximab- and anthracycline-containing regimen will be evaluated by FDG-PET at the completion of therapy. Patients with a negative scan (defined by Deauville 1–3) will be randomized to consolidative RT vs. no further therapy.

Gray Zone Lymphoma

Studies evaluating radiation in GZL are extremely limited, and most treatment decisions mirror that of PMBCL. In the prospective trial of DA-EPOCH-R in GZL, patients did not receive consolidative radiation [45]. In a large adult retrospective series of GZL, radiation was administered to 33% of patients, most of whom had early-stage and/or bulky disease [47]. In this series, response rates did not differ by the administration of RT; however, given the retrospective nature of this study, interpretations are limited. Given the limited data in GZL, an appropriate approach to RT is to use a PMCBL-like protocol.

Management of Relapsed and Refractory Disease

The majority of relapses and progressions in PMBCL and GZL occur early, with most events occurring within 1 year of diagnosis [23, 26, 42]. Disease can be localized to the mediastinum or can spread to distant sites including extranodal sites and the central nervous system. In pediatrics, given the rarity of the disease, there are no retrospective or prospective trials on relapsed disease to guide management.

In adults with relapsed/refractory PMBCL or GZL that is confined to the mediastinum, radiation therapy alone can be curative [41, 42, 45]. Patients with disseminated disease are typically treated with high-dose chemotherapy followed by autologous or allogeneic stem cell transplantation. A retrospective series of 44 adults with relapsed/refractory PMBCL who underwent auto-SCT reported a 4-year PFS of 61% [54]. Similar outcomes have been reported in smaller series [55–57]. A multicenter retrospective series of adults with GZL who underwent auto-SCT has also been reported [58]. Among 33 patients, the 3-year PFS and OS were 69% and 78%, respectively. Outcomes for patients with PMCBL or GZL that does not respond to second-line chemotherapy therapy are poor. In these cases, novel agents should be considered.

Novel Therapies

With the recent advances in our understanding of the biology of PMBCL and GZL, several novel therapeutic targets have been identified and studied in clinical trials (Fig. 14.2). Considering the close clinical and biologic overlap between PMBCL, GZL, and cHL, agents with activity in one lymphoma subtype may be of benefit in others; however, this is not always the case, and prospective trials including these rare subtypes are needed.

The anti-CD30 antibody-drug conjugate brentuximab vedotin, which is known to have activity in cHL, was recently studied in a phase II trial in adults with relapsed/refractory PMBCL. Despite the encouraging results in cHL, responses in PMCBL were only observed in only 2 of 15 patients (13%), and the trial was terminated [59]. In contrast, patients with GZL were included in a separate phase II trial of brentuximab in B-cell NHL [60]. Among six patients with GZL, responses were observed in three, one CR and two PRs. Similar activity was observed in a case series of patients with GZL treated with brentuximab where responses were observed in three of four patients [61].

The majority of PMBCL and GZL cases harbor molecular alterations in 9p24 which include the programmed death ligands 1 and 2 (PD-L1 and PD-L2) and Janus kinase 2 (JAK2), both of which can be therapeutically targeted [11, 20, 62]. Amplification of PD-L1 and PD-L2 likely contributes to tumor immune evasion which could potentially be reversed with monoclonal antibodies targeting the PD-1 checkpoint pathway. The PD-1 inhibitor pembrolizumab has been studied in adults with relapsed/refractory PMCBL in the phase I and phase II settings. In the phase I trial, responses were observed in 7 of 17 patients (41%) including 2 patients

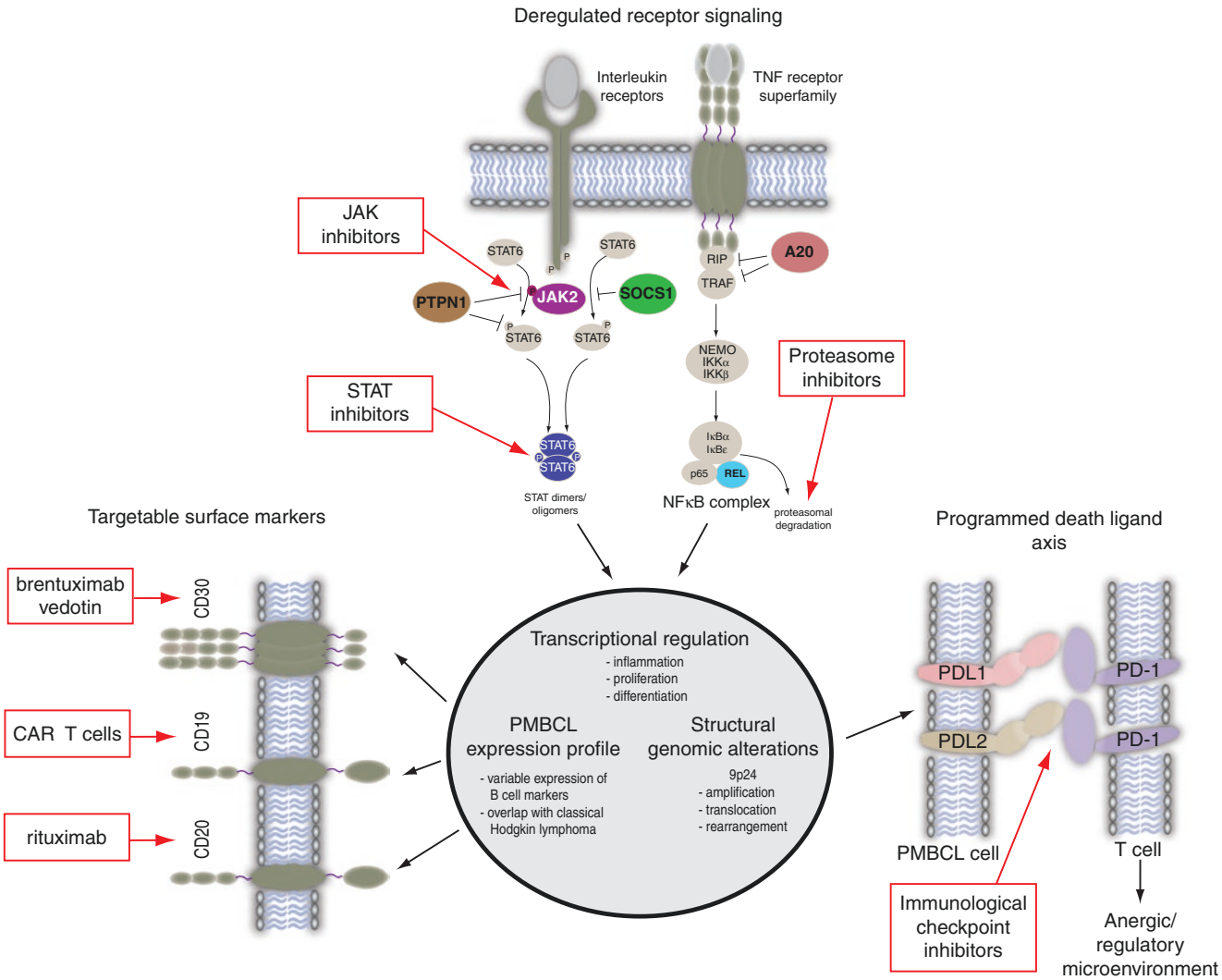

Fig. 14.2 Potential therapeutic targets in primary mediastinal B-cell lymphoma. (used with permission from Elsevier, Non-Hodgkin's Lymphoma in Childhood and Adolescence, 52, Dunleavy and Steidl [8])

who remained in remission beyond the 2-year maximum treatment duration [63]. An interim analysis of the ongoing phase II trial reported a similar response rate (41%) among 49 patients including 4 CRs and 8 PRs [64]. Case reports of GZL also suggest activity for PD-1 blockade [65], and additional checkpoint inhibitors are being developed in B-cell lymphomas [66, 67]. Inhibitors of JAK2, such as ruxolitinib, and others may also have activity in PMBCL and GZL; however too few patients have been treated at this time to draw definitive conclusions [68, 69].

T-cells engineered to express chimeric antigen receptors (CARs) directed at CD-19 have demonstrated activity in adults with refractory B-cell lymphoma including PMBCL. In a phase II trial of the CD-19 CAR axicabtagene ciloleucel, an objective response rate of 84% and a complete response rate of 54% were observed among 111 patients with B-NHL [70]. Twenty-four patients in this trial had PMBCL or transformed follicular lymphoma, among which the ORR was 83%. Other CAR-CD19 platforms have also been evaluated in trials that include PMCBL with similar encouraging results [71, 72]. Two CD-19 CAR-T therapies (axicabtagene ciloleucel and tisagenlecleucel) are now FDA approved in adults with high-grade B-cell lymphoma after two or more lines of systemic therapy.

Summary

PMBCL (and MGZL) represent distinct clinicopathologic entities. Emerging data suggest that they should have a distinct management approach, different to that of other subtypes of DLBCL. These diseases have a high cure rate, but considering the young population they afflict, it is critical to develop strategies that are highly effective but remove the

need for mediastinal radiation which causes significant long-term toxicities. Our understanding of the critical molecular pathways in these lymphomas has advanced significantly over recent times, and, in particular, realizing the role of PD1 ligands and JAK-STAT pathways has paved the way for the novel approaches in PMBCL and MGZL.

References

1. Rosenwald A, Wright G, Leroy K, et al. Molecular diagnosis of primary mediastinal B cell lymphoma identifies a clinically favorable subgroup of diffuse large B cell lymphoma related to Hodgkin lymphoma. J Exp Med. 2003;198(6):851–62.
2. Gunawardana J, Chan FC, Telenius A, et al. Recurrent somatic mutations of PTPN1 in primary mediastinal B cell lymphoma and Hodgkin lymphoma. Nat Genet. 2014;46(4):329–35.
3. Eberle FC, Salaverria I, Steidl C, et al. Gray zone lymphoma: chromosomal aberrations with immunophenotypic and clinical correlations. Mod Pathol. 2011;24(12):1586–97.
4. Dunleavy K, Gross TG. Management of aggressive B cell NHLs in the AYA population: an adult versus pediatric perspective. Blood. 2018;132(4):369–75.
5. Swerdlow SH, Campo E, Harris NL, et al. WHO classification of tumours of haematopoietic and lymphoid tissues. IARC: Lyon; 2008.
6. Grant C, Dunleavy K, Eberle FC, Pittaluga S, Wilson WH, Jaffe ES. Primary mediastinal large B-cell lymphoma, classic Hodgkin lymphoma presenting in the mediastinum, and mediastinal gray zone lymphoma: what is the oncologist to do? Curr Hematol Malig Rep. 2011;6(3):157–63.
7. Steidl C, Gascoyne RD. The molecular pathogenesis of primary mediastinal large B-cell lymphoma. Blood. 2011;118(10):2659–69.
8. Dunleavy K, Steidl C. Emerging biological insights and novel treatment strategies in primary mediastinal large B-cell lymphoma. Semin Hematol. 2015;52(2):119–25.
9. Dubois S, Viailly PJ, Mareschal S, et al. Next-generation sequencing in diffuse large B-cell lymphoma highlights molecular divergence and therapeutic opportunities: a LYSA study. Clin Cancer Res. 2016;22(12):2919–28.
10. Guiter C, Dusanter-Fourt I, Copie-Bergman C, et al. Constitutive STAT6 activation in primary mediastinal large B-cell lymphoma. Blood. 2004;104(2):543–9.
11. Rui L, Emre NC, Kruhlak MJ, et al. Cooperative epigenetic modulation by cancer amplicon genes. Cancer Cell. 2010;18(6):590–605.
12. Melzner I, Bucur AJ, Bruderlein S, et al. Biallelic mutation of SOCS-1 impairs JAK2 degradation and sustains phospho-JAK2 action in the MedB-1 mediastinal lymphoma line. Blood. 2005;105(6):2535–42.
13. Mottok A, Renne C, Willenbrock K, Hansmann ML, Brauninger A. Somatic hypermutation of SOCS1 in lymphocyte-predominant Hodgkin lymphoma is accompanied by high JAK2 expression and activation of STAT6. Blood. 2007;110(9):3387–90.
14. Weniger MA, Melzner I, Menz CK, et al. Mutations of the tumor suppressor gene SOCS-1 in classical Hodgkin lymphoma are frequent and associated with nuclear phospho-STAT5 accumulation. Oncogene. 2006;25(18):2679–84.
15. Ritz O, Guiter C, Castellano F, et al. Recurrent mutations of the STAT6 DNA binding domain in primary mediastinal B-cell lymphoma. Blood. 2009;114(6):1236–42.
16. Savage KJ, Monti S, Kutok JL, et al. The molecular signature of mediastinal large B-cell lymphoma differs from that of other diffuse large B-cell lymphomas and shares features with classical Hodgkin lymphoma. Blood. 2003;102(12):3871–9.
17. Fowler NH, Cheah CY, Gascoyne RD, et al. Role of the tumor microenvironment in mature B-cell lymphoid malignancies. Haematologica. 2016;101(5):531–40.
18. Rigaud G, Moore PS, Taruscio D, et al. Alteration of chromosome arm 6p is characteristic of primary mediastinal B-cell lymphoma, as identified by genome-wide allelotyping. Genes Chromosomes Cancer. 2001;31(2):191–5.
19. Roberts RA, Wright G, Rosenwald AR, et al. Loss of major histocompatibility class II gene and protein expression in primary mediastinal large B-cell lymphoma is highly coordinated and related to poor patient survival. Blood. 2006;108(1):311–8.
20. Green MR, Monti S, Rodig SJ, et al. Integrative analysis reveals selective 9p24.1 amplification, increased PD-1 ligand expression, and further induction via JAK2 in nodular sclerosing Hodgkin lymphoma and primary mediastinal large B-cell lymphoma. Blood. 2010;116(17):3268–77.
21. Twa DDW, Chan FC, Ben-Neriah S, et al. Genomic Rearrangements Involving Programmed Death Ligands Are Recurrent In Primary Mediastinal Large B-Cell Lymphoma. Blood. 2013;122(21):635.
22. Steidl C, Shah SP, Woolcock BW, et al. MHC class II transactivator CIITA is a recurrent gene fusion partner in lymphoid cancers. Nature. 2011;471(7338):377–81.
23. Seidemann K, Tiemann M, Lauterbach I, et al. Primary mediastinal large B-cell lymphoma with sclerosis in pediatric and adolescent patients: treatment and results from three therapeutic studies of the Berlin-Frankfurt-Munster Group. J Clin Oncol. 2003;21(9):1782–9.
24. Patte C, Auperin A, Gerrard M, et al. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. Blood. 2007;109(7):2773–80.
25. Cairo MS, Gerrard M, Sposto R, et al. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. Blood. 2007;109(7):2736–43.
26. Gerrard M, Waxman IM, Sposto R, et al. Outcome and pathologic classification of children and adolescents with mediastinal large B-cell lymphoma treated with FAB/LMB96 mature B-NHL therapy. Blood. 2013;121(2):278–85.
27. Pillon M, Di Tullio MT, Garaventa A, et al. Long-term results of the first Italian Association of Pediatric Hematology and Oncology protocol for the treatment of pediatric B-cell non-Hodgkin lymphoma (AIEOP LNH92). Cancer. 2004;101(2):385–94.
28. Woessmann W, Seidemann K, Mann G, et al. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. Blood. 2005;105(3):948–58.
29. Reiter A, Schrappe M, Parwaresch R, et al. Non-Hodgkin's lymphomas of childhood and adolescence: results of a treatment stratified for biologic subtypes and stage--a report of the Berlin-Frankfurt-Munster Group. J Clin Oncol. 1995;13(2):359–72.
30. Reiter A, Schrappe M, Tiemann M, et al. Improved treatment results in childhood B-cell neoplasms with tailored intensification of therapy: a report of the Berlin-Frankfurt-Munster Group Trial NHL-BFM 90. Blood. 1999;94(10):3294–306.
31. Patte C, Auperin A, Michon J, et al. The Societe Francaise d'Oncologie Pediatrique LMB89 protocol: highly effective multiagent chemotherapy tailored to the tumor burden and initial response in 561 unselected children with B-cell lymphomas and L3 leukemia. Blood. 2001;97(11):3370–9.
32. Patte C, Auperin A, Reiter A. Primary mediastinal large B-cell lymphoma in children and adolescents: Data of the European and American Groups, for SFOP LMB, FAB LMB96, BFM, and AEIO study groups. Ann Oncol. 2005;16

33. Avigdor A, Sirotkin T, Kedmi M, et al. The impact of R-VACOP-B and interim FDG-PET/CT on outcome in primary mediastinal large B cell lymphoma. Ann Hematol. 2014;93(8):1297–304.
34. Gleeson M, Hawkes EA, Cunningham D, et al. Rituximab, cyclophosphamide, doxorubicin, vincristine and prednisolone (R-CHOP) in the management of primary mediastinal B-cell lymphoma: a subgroup analysis of the UK NCRI R-CHOP 14 versus 21 trial. Br J Haematol. 2016;175(4):668–72.
35. Rieger M, Osterborg A, Pettengell R, et al. Primary mediastinal B-cell lymphoma treated with CHOP-like chemotherapy with or without rituximab: results of the Mabthera International Trial Group study. Ann Oncol. 2011;22(3):664–70.
36. Savage KJ, Al-Rajhi N, Voss N, et al. Favorable outcome of primary mediastinal large B-cell lymphoma in a single institution: the British Columbia experience. Ann Oncol. 2006;17(1):123–30.
37. Soumerai JD, Hellmann MD, Feng Y, et al. Treatment of primary mediastinal B-cell lymphoma with rituximab, cyclophosphamide, doxorubicin, vincristine and prednisone is associated with a high rate of primary refractory disease. Leuk Lymphoma. 2014;55(3):538–43.
38. Vassilakopoulos TP, Pangalis GA, Katsigiannis A, et al. Rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone with or without radiotherapy in primary mediastinal large B-cell lymphoma: the emerging standard of care. Oncologist. 2012;17(2):239–49.
39. Zinzani PL, Broccoli A, Casadei B, et al. The role of rituximab and positron emission tomography in the treatment of primary mediastinal large B-cell lymphoma: experience on 74 patients. Hematol Oncol. 2015;33(4):145–50.
40. Zinzani PL, Stefoni V, Finolezzi E, et al. Rituximab combined with MACOP-B or VACOP-B and radiation therapy in primary mediastinal large B-cell lymphoma: a retrospective study. Clin Lymphoma Myeloma. 2009;9(5):381–5.
41. Dunleavy K, Pittaluga S, Maeda LS, et al. Dose-adjusted EPOCH-rituximab therapy in primary mediastinal B-cell lymphoma. N Engl J Med. 2013;368(15):1408–16.
42. Giulino-Roth L, O'Donohue T, Chen Z, et al. Outcomes of adults and children with primary mediastinal B-cell lymphoma treated with dose-adjusted EPOCH-R. Br J Haematol. 2017;179(5):739–47.
43. Pinnix CC, Dabaja B, Ahmed MA, et al. Single-institution experience in the treatment of primary mediastinal B cell lymphoma treated with immunochemotherapy in the setting of response assessment by 18fluorodeoxyglucose positron emission tomography. Int J Radiat Oncol Biol Phys. 2015;92(1):113–21.
44. Burke GAA, Gross TG, Pillon M, et al. Results of Inter-B-NHL Ritux 2010 – Phase II Study of DA-EPOCH-R for Children and Adolescents with Primary Mediastinal Large B-Cell Lymphoma (PMLBL) on Behalf of European Intergroup for Childhood Non Hodgkin's Lymphoma (EICNHL) and Children's Oncology Group (COG). Blood. 2017;130(Suppl 1):4124.
45. Wilson WH, Pittaluga S, Nicolae A, et al. A prospective study of mediastinal gray-zone lymphoma. Blood. 2014;124(10):1563–9.
46. Sarkozy C, Molina T, Ghesquieres H, et al. Mediastinal gray zone lymphoma: clinico-pathological characteristics and outcomes of 99 patients from the Lymphoma Study Association. Haematologica. 2017;102(1):150–9.
47. Evens AM, Kanakry JA, Sehn LH, et al. Gray zone lymphoma with features intermediate between classical Hodgkin lymphoma and diffuse large B-cell lymphoma: characteristics, outcomes, and prognostication among a large multicenter cohort. Am J Hematol. 2015;90(9):778–83.
48. Chihara D, Westin JR, Miranda RN, et al. Dose adjusted-EPOCH-R and mediastinal disease may improve outcomes for patients with gray-zone lymphoma. Br J Haematol. 2017;179(3):503–6.
49. Moskowitz C, Hamlin PA, Maragulia J, Meikle J, Zelenetz AD. Sequential dose-dense RCHOP followed by ICE consolidation (MSKCC protocol 01–142) without radiotherapy for patients with primary mediastinal large B cell lymphoma. Blood. 2010;116(21):420.
50. Moskowitz CH, Schoder H, Teruya-Feldstein J, et al. Risk-adapted dose-dense immunochemotherapy determined by interim FDG-PET in Advanced-stage diffuse large B-Cell lymphoma. J Clin Oncol. 2010;28(11):1896–903.
51. Melani C, Advani R, Roschewski M, et al. End-of-treatment and serial PET imaging in primary mediastinal B-cell lymphoma following dose-adjusted-EPOCH-R: a paradigm shift in clinical decision making. Haematologica. 2018;103(8):1337–44.
52. Savage KJ, Yenson PR, Shenkier T, et al. The outcome of primary mediastinal large B-cell lymphoma (PMBCL) in the R-CHOP treatment era. Blood. 2012;120(21):303.
53. Cavalli F, Ceriani L, Zucca E. Functional imaging using 18-Fluorodeoxyglucose PET in the management of primary mediastinal large B-cell lymphoma: the contributions of the International Extranodal Lymphoma Study Group. Am Soc Clin Oncol Educ Book. 2016;35:e368–75.
54. Aoki T, Shimada K, Suzuki R, et al. High-dose chemotherapy followed by autologous stem cell transplantation for relapsed/refractory primary mediastinal large B-cell lymphoma. Blood Cancer J. 2015;5:e372.
55. Hamlin PA, Portlock CS, Straus DJ, et al. Primary mediastinal large B-cell lymphoma: optimal therapy and prognostic factor analysis in 141 consecutive patients treated at Memorial Sloan Kettering from 1980 to 1999. Br J Haematol. 2005;130(5):691–9.
56. Kuruvilla J, Pintilie M, Tsang R, Nagy T, Keating A, Crump M. Salvage chemotherapy and autologous stem cell transplantation are inferior for relapsed or refractory primary mediastinal large B-cell lymphoma compared with diffuse large B-cell lymphoma. Leuk Lymphoma. 2008;49(7):1329–36.
57. Sehn LH, Antin JH, Shulman LN, et al. Primary diffuse large B-cell lymphoma of the mediastinum: outcome following high-dose chemotherapy and autologous hematopoietic cell transplantation. Blood. 1998;91(2):717–23.
58. Kharfan-Dabaja MA, Raj R, Nikolaenko L, et al. Efficacy of high-dose therapy and autologous hematopoietic cell transplantation in gray zone lymphoma: a US Multicenter Collaborative Study. Biol Blood Marrow Transplant. 2018;24(3):486–93.
59. Zinzani PL, Pellegrini C, Chiappella A, et al. Brentuximab vedotin in relapsed primary mediastinal large B-cell lymphoma: results from a phase 2 clinical trial. Blood. 2017;129(16):2328–30.
60. Jacobsen ED, Sharman JP, Oki Y, et al. Brentuximab vedotin demonstrates objective responses in a phase 2 study of relapsed/refractory DLBCL with variable CD30 expression. Blood. 2015;125(9):1394–402.
61. Li J, Sim J, Leung AYH, Kwong YL. Favorable response of relapsed/refractory gray-zone lymphoma to brentuximab vedotin. Ann Hematol. 2018;97(3):551–4.
62. Joos S, Otano-Joos MI, Ziegler S, et al. Primary mediastinal (thymic) B-cell lymphoma is characterized by gains of chromosomal material including 9p and amplification of the REL gene. Blood. 1996;87(4):1571–8.
63. Zinzani PL, Ribrag V, Moskowitz CH, et al. Safety and tolerability of pembrolizumab in patients with relapsed/refractory primary mediastinal large B-cell lymphoma. Blood. 2017;130(3):267–70.
64. Zinzani PL, Thieblemont C, Melnichenko V, et al. Efficacy and safety of pembrolizumab in relapsed/refractory primary mediastinal large B-cell lymphoma (rrPMBCL): updated analysis of the Keynote-170 Phase 2 Trial. Blood. 2017;130(Suppl 1):2833.
65. Melani C, Major A, Schowinsky J, et al. PD-1 blockade in mediastinal gray-zone lymphoma. N Engl J Med. 2017;377(1):89–91.
66. Armand P, Shipp MA, Ribrag V, et al. Programmed death-1 blockade with pembrolizumab in patients with classical hodgkin lymphoma after brentuximab vedotin failure. J Clin Oncol. 2016;34(31):3733–9.

67. Lesokhin AM, Ansell SM, Armand P, et al. Nivolumab in patients with relapsed or refractory hematologic malignancy: preliminary results of a phase Ib study. J Clin Oncol. 2016;34(23):2698–704.
68. Kim SJ, Kang HJ, Dong-Yeop S, et al. The efficacy of JAK2 inhibitor in heavily pretreated classical Hodgkin lymphoma: a prospective pilot study of ruxolitinib in relapsed or refractory classical Hodgkin lymphoma and primary mediastinal large B-cell lymphoma. Blood. 2016;128(22):1820.
69. Younes A, Hilden P, Coiffier B, et al. International Working Group consensus response evaluation criteria in lymphoma (RECIL 2017). Ann Oncol. 2017;28(7):1436–47.
70. Neelapu SS, Locke FL, Bartlett NL, et al. Axicabtagene ciloleucel CAR T-cell therapy in refractory large B-cell lymphoma. N Engl J Med. 2017;377(26):2531–44.
71. Abramson JS, Palomba ML, Gordon LI, et al. CR rates in relapsed/refractory (R/R) aggressive B-NHL treated with the CD19-directed CAR T-cell product JCAR017 (TRANSCEND NHL 001). J Clin Oncol. 2017;35(15_suppl):7513.
72. Kochenderfer JN, Dudley ME, Kassim SH, et al. Chemotherapy-refractory diffuse large B-cell lymphoma and indolent B-cell malignancies can be effectively treated with autologous T cells expressing an anti-CD19 chimeric antigen receptor. J Clin Oncol. 2015;33(6):540–9.
73. Wöessmann W, Lisfeld J, Burkhardt B, NHL-BFM Study Group. Therapy in primary mediastinal B-cell lymphoma. N Engl J Med. 2013;369(3):282. https://doi.org/10.1056/NEJMc1305983.

15 Epstein-Barr Virus-Associated Post-Transplantation Lymphoproliferative Disease

Ashley V. Geerlinks, Thomas G. Gross, and Angela S. Punnett

Introduction

Post-transplantation lymphoproliferative disorders (PTLD) encompass a diverse spectrum of pathologic and clinical entities that may develop in the setting of decreased T-cell function and altered immune surveillance following haematopoietic stem cell transplantation (HSCT) and solid organ transplantation (SOT). PTLD is most commonly of B-cell origin and EBV-associated, particularly among those patients post HSCT or early after SOT. Late-onset disease in the solid organ transplant population is increasingly described and is more likely to be EBV-negative and of monomorphic pathology. In spite of growing consensus on diagnosis and management, PTLD continues to be a major cause of morbidity and mortality following transplantation. This chapter will review the pathobiology, epidemiology, presentation and therapy for EBV-associated B-cell PTLD.

Pathobiology

EBV infection is identified in many cases of PTLD and appears to play an important role in the etiology of these disorders [35, 153]. Post-transplant immunosuppression in a patient who carries EBV reduces the activity of the patient's EBV-specific cytotoxic T-cell surveillance, which increases the chances of uncontrolled proliferation of EBV-infected B cells and subsequent progression to PTLD [35]. Patients who experience primary EBV infection, either via allograft or natural route of transmission via oral secretions from a EBV-infected individual, appear to be particularly susceptible to developing EBV-positive PTLD [168], presumably due to inability to develop an adequate anti-EBV immune response soon enough to control B-cell proliferation.

However, EBV is not found in all PTLD, and there is some evidence that the incidence of EBV-negative PTLD may be increasing [100]. EBV-negative PTLD is rare following haematopoietic stem cell transplantation (HSCT), is more common in adults and in late-onset PTLD (>2 years post-transplant) [35]. The pathobiology of EBV-negative PTLD is less clear. Transplantation increases the risk of cancer of all types [66]. The chronicity and/or intensity of immunosuppression certainly may decrease the anti-tumour immune surveillance, but some immunosuppressants, e.g. anti-metabolites such as azathioprine or mycophenolate mofetil, may cause DNA damage. In support of this hypothesis, EBV-negative PTLD appears to have more genomic alterations, including alterations in known genes in lymphoma genesis, i.e. c-MYC, TP53, BCL6 and RAS [47, 109].

Classification of PTLD is difficult as it refers to a heterogeneous group of lymphoproliferative diseases that can range from uncomplicated, self-limiting disease to widespread nodal and often extranodal disease. The most widely used classification system is the WHO classification, which is based on histology; the latest revision was in 2016 [153]. This classification includes an expansion of "early lesions" defined as lesions that despite cellular proliferation, retain their normal histologic architecture. Such lesions now include: plasmacytic hyperplasia PTLD, infectious mononucleosis PTLD and florid follicular hyperplasia PTLD. These are rarely monoclonal and often EBV-positive, though plasmacytic hyperplasia can be EBV-negative. Next is polymorphic PTLD, defined by disruption of normal architecture but containing a heterogenous admixture of cells. Polymorphic PTLD is often monoclonal and EBV-positive. Monomorphic PTLD is defined as PTLD that is histologically identical to Non-Hodgkin lymphoma (NHL), i.e. diffuse large B-cell lymphoma (DLBCL),

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Burkitt lymphoma (BL), plasma cell PTLD (myeloma-like or plasmacytoma-like), peripheral T-cell lymphoma or hepatosplenic T-cell lymphoma. Monomorphic PTLD is almost always monoclonal. B-cell and T-cell PTLD are often EBV-positive, but plasma cell PTLD is rarely EBV-positive. And finally, there is classical Hodgkin lymphoma (cHL)-like PTLD. This has the same immunophenotype as cHL, i.e. CD45(−), CD15(+). Some polymorphic PTLD can have Reed-Sternberg-like cells, but the immunophenotype is CD45(+) and CD15(−) and should be verified before making the diagnosis of cHL-like PTLD. This classification appears to have no clinical role for PTLD following HSCT, and it is unclear how clinically relevant this histologic classification is for prognosis or treatment decisions for PTLD following SOT. A biopsy from a single lesion can have a mix of histologies, and there can be a discordance in histology between different lesions in the same patient [63]. In general, early lesions tend to respond to less aggressive therapy, i.e. reduction of immunosuppression. Polymorphic lesions also tend to respond more often to less aggressive therapies [71]. Unfortunately, histology has been difficult to correlate with outcome [63].

Epidemiology

With the increasing number of solid organ transplants performed and improved long-term survival, PTLD has become the most common cancer following SOT during childhood and adolescence [48, 145]. It is a significant contributor to the burden of NHL (specifically DLBCL and BL) in this age group with a risk 100 to 200 times that of the general population [178]. A bimodal distribution of presentation is described with early disease diagnosed within the first 1–2 years of transplantation and predominantly EBV-associated with B-cell histology. A later peak of presentation occurs at 3–5 years from transplant with ongoing risk noted out to 10 years from transplant [43, 116, 140, 170]. This later disease is not necessarily associated with EBV and is more likely to include BL, cHL and T-cell lymphoma. With clinical experience in monitoring and titrating immunosuppressive medications, and pre-emptive management with new or rising blood EBV titres, the incidence of early disease may be decreasing [24, 111] while, as already noted, that of later disease may be increasing [100]. PTLD following HSCT is the most common de novo malignancy occurring early after HSCT, typically within 3–4 months, and is relatively uncommon thereafter. Rates of disease are also noted to be decreasing in the most recent treatment eras [94].

The assessment of risk factors for PTLD is complicated by the complex nature of the disease and the variability among studies with respect to disease definitions, immunosuppression protocols and the length of follow up. Identified risk factors include EBV status at the time of transplant, host factors including age, and the degree of immunosuppression as determined primarily by the organ transplanted or donor source for hematopoietic stem cell transplant and cumulative exposure to immunosuppressive medications.

EBV Status

Among SOT recipients, the highest risk for PTLD results from transplant of an EBV-positive donor to an EBV-naïve recipient [24, 33, 78, 135, 136]. The similar serological EBV mismatch is an established risk factor for PTLD following HSCT [161]. Newly detected and/or rapidly rising EBV load is used by most SOT and HSCT centres as an indication to reduce immunosuppression (RI) and/or begin rituximab, respectively (see below). The association between degree and duration of EBV load and development of PTLD after SOT is less clear with chronic high load EBV carriers post liver and kidney transplant not necessarily at increased risk of disease and, conversely, EBV-PTLD reported in the context of low EBV load [59, 78]. Chronic high load EBV carriers may be at increased risk following heart transplant [13], however, illustrating the interplay of allograft, degree of immunosuppression and perhaps other risk factors for the development of disease.

The role of co-infection in PTLD risk with other viruses, including CMV and other herpes viruses, is controversial [124, 136, 176] though may be more significant after HSCT [176].

Host Factors

Age at transplant is a significant risk factor for PTLD post SOT with younger children at increased risk in most studies [24, 172], likely reflecting the proportion of patients who are EBV naïve. A history of an autoimmune condition may be associated with increased risk of PTLD reflecting the role of chronic immunologic stimulation in the disease process and/or degree of immunosuppression [144, 179]. Prior malignancy has also been associated with increased risk among SOT recipients [113].

Cytokine gene polymorphisms affecting synthesis of IFN gamma, IL-10, TNF alpha and TGF beta have been identified to correlate with EBV-PTLD risk in small studies and may represent disturbances in the innate immune system response with alterations in the Th1/Th2 balance [6, 97, 104]. HLA gene polymorphisms in both recipient and donor may similarly be associated with the risk of PTLD risk but require further study in larger cohorts [78, 87, 126].

Immunosuppression

The incidence of PTLD differs by organ transplanted and by stem cell source reflecting both the intensity of immunosuppression required to prevent rejection and graft versus host disease (GvHD) respectively, and the potential number of EBV-carrying donor B cells in associated lymphoid tissue within the allograft.

Solid Organ Transplantation

The reported incidences of PTLD post SOT in children by transplanted organ include: 2–3% for kidney [48, 78], 2–10% for liver [110, 111], up to 6–7% for heart [24, 72, 170], up to 15% for lung [110] and 10–15% for intestinal transplant [112, 125].

The increased incidence of PTLD among recipients of SOT with history of rejection [111] and among HSCT recipients with GvHD [94] supports the concern for intensity and cumulative exposure of immunosuppression for risk of PTLD [51]. It is difficult to appreciate the absolute risks associated with specific immunosuppressive agents. A 'learning curve' with new agents has been described such that the incidence of PTLD may be higher when such agents are first introduced to clinical care [111, 124]. Registry data is limited in detail and a universally accepted measure of intensity of immunosuppression does not exist, although an analysis of type, dosage and trough levels has been proposed [78]. Historically, the use of monoclonal anti-T-cell antibodies (OKT3 and antithymocyte globulin (ATG)) have been associated with the highest risk of PTLD [88, 115]. However, recent registry data describe no increased risk or lower risk of PTLD following induction immunosuppression [51, 72], and a systematic review did not find an association between ATG dose and risk of PTLD, with the lowest risk reported for those receiving antiviral prophylaxis [103]. The highest risk was reported with the use of OKT3, likely due to its long-lasting depletional effects on T lymphocytes. IL-2 receptor inhibitor antibodies (daclizumab, basiliximab) do not appear to significantly increase PTLD risk [56]. Alemtuzumab, targeting both T and B cells, has not been associated with an increased risk [88]. Belatacept selectively blocks T-cell co-stimulation and has been associated with a greater risk of PTLD in the central nervous system (CNS) among EBV-seronegative kidney transplant recipients [61].

The use of calcineurin inhibitors (CNI) seems to confer the next highest risk for PTLD. Although early studies suggested the use of tacrolimus increases the risk of PTLD compared with cyclosporine, this risk may be mitigated with close monitoring of serum levels with dose adjustment and the use of additional CNI-sparing immunosuppressive agents [32, 78, 171]. Higher trough tacrolimus levels have been reported in the 2 months preceding PTLD diagnosis among paediatric liver transplant recipients compared with those with asymptomatic EBV infection or other viral infections [50]. The MTOR inhibitor sirolimus has been unexpectedly associated with increased risk of PTLD in kidney transplant recipients, particularly among those recipients who are EBV-naïve [88, 113]. The use of antimetabolites, including mycophenolate mofetil, in maintenance immunosuppression does not appear to be associated with increased risk of PTLD and indeed may decrease risk by decreasing CNI exposure [32, 51].

Haematopoietic Stem Cell Transplantation

The frequency of PTLD after HSCT has been reported at 3.2% in a retrospective review of adult and paediatric data from the European Bone Marrow Transplant (EBMT) registry [149] and in keeping with rates of 2–4% reported in single institution series [82, 161]. Donor source and T-cell depletion strategies have been identified as the most important risk factors for development of PTLD following HSCT in a large Centre for International Blood and Marrow Transplant Research (CIBMTR) database study [94]. The incidence of PTLD ranged from 1% in matched related donor recipients to 11% in mismatched unrelated donor recipients in the EBMT study [149]. Although a similar increase in incidence with increasing mismatch was found in the CIMBTR study, there was a strong interaction with T-cell depletion and ATG use [94]. Selective T-cell depletion conferred the highest risk for PTLD followed by ATG use for prophylaxis or treatment of GvHD. One solution proposed to mitigate this risk is the use of a TCR alpha beta-/CD19-depleted graft and the use of rituximab in conditioning [92].

The presence of acute or chronic GvHD is associated with a moderate risk for development of PTLD [94]. Initial reports of the use of T-cell replete grafts with post-transplant cyclophosphamide for GvHD prophylaxis are very favourable with no reported cases of PTLD in the early transplant period. Investigators hypothesize that this approach destroys both donor and recipient EBV-infected B cells, preserves viral immunity with relative sparing of specific memory T cells and allows for rapid immune recovery [83, 155].

Clinical Presentation

The clinical presentation of PTLD is highly variable, depending on the underlying pathology, the type of transplant and the time since transplant. It is important to maintain a high index of suspicion because the onset of PTLD can be insidious and nonspecific. Frequently, patients present with relatively benign findings (episodic and unex-

plained fever, weight loss, fatigue) before developing more significant symptomatology. Fulminant PTLD is relatively rare, presenting as a rapidly progressive disease resulting in multiorgan failure and is more commonly seen following HSCT [64]. Other EBV-associated diseases must be differentiated from PTLD, although the initial management is similar. Early-onset disease is more likely to be extranodal and involve the allograft [140]. The major differential diagnostic considerations for early-onset PTLD include allograft rejection and infection [89]. Late-onset disease is more likely to involve nodal sites or present with dissemination [140].

Outside the allograft, common areas affected by PTLD include lymphoid tissues, GI tract, lung and liver [3]. Disease classified pathologically as early lesions often presents with adenotonsillar involvement [1] with associated obstructive symptoms (new-onset snoring or mouth breathing). Involvement of the GI tract may present with vomiting, diarrhoea, bleeding, intussusception or obstruction. Perforation may occur at presentation or immediately following initiation of therapy in the presence of transmural lesion necrosis. Chronic ulceration in intestinal transplant recipients should prompt a biopsy to rule out PTLD with samples from the ulcer edge and the intervening mucosa [142]. New-onset anaemia or hypoalbuminaemia may indicate GI involvement. Lung disease may result in respiratory insufficiency or asymptomatic nodules. Although bronchoalveolar lavage fluid analysis for EBV was initially proposed to be predictive for PTLD of the lung, this has not been replicated in a larger study [119]. Liver disease may present as diffuse hepatitis or nodular lesions. PTLD of the central nervous system (CNS), isolated or as part of multiorgan disease, has been described and is often a late presentation with monomorphic and EBV-positive pathology. CNS PTLD appears to be more frequent in renal transplant recipients [45]. Patients may present with headache, seizures or focal neurologic findings. Examination of the CSF may be helpful to differentiate CNS PTLD from encephalitis [54, 175].

The use of EBV viral load monitoring has provided a powerful tool for surveillance and pre-emptive management of patients at risk for PTLD. Such monitoring shows high sensitivity but is limited by poor specificity for determining PTLD risk. In addition, the optimal sample type, reporting units, trigger points and monitoring algorithms have yet to be defined, though there is a newly developed reference standard for inter-laboratory calibration for EBV NAT assays and some clinical recommendations exist [49, 119, 134, 146, 147]. Complementary biomarkers have been proposed to refine risk prediction and diagnosis but remain under study. The most promising to date include plasma levels of IL-6 and IL-10 [77], sCXCL13[139], markers of B-cell activation including sCD30 and immunoglobulin-free light chains [44, 67] and potentially functional NK cell changes including PD1 expression [173]. Given the heterogeneity of the disease, a single predictive model may not be realistic [40].

Algorithms for diagnostic and staging evaluations have been published and mirror the evaluation for suspected NHL (see for example the NCCN Guidelines 2019 available at https://www.nccn.org/professionals/physician_gls/default.aspx). Initial assessment includes a full physical examination, screening blood tests, including a complete blood count with differential, chemistry panel to assess for tumour lysis syndrome, allograft function screening, and EBV viral-load studies. Bone marrow evaluation and diagnostic lumbar puncture may be deferred in the absence of monomorphic disease if clinical, laboratory and imaging studies do not support their involvement. Ultrasound seems to be effective for initial imaging in patients with suspected abdominal or soft-tissue PTLD [128]. PET-CT is increasingly used to follow up equivocal findings, to guide biopsy site selection, for staging and for response assessment [8, 163].

PTLD Therapy

Treatment strategies for PTLD must be tailored to individual patient contexts and take into consideration multiple factors including disease presentation and pathology, patient comorbidities and performance, risk of rejection, organ graft function and immunosuppressive regimen. Clinical decision-making requires input from the interdisciplinary team including an oncologist, a transplant specialist, and an infectious disease specialist. While there is no universally accepted standard treatment for PTLD, both a risk-adapted and response-adapted approach are increasingly being used.

Existing studies examining prognostic factors are based on small numbers and heterogeneous patient populations with sometimes conflicting results. Most of these studies point to EBV-negative, monomorphic, fulminant disease, late-onset and CNS disease as poor prognostic factors [20, 34, 37, 39, 62, 71, 101, 159, 170]. Interestingly, Gross et al. in their prospective multicentre Phase II paediatric therapeutic study did not identify any difference in outcome for paediatric patients with polymorphic versus monomorphic disease [63]. Poor performance status, increased number of involved sites and CD20-negative PTLD have also been suggested to be poor prognostic markers [96, 107].

Reduction of Immunosuppression (RI)

The initial approach to managing patients with PTLD after SOT is RI when graft function allows, with the goal to restore EBV-reactive cytotoxic T lymphocytes (CTL) function. There is no standard definition of RI, including how much and which agents to reduce or discontinue and decisions are

based on the type of allograft, time from transplant and previous history of rejection. Generally, the recommendation is first to reduce immunosuppression by 25–50% with the expectation of clinical response within 2–4 weeks [129]. Recent prospective clinical studies evaluating second-line therapies define failed RI as greater than or equal to 50 per cent reduction for greater than 1 week [63, 157]. RI is described as most effective for early lesions, for polymorphic disease, and for those patients diagnosed early following transplantation; however success with monomorphic PTLD has been described [71, 170].

Reported response rates for RI vary greatly, ranging from 20% to 89% [5, 71, 127, 170]. However, any amount of RI appears to be beneficial as Aull et al. described in adult cardiac transplant patients prescribed RI as a component of treatment had significantly higher survival compared with those not prescribed RI as a part of therapy [5]. For HSCT recipients, RI may not allow for timely recovery of the new immune system to manage PTLD, and current recommendations include the use of rituximab as first-line therapy with RI if possible [132, 150]. Styczynski et al. found that HSCT patients that had RI as part of their therapy for PTLD experienced significantly less PTLD-related mortality compared to those who received rituximab alone [149].

Mammalian target of rapamycin (mTOR) inhibitors sirolimus (rapamycin) and everolimus have been proposed to have a role in both prevention and treatment PTLD. Use of these agents allows for CNI use for primary immunosuppression and they are known to have both antiproliferative and antineoplastic activity [73]. Given the increased risk described among EBV-naïve kidney transplant recipients receiving sirolimus as part of their immunosuppression regimen, more studies are needed to understand the role of mTOR inhibitors for patients with PTLD.

The decision around reduction versus complete withdrawal of immunosuppression for those patients also receiving chemotherapy is allograft and patient specific. The lymphodepleting effects of most chemotherapy regimens should protect the allograft, but rejection during chemotherapy treatment does occur. Although kidney transplant recipients have been reported to tolerate complete withdrawal while receiving chemotherapy [154], Serre et al. identified better allograft outcomes when maintaining CNI at a reduced level [143]. Thus maintaining some immunosuppression during therapy may be best for managing PTLD and the risk of allograft rejection.

Surgery and Radiation

Complete resection of a solitary lesion may be curative, but is usually combined with RI [4]. Paediatric patients with localized adenotonsillar disease treated with RI and surgery were shown to have promising responses without the need for additional therapy [91]. Radiation is rarely used but may be considered when rapid local responses are required (e.g. airway compression) and in CNS PTLD [45, 129].

Antivirals

The role of antivirals, including acyclovir and ganciclovir, in the treatment of PTLD is controversial. Antivirals inhibit lytic EBV DNA replication by targeting viral tyrosine kinase, however PTLD and other EBV-driven lymphomas develop in the latent stage of EBV infection when thymidine kinase is no longer expressed. One small study investigating the use of arginine butyrate to induce EBV thymidine kinase activity in latently infected B cells did demonstrate complete remission in 4 out of 15 patients and partial response in an additional 6 patients [120]. However, arginine butyrate is no longer available for use in clinical settings.

Rituximab

Monoclonal B-cell antibody therapy alone is the standard of care for PTLD after HSCT and is used as a single agent or part of combined therapy for PTLD after SOT. Monoclonal B-cell antibodies were first demonstrated to be useful in PTLD following HSCT and SOT, using anti-CD21 and anti-CD24 antibodies in 1998 [12]. Benkerrou et al. showed 61% of patients achieved complete response with limited toxicity in this paediatric and adult multicentre study. After this study anti-CD21 and anti-CD24 antibodies were not developed commercially and studies instead started using rituximab, a chimeric anti-CD20 IgG monoclonal antibody consisting of human constant regions linked to murine variable domains. The first case report of single-agent rituximab achieving complete remission (CR) in a paediatric patient with tonsillar PTLD post HSCT was described in 1998 [46].

There are a growing number of adult and paediatric studies using rituximab as a single agent or part of multimodal treatment (Table 15.1) since rituximab has been demonstrated to be efficacious and well tolerated after HSCT and SOT [152]. As a single agent rituximab has been associated with CR rates of 44–71% in paediatric SOT recipients and is less toxic than systemic chemotherapy [106, 169]. Rituximab combined with chemotherapy has been examined in an attempt to improve initial response rates. Gross et al. conducted the first paediatric multicentre, prospective, single-arm study, COG ANHL0221, combining low-dose chemotherapy with rituximab for patients with PTLD post SOT and reported a 2-year event-free survival (EFS) of 71% and overall survival (OS) of 83% [63]. This study demonstrated that

Table 15.1 Chemotherapy and Rituximab Studies in PTLD

Author	Age	Study size	Therapy	Transplant type	Pathology	EBV status	Outcome
Webber et al. [169]	Ped	40	Rituximab 375 mg/m^2 IV × 4 doses	SOT (40)	Polymorphic (27) Monomorphic (10) Hodgkin like (2) Nonspecific (1)	Positive (38) Negative (2)	CR 71% 1.5-yr OS 76%
Gross et al. [62]	Ped	36	Cyclophosphamide 600 mg/m^2 IV day 1 × 6 cycles Prednisone 2 mg/kg PO day 1–5 × 6 cycles	SOT (36)	Not reported	Positive (36)	CR 75% 2-yr EFS 67% 2-yr OS 73%
Messahel et al. [106]	Ped	18	Rituximab 375 mg/m^2 IV × 1–4 doses	HSCT (8) SOT (10)	Polymorphic (13) Monomorphic (9)	Positive (18)	CR 44% 2-yr OS 61%
Gross et al. [63]	Ped	55	Cyclophosphamide 600 mg/m^2 IV day 1 × 6 cycles Prednisone 2 mg/kg PO day 1–5 × 6 cycles Rituximab 375 mg/m^2 day 1, 8, 15 each cycle, total of 6 doses	SOT (55)	Polymorphic (8) Monomorphic (29) Poly- and monomorphic (3)	Positive (55)	CR 69% 2-yr EFS 71% 2-yr OS 83%
Trappe et al. [158]	Adult	70	Rituximab 375 mg/m^2 IV weekly × 4 doses then 4 week break with no treatment then CHOP 21 × 4 cycles (cyclophosphamide 750 mg/m^2 IV day 1, doxorubicin 50 mg/m^2 IV day 1, vincristine 1.4 mg/m^2 IV day 1, and prednisone 50 mg/m^2 PO days 1–5)	SOT (70)	Polymorphic (3) Monomorphic (67)	Positive (29) Negative (37)	CR 57% 3-yr PFS 54% 3-yr OS 61%
Maecker-Kolhoff et al. [102]	Ped	49	Rituximab 375 mg/m^2 IV weekly × 3 doses If CR or PR (32): Rituximab 375 mg/m^2 IV × 3 doses If SD or PD (17): vincristine[a] day 1, prednisone[a] day 1, cyclophosphamide[a] day 1, methotrexate[a] day 15 × 6 cycles	SOT (49)	Polymorphic (12) Monomorphic (37)	Positive (44)	CR 73% 2-yr EFS 67% 2-yr OS 86%
Trappe et al. [157]	Adult	152	Rituximab 375 mg/m^2 IV weekly × 4 doses If CR(37): Rituximab 375 mg/m^2 IV q21 days × 4 doses If PR, SD or PD (111): R-CHOP 21 × 4 cycles (rituximab 375 mg/m^2 IV day 1, cyclophosphamide 750 mg/m^2 IV day 1, doxorubicin 50 mg/m^2 IV day 1, vincristine 1.4 mg/m^{2}IV day 1, and prednisone 50 mg/m^2 PO days 1–5)	SOT (152)	Early (2) Polymorphic (20) Monomorphic (129)	Positive (67) Negative (77)	CR 70% 3-yr PFS 75% 3-yr OS 70%

Ped paediatric study, *Adult* adult study, *SOT* solid organ transplant, *HSCT* hematopoietic stem cell transplant, *CR* complete remission, *PR* partial response, *SD* stable disease, *PD* progressive disease, *EFS* event-free survival, *PFS* progression-free survival, *OS* overall survival, *IV* intravenous, *PO* orally
[a]doses not available

rituximab was well tolerated when combined with low-dose chemotherapy [63]. Gupta et al., in a single-centre retrospective chart review study, demonstrated significant improvement with this rituximab and low-dose chemotherapy regimen including higher complete response rate and lower risk of recurrence and mortality when compared to historical treatment regimens [65].

Newer studies are demonstrating that disease response after rituximab may be used to risk stratify patients to more aggressive or less aggressive treatments. Trappe et al. studied 152 adult SOT recipients that failed RI and were started on rituximab induction [157]. These patients were then stratified to receive single-agent rituximab consolidation if CR was achieved or rituximab with conventional chemotherapy if there was less than CR [157]. The proportion without progression and OS at 3 years were 75% and 70%, respectively, and 25% of patients were treated with rituximab alone with a 3-year proportion without progression of 89% [157]. A similar paediatric study using disease response after rituximab to risk stratify patients is still being conducted in Europe (Ped-PTLD Pilot 2005) with the use of moderate-dose chemotherapy for those patients without a CR [102]. Preliminary results for the first 49 patients show 64% were treated with single agent rituximab and the 2-year EFS was 67% and OS was 86% [102].

Monoclonal antibody use in paediatric HSCT recipients dates back to 1988 in which a case report describes two paediatric HSCT recipients successfully treated for PTLD with anti-CD21 and anti-CD24 antibodies [14]. In HSCT recipients, rituximab is a recommended first-line therapy for biopsy-proven PTLD [150]. A recent analysis by the European Group for Blood and Bone Marrow Transplantation

of 144 cases of PTLD, adult and paediatric, showed an overall survival of 69.4% after rituximab-based treatment [149].

Loss of CD20 expression on tumour cells (CD20- tumour cells) has been described with rituximab use [28, 177]. Clinicians needs to be aware of this potential especially in patients who have progression or refractory disease with rituximab-based treatment.

Chemotherapy

Historically, conventional lymphoma treatment protocols were used for upfront therapy of PTLD. Adult patients experienced significant toxicities with these regimens including infections and multiorgan failure causing significant treatment-related mortality [27, 37, 42]. Although paediatric patients tolerate conventional lymphoma regimens better compared to adults, myelotoxicity and organ toxicity are still major concerns [71]. Current treatment approaches include initial low-dose chemotherapy, salvage chemotherapy after failed treatment with rituximab or initial conventional lymphoma regimens for aggressive PTLD subtypes (such as Burkitt lymphoma). See Table 15.1 for a summary of studies.

Low-dose chemotherapy regimens have been developed and studied in paediatric SOT recipients based on the hypothesis that these regimens would be effective by simultaneously controlling the lymphoproliferative process, preventing allograft rejection, and minimizing treatment-related mortality. Gross et al. demonstrated that paediatric patients with EBV-positive PTLD who failed to respond to RI can be treated with low-dose chemotherapy with CR rate of 75% and relapse reported in only 19% [62]. Treatment included cyclophosphamide and prednisone every 3 weeks for six cycles. COG ANHL 0221, which combined low-dose chemotherapy and rituximab, achieved a similar CR rate of 69% and only 6% relapsed [63]. These low-dose regimens are well tolerated and easy to administer. Interestingly, Gross' COG study identified some patients at the end of treatment who did not achieve a CR but were eventually in CR 28 weeks later without further therapy suggesting some patients may respond at a slower rate [63]. There are no studies directly comparing low-dose chemotherapy versus rituximab.

It is controversial if patients that progress or relapse post rituximab may have benefited from more aggressive chemotherapy regimens upfront. Trappe et al. initially reported favourable OR rates after chemotherapy in adults and concluded PTLD generally remains chemotherapy-sensitive after progression following first-line rituximab [156]. More recent studies have reported on a significant portion of patients that cannot be salvaged despite more aggressive chemotherapy after rituximab [63, 157]. Better risk stratification is needed to provide the most appropriate therapy upfront.

Conventional cytotoxic chemotherapy continues to be used by many as the first-line treatment for Burkitt lymphoma [36, 122]. COG ANHL 0221 did include patients with Burkitt lymphoma with some reported responses; however, cytogenetics and/or c-myc rearrangement status was not known in all of these cases [63]. Ped-PTLD 2005 trial data indicates that 6/7 patients with Burkitt histology required the moderate chemotherapy regimen; however this is preliminary data and long-term results from this study are not available [102]. It is not clear what the optimal treatment regimen is for Burkitt lymphoma PTLD in both paediatric and adult patients [36].

Cellular Therapy

Cytotoxic EBV-T cell therapy (EBV-CTL) targets cells expressing EBV viral antigens thus correcting the underlying immune defect leading to the development of EBV-PTLD. Cellular therapy has been shown to be associated with minimal toxicity and unlike rituximab and chemotherapy, reconstitutes immunity to EBV instead of causing further immune suppression. CTLs can be collected and manufactured from 3 sources (donor, autologous and third party) and there are a variety of manufacturing processes being studied, including activation with lymphoblastoid cell lines, rapid EBV-CTLS, multimer-selected EBV-CTLs and gamma capture EBV-CTLs, with the hope of decreasing the manufacturing time [16]. To date the cost and availability of EBV-CTLs have limited its used as first-line treatment, although this may be changing as third-party donor pools become more available [166, 174].

In HSCT recipients, donor lymphocyte infusions (DLI) were initially studied as therapy for PTLD. Patients received unmanipulated lymphocytes from their EBV-positive donors that presumably contained a small proportion of EBV-CTLs. This approach did result in tumour regression, however it was also associated with significant inflammation leading to death and significant GvHD due to alloreactive T cells [75, 117]. The other limitation is the time required for the EBV-CTL precursors in the infused population to expand in vivo to an amount that would be sufficient to control lymphoproliferation. Although DLI is still being explored with the addition of suicide genes, attention has moved to minimizing GvHD risk by manufacturing EBV-CTLs in which the CD4+ and CD8+ T lymphocytes are expanded against EBV-specific antigens. An initial pilot study from 1995 demonstrated safe and effective control of EBV-PTLD with EBV-CTLs [130]. Since then larger studies (see Table 15.2) have shown CR rates between 68% and 85%, persistence of the EBV-CTLs in vivo up to 10 years and minimal toxicity including no significant GvHD [76, 131]. The drawback of this approach is the time required to wait while donor-specific CTLs are

Table 15.2 Treatment of EBV-related PTLD with EBV cytotoxic T lymphocytes

CTL Type	Infusions (n)	Study size (n)	Age (years)	Type of transplant	Pathology	Outcome	Author
Donor	4	1	17	HSCT	Monomorphic	8 months no recurrence	Rooney et al. [130]
Donor	1–4	13	Not indicated	HSCT	Not indicated	CR 85% PD 15% 2 yr. OS 62%	Heslop et al. [76]
Donor	1	6	19–46	HSCT	Not indicated	CR 50% 2 yr. OS 33%	Moosmann et al. [108]
Donor	1–3	19	13 (8–44)	HSCT	Early (1) Monomorphic (17) Hodgkin (1)	CR 68%	Doubrovina et al. [38]
Donor	1–2	8	8–51	HSCT	Monomorphic (3) Unknown (5)	CR 75%	Icheva et al. [80]
Autologous	2–5	5	2–14	SOT	Early (1) Monomorphic (4)	CR 100%	Comoli et al. [30]
Autologous	1–4	2	1, 3.3	SOT	Not indicated	CR 50% PR 50%	Savoldo et al. [137]
Third party	3	2	Not indicated	SOT	Monomorphic (2)	CR 100%	Sun et al. [151]
Third party	1–8	33	1–76	HSCT and SOT	Early (7) Polymorphic (9) Monomorphic (12) Hodgkin (5)	CR 42% NR 48% 6mo OS 79%	Haque et al. [68]
Third party	1, 4, 8	3	18, 52, 58	SOT	Monomorphic (3)	CR 66%	Gandhi et al. [53]
Third party	5, 9	2	10, 32	HSCT	Monomorphic (2)	CR 100% 15mo OS 100%	Barker et al. [10]
Third party	1–3	5	6–44	HSCT	Monomorphic (5)	CR 40%	Gallot et al. [52]
Third party	1–4	10	3 (1–12)	SOT	Polymorphic (2) Monomorphic (7) Hodgkin (1)	CR 80% 2yo OS 89%	Chiou et al. [25]

generated, which is not always an option in this patient population. Thus "off the shelf" third party EBV-CTLs closely matched by human leukocyte antigens (HLA) are being manufactured and studied since these could be readily available for patients. There are a few small, single-centre reports describing efficacy in HSCT recipients. Better response rates are noted with closer HLA matches, and no immediate or delayed toxicity using third-party EBV-CTLs has been described [10, 52, 68]. Some experts suggest reserving third party banked EBV-CTLs for SOT recipients and HSCT recipients with unrelated donors [16]. Regardless of EBV-CTL source, clinicians need to be proactive in procuring EBV-CTLs to minimize the time to therapy [166].

The implementation of CTL therapy in SOT recipients with PTLD is more problematic as the prolonged duration of immunosuppressive drugs limit the ability of CTLs to generate, expand and persist in vivo. Savoldo et al. demonstrated that autologous EBV-CTLS in SOT recipients can be used to treat PTLD; however expansion was decreased to 3- to five-fold compared to 32-fold in HSCT recipients, there was a decrease of EBV-CTLs within 2–6 months and an inability to generate CTLs in two patients [137]. Despite these limitations, Savoldo and Comoli demonstrated infusions of EBV-CTLs in SOT patients are safe and do not cause graft rejection [30, 137]. Due to similar concerns of the time limitations for generating CTLs and the added manufacturing challenges, "off the shelf" third-party EBV CTLs have been studied in SOT recipients and again found to be safe and efficacious [53, 68, 151]. The most recent publication from Chiou et al. demonstrated a median interval from diagnosis to first EBV-CTL infusion of 26 days, shortest 14 days, and a significant 2-year OS of 89% in paediatric patients who had persistent or progressive disease after RI and/or rituximab [25]. This significantly improved response is speculated to be due to high degrees of HLA matching between recipient and donor EBV-CTLs [25, 68]. Many of the patients described in these publications had failed multiple previous lines of therapy prior to using EBV-CTLs, thus leading to questions about possible improved outcomes if EBV-CTLs were used as upfront therapy. A current Children's Oncology Group pilot study, ANHL1522, is studying the feasibility of using third-party EBV-CTLs at multiple paediatric centres for patients with incomplete response to three cycles of rituximab; there are no preliminary results yet available. These EBV-CTLs will be latent membrane protein (LMP)-specific T cells which are generated using a reproducible and standardized technology [17]. The study has excluded patients with Burkitt morphology, bone marrow (>25%) or CNS involvement, and fulminant PTLD.

There have been no reports describing the use of other immunotherapies for PTLD to date. B-cell disease provides appropriate targets for bispecific T-cell engagers (e.g. blinatumomab) or chimeric antigen receptor (CAR) T cells, but there is a theoretical concern about their activity in the immunosuppressed host. Checkpoint inhibitors (e.g. nivolumab, pembrolizumab) and immunomodulators (e.g. lenalidomide) similarly may be considered for therapy but with significant concern for rejection and risk of GvHD. New immune conjugates with directed cytotoxicity and less off-target toxicity may be beneficial (e.g. new CD22 toxins). In addition, CD30 is highly expressed in PTLD samples and thus may be a potential therapeutic target [165].

Exceptional Sites

CNS-PTLD following SOT and HSCT is rare, and published data include case reports and small retrospective studies. Most paediatric prospective studies have excluded patients with CNS involvement. PTLD with CNS involvement has been described to have a very poor prognosis; however patients with isolated CNS PTLD are described to have a better prognosis compared to multiple site involvement [20]. In addition, at initial diagnosis CNS disease is usually already multifocal [21]. Diagnostic biopsy is important for planning therapy, however complete or near complete surgical resection of brain lesions is discouraged in cases of suspected CNS-PTLD [45, 93]. The use of MRI, in particular diffusion-weighted imaging, and the EBV-DNA load in the CSF may play an important role in diagnosis and function as biomarkers for monitoring treatment [15, 55].

Although RI alone has been shown to achieve complete remissions in a few patients with CNS-PTLD, the reports were complicated by lack of information on the WHO classification for the lesions [21]. Instead, RI is typically used in conjunction with other therapies [45]. Retrospective studies have described radiation, systemic chemotherapy, including high-dose methotrexate, intravenous rituximab and/or intrathecal chemotherapy, and all have been described to achieve complete remissions in some patients, but none have been associated with a significantly better response or survival when compared to the other [20, 21, 45].

Intravenous rituximab alone has been shown to result in CR [45, 99] even though the concentration of rituximab achieved in the cerebral spinal fluid is only 0.1–0.2% of intravenous concentration [69]. Since CNS penetration is poor, intrathecal rituximab, ranging between 2 and 14 doses of 10-40 mg/dose, has been used and shown to be safe and efficacious [18, 22, 31, 164]. These patients received intrathecal rituximab in combination with systemic therapy, either conventional chemotherapy or rituximab. Six of eleven patients have been described to achieve complete remissions with only intravenous and intrathecal rituximab [22, 31]. The most significant side effects described were grade III neuropathy and seizures that were self-limited [22]. EBV-CTLs have also been shown to be efficacious and safe in a few patients with CNS-PTLD [10, 38, 80, 151]. Haque et al. demonstrated two of four patients with CNS-PTLD achieved a complete response with EBV-CTLs alone [68]. Thus, some patients may achieve complete remission without systemic chemotherapy and whole-brain radiation even if we do not have a reliable means of identifying such patients upfront.

Intraocular PTLD is another very rare complication after transplant and as of 2018 only 20 cases have been described. Bilateral ocular involvement and systemic disease are common at diagnosis [81]. Described treatments included RI, radiation, chemotherapy and enucleation. EBV CTLs and intravenous rituximab have also been described to successfully treat patients, and it has been hypothesized that inflammation caused by the PTLD leads to breakdown of the blood-aqueous barrier allowing effective ocular penetration [81]. Intraocular rituximab injections were also safely used in one paediatric patient with PTLD [11].

PTLD must be considered in the differential diagnosis of mucocutaneous ulcers in transplant recipients. Interestingly, multiple patients have been found to have mucocutaneous lesions that are EBV-positive PTLD in the absence of EBV-DNA in their blood [70]. Most reports describe starting with RI or RI with rituximab which can result in complete remission and no recurrence in some patients, including patients with monomorphic PTLD [70]. There are reports describing patients that failed both RI and rituximab, and instead required conventional lymphoma treatment to achieve CR [7, 90].

Preventative Management

Donor EBV Serologic Status

When feasible, avoiding EBV-positive donors in EBV-naive recipients may prevent PTLD. In HSCT, EBV status is used to guide donor selection; however in SOT this is impractical given the very small proportion of EBV-negative donors (6%) [95, 150].

Prophylactic Antivirals

The role of antiviral prophylaxis for prevention of EBV-related PTLD is controversial. Antiviral agents limit the lytic phase of EBV and thus are felt to have some importance in the early phase of transformation [121]. A recent systemic review and meta-analysis of prophylactic antiviral agents in paediatric and adult EBV-naïve SOT recipients found no significant difference in the rate of EBV-associated PTLD

across types of organ transplants and age groups [2]. For HSCT recipients, antiviral agents are not recommended as prophylaxis [150].

Prophylactic Passive and Active Immunization

Increasing levels of anti-Epstein-Barr nuclear antigens (EBNA) antibodies, including those introduced through transfusion, have correlated with decreasing EBV load, suggesting a potential role for intravenous immunoglobulins (IVIG) in controlling EBV-infected cells [60]. The potential prophylactic benefit of CMV-IVIG against the development of EBV-related PTLD in paediatric liver transplant recipients has been evaluated in a randomized multicentre trial, and no statistically significant differences were observed [58]. In contrast, a significantly larger study including 44,828 kidney transplant recipients found that anti-CMV immunoglobulin was effective in the prevention of early-onset PTLD in kidney transplant patients, but not in the prevention of late-onset PTLD [114]. For HSCT recipients, immunoglobulin is not recommended for prevention of EBV reactivation or disease [150].

EBV vaccination may be effective in PTLD prevention, especially in EBV-seronegative transplant candidates, but the vaccines to date have had no reliable long-lasting effects on immunity. This therapy is not commercially available and its potential is controversial [123].

Prophylactic B-Cell Depletion

In SOT recipients, Schachtner et al. gave EBV-seronegative kidney transplant recipients one dose of rituximab 4 weeks prior to receiving a transplant from a living EBV-seropositive donor. None of these patients developed EBV viremia, and 60% remained EBV seronegative after transplant [138]. Pretransplant rituximab may prove useful to prevent PTLD in patients receiving SOT from living donors but further studies are needed.

Pretransplant rituximab in the context of T-cell depleted graft was noted to be associated with a decreased risk of PTLD among HSCT recipients in one study [92]. Similarly, some studies have shown a low risk of PTLD in patients that received post transplant cyclophosphamide for GvHD prophylaxis [83, 155].

Pre-Emptive Management

Monitoring EBV-DNA Load

Development of EBV-associated PTLD is often preceded by increased and/or rising levels of EBV-DNA load in peripheral blood usually 2–16 weeks before EBV-positive PTLD diagnosis [133, 141, 162]. However, the sensitivity and specificity of EBV-DNA load measurements range between 69–100% and 50–86%, respectively, depending on the study, the method of EBV-DNA measurement and the local cut-off used at individual institutions [4, 141, 160, 167].

It is common practice to perform frequent monitoring of EBV-PCR after both SOT and HSCT, although there is no consensus on the frequency of such monitoring after SOT [84, 85, 118]. The American Society of Transplantation 2006 guidelines suggests all SOT recipients who are seronegative or less than 1 year of age should at a minimum have screening at baseline, every 1 month for 12 months, and with presentation of symptoms [79]. For HSCT recipients, guidelines recommend at least weekly titres starting 4 weeks after HSCT and continuing for at least 4 months [150]. The goal of monitoring is to start pre-emptive management as early as possible and to identify less advanced histiological disease. After implementation of an EBV monitoring program, paediatric liver transplant recipients were more likely to be diagnosed with early-lesion and polymorphic PTLD and achieved better outcomes, when compared to historical controls [86]. Patients that received in vivo T-cell depletion or umbilical cord transplants have been described as having a higher risk of EBV viremia; however EBV viremia and PTLD were successfully managed with monitoring and pre-emptive treatment [19, 41]. Clinicians should be aware that EBV DNA load alone cannot be used to exclude the diagnosis of PTLD [74].

Pre-Emptive Therapies

The first-line approach in the management of rising EBV DNA measurements is RI if possible. Studies in paediatric liver transplant and intestinal transplant recipients have demonstrated a 4–14% decrease in incidence of PTLD compared to historical controls; however, these studies were confounded by the lack of standardization of RI [57, 98, 105, 147, 148]. This approach has also been described as effective and safe in other transplant recipients, including lung transplant and allogeneic HSCT [9, 19, 23]. The role of antivirals is controversial and all studies combine this approach with RI, however AlDabbagh's systemic review concluded that there was no significant difference in the rate of EBV-associated PTLD with pre-emptive antiviral therapy [2].

Rituximab has been successfully used as a pre-emptive treatment for PTLD in the HSCT population and is a recommended practice [28, 150]. Pre-emptive rituximab has not been well studied in the SOT population with only one adult cardiac transplant study in which 9 patients received pre-emptive rituximab after failed RI. One patient developed PTLD and there were no significant side effects described

[26]. More studies are needed to evaluate the efficacy and safety of pre-emptive rituximab in SOT recipients. As for EBV-CTLs, multiple studies have now demonstrated in HSCT recipients and a few SOT recipients that EBV-CTLs are safe and effective with no patients developing PTLD [28, 29, 76, 137]. EBV-CTLs are recommended for pre-emptive treatment in HSCT recipients [150].

Conclusion

PTLD is a heterogeneous disease with varied pathologies and presentations. Increasing knowledge of the risk factors associated with PTLD have led to some successful pre-emptive practices and a decreased incidence of early disease. Therapy is assuming a risk and response-based approach with less toxicity and improved survival [34]. However, PTLD remains a significant cause of morbidity and mortality following paediatric transplant and significant contributor to the paediatric NHL burden. An improved understanding of the molecular pathobiology of the disease and the complex interplay of host, EBV and tumour is necessary to better define high risk groups, refine pathological diagnosis and tailor management of PTLD. Prevention of disease remains the ultimate goal.

References

1. Akbas A, Tiede C, Lemound J, Maecker-Kolhoff B, Kreipe H, Hussein K. Post-transplant lymphoproliferative disorders with naso- and oropharyngeal manifestation. Transpl Int. 2015;28:1299–307.
2. AlDabbagh MA, Gitman MR, Kumar D, Humar A, Rotstein C, Husain S. The role of antiviral prophylaxis for the prevention of Epstein-Barr virus-associated Posttransplant lymphoproliferative disease in solid organ transplant recipients: a systematic review. Am J Transplant. 2017;17:770–81.
3. Allen UD, Farkas G, Hebert D, Weitzman S, Stephens D, Petric M, Tellier R, Ngan B, Fecteau A, West L, Wasfy S. Risk factors for post-transplant lymphoproliferative disorder in pediatric patients: a case-control study. Pediatr Transplant. 2005;9:450–5.
4. Allen U, Diane H, Dorothy M, Yigal D, Samia W. Epstein–Barr virus-related post-transplant lymphoproliferative disease in solid organ transplant recipients, 1988–97: a Canadian multi-centre experience. Pediatr Transplant. 2001;5:198–203.
5. Aull MJ, Buell JF, Jennifer T, Roy First M, Alloway RR, Hanaway MJ, Wagoner L, Gross TG, Beebe T, Steve Woodle E. Experience with 274 cardiac Transplant recipients with posttransplant lymphoproliferative disorder: a report from the Israel Penn International Transplant Tumor Registry. Transplantation. 2004;78:1676–82.
6. Babel N, Vergopoulos A, Trappe RU, Oertel S, Hammer MH, Karaivanov S, Schneider N, Riess H, Papp-Vary M, Neuhaus R, Gondek LP, Volk HD, Reinke P. Evidence for genetic susceptibility towards development of posttransplant lymphoproliferative disorder in solid organ recipients. Transplantation. 2007;84:387–91.
7. Bahmanyar M, Shakibazad N, Imanieh MH, Dehghani SM. Soft palate ulcer: an unusual presentation of a posttransplant lymphoproliferative disorder. J Pediatr Hematol Oncol. 2017;39:e97–9.
8. Bakker NA, Pruim J, de Graaf W, van Son WJ, van der Jagt EJ, van Imhoff GW. PTLD visualization by FDG-PET: improved detection of extranodal localizations. Am J Transplant. 2006;6:1984–5.
9. Bakker NA, Verschuuren EA, Erasmus ME, Hepkema BG, Veeger NJ, Kallenberg CG, van der Bij W. Epstein-Barr virus-DNA load monitoring late after lung transplantation: a surrogate marker of the degree of immunosuppression and a safe guide to reduce immunosuppression. Transplantation. 2007;83:433–8.
10. Barker JN, Doubrovina E, Sauter C, Jaroscak JJ, Perales MA, Doubrovin M, Prockop SE, Koehne G, O'Reilly RJ. Successful treatment of EBV-associated posttransplantation lymphoma after cord blood transplantation using third-party EBV-specific cytotoxic T lymphocytes. Blood. 2010;116:5045–9.
11. Bata BM, Pulido JS, Patel SV, Khan SP, Salomao D, Boyce TG, Bothun ED. Combined intraocular and systemic rituximab for ocular lymphoproliferative disorder with extranodal marginal zone lymphoma–type morphology after heart transplant. J Am Assoc Pediatr Ophthalmol Strabismus {JAAPOS}. 2018;22:159–61.
12. Benkerrou M, Jais JP, Leblond V, Durandy A, Sutton L, Bordigoni P, Garnier JL, Le Bidois J, Le Deist F, Blanche S, Fischer A. Anti-B-cell monoclonal antibody treatment of severe posttransplant B-lymphoproliferative disorder: prognostic factors and long-term outcome. Blood. 1998;92:3137–47.
13. Bingler MA, Feingold B, Miller SA, Quivers E, Michaels MG, Green M, Wadowsky RM, Rowe DT, Webber SA. Chronic high Epstein-Barr viral load state and risk for late-onset posttransplant lymphoproliferative disease/lymphoma in children. Am J Transplant. 2008;8:442–5.
14. Blanche S, Le Deist F, Veber F, Lenoir G, Fischer AM, Brochier J, Boucheix C, Delaage M, Griscelli C, Fischer A. Treatment of severe Epstein-Barr virus-induced polyclonal B-lymphocyte proliferation by anti-B-cell monoclonal antibodies: two cases after HLA-mismatched bone marrow transplantation. Ann Intern Med. 1988;108:199–203.
15. Bocian J, Januszkiewicz-Lewandowska D. Utility of quantitative EBV DNA measurements in cerebrospinal fluid for diagnosis and monitoring of treatment of central nervous system EBV-associated post-transplant lymphoproliferative disorder after allogenic hematopoietic stem cell transplantation. Ann Transplant. 2014;19:253–6.
16. Bollard CM, Rooney CM, Heslop HE. T-cell therapy in the treatment of post-transplant lymphoproliferative disease. Nat Rev Clin Oncol. 2012;9:510–9.
17. Bollard CM, Stephen G, Helen Huls M, Leen AM, Gee AP, Rooney CM. Good manufacturing practice-grade cytotoxic T lymphocytes specific for latent membrane proteins (LMP)-1 and LMP2 for patients with Epstein–Barr virus-associated lymphoma. Cytotherapy. 2011;13:518–22.
18. Bonney DK, Htwe EE, Turner A, Kelsey A, Shabani A, Hughes S, Hughes I, Wynn RF. Sustained response to intrathecal rituximab in EBV associated post-transplant lymphoproliferative disease confined to the central nervous system following haematopoietic stem cell transplant. Pediatr Blood Cancer. 2012;58:459–61.
19. Bordon V, Padalko E, Benoit Y, Dhooge C, Laureys G. Incidence, kinetics, and risk factors of Epstein–Barr virus viremia in pediatric patients after allogeneic stem cell transplantation. Pediatr Transplant. 2012;16:144–50.
20. Buell JF, Gross TG, Hanaway MJ, Trofe J, Roy-Chaudhury P, First MR, Woodle ES. Posttransplant lymphoproliferative disorder: significance of central nervous system involvement. Transplant Proc. 2005;37:954–5.
21. Cavaliere R, Petroni G, Lopes MB, Schiff D. Primary central nervous system post-transplantation lymphoproliferative disorder. Cancer. 2010;116:863–70.
22. Ceppi F, Weitzman S, Woessmann W, Davies K, Lassaletta A, Reismuller B, Mellgren K, Uyttebroeck A, Maia I, Abdullah S,

Miakova N, Glaser D, Cohn R, Abla O, Attarbaschi A, Alexander S. Safety and efficacy of intrathecal rituximab in children with B cell lymphoid CD20+ malignancies: an international retrospective study. Am J Hematol. 2016;91:486–91.
23. Chiereghin A, Prete A, Belotti T, Gibertoni D, Piccirilli G, Gabrielli L, Pession A, Lazzarotto T. Prospective Epstein–Barr virus-related post-transplant lymphoproliferative disorder prevention program in pediatric allogeneic hematopoietic stem cell transplant: virological monitoring and first-line treatment. Transpl Infect Dis. 2016;18:44–54.
24. Chinnock R, Webber SA, Dipchand AI, Brown RN, George JF, Transplant SPH. A 16-year multi-institutional study of the role of age and EBV status on PTLD incidence among pediatric heart transplant recipients. Am J Transplant. 2012;12:3061–8.
25. Chiou FK, Beath SV, Wilkie GM, Vickers MA, Morland B, Gupte GL. Cytotoxic T-lymphocyte therapy for post-transplant lymphoproliferative disorder after solid organ transplantation in children. Pediatr Transplant. 2018;22(2), e13133.
26. Choquet S, Varnous S, Deback C, Golmard JL, Leblond V. Adapted treatment of Epstein–Barr virus infection to prevent posttransplant lymphoproliferative disorder after heart transplantation. Am J Transplant. 2014;14:857–66.
27. Choquet S, Trappe R, Leblond V, Jager U, Davi F, Oertel S. 'CHOP-21 for the treatment of post-transplant lymphoproliferative disorders (PTLD) following solid organ transplantation. haematologica. 2007;92:273–4.
28. Comoli P, Basso S, Zecca M, Pagliara D, Baldanti F, Bernardo ME, Barberi W, Moretta A, Labirio M, Paulli M, Furione M, Maccario R, Locatelli F. Preemptive therapy of EBV-related lymphoproliferative disease after pediatric haploidentical stem cell transplantation. Am J Transplant. 2007;7:1648–55.
29. Comoli P, Labirio M, Basso S, Baldanti F, Grossi P, Furione M, Vigano M, Fiocchi R, Rossi G, Ginevri F, Gridelli B, Moretta A, Montagna D, Locatelli F, Gerna G, Maccario R. Infusion of autologous Epstein-Barr virus (EBV)-specific cytotoxic T cells for prevention of EBV-related lymphoproliferative disorder in solid organ transplant recipients with evidence of active virus replication. Blood. 2002;99:2592–8.
30. Comoli P, Maccario R, Locatelli F, Valente U, Basso S, Garaventa A, Tomà P, Botti G, Melioli G, Baldanti F. Treatment of EBV-related post-renal Transplant lymphoproliferative disease with a tailored regimen including EBV-specific T cells. Am J Transplant. 2005;5:1415–22.
31. Czyzewski K, Styczynski J, Krenska A, Debski R, Zajac-Spychala O, Wachowiak J, Wysocki M. Intrathecal therapy with rituximab in central nervous system involvement of post-transplant lymphoproliferative disorder. Leuk Lymphoma. 2013;54:503–6.
32. Dharnidharka VR, Ho PL, Stablein DM, Harmon WE, Tejani AH. Mycophenolate, tacrolimus and post-transplant lymphoproliferative disorder: a report of the north American pediatric renal Transplant cooperative Study. Pediatr Transplant. 2002;6:396–9.
33. Dharnidharka VR, Lamb KE, Gregg JA, Meier-Kriesche HU. Associations between EBV serostatus and organ transplant type in PTLD risk: an analysis of the SRTR National Registry Data in the United States. Am J Transplant. 2012;12:976–83.
34. Dharnidharka VR, Martz KL, Stablein DM, Benfield MR. Improved survival with recent post-transplant lymphoproliferative disorder (PTLD) in children with kidney transplants. Am J Transplant. 2011;11:751–8.
35. Dharnidharka VR, Webster AC, Martinez OM, Preiksaitis JK, Leblond V, Choquet S. Post-transplant lymphoproliferative disorders. Nat Rev Dis Primers. 2016;2:15088.
36. Dierickx D, Tousseyn T, Gheysens O. How I treat posttransplant lymphoproliferative disorders. Blood. 2015;126:2274–83.
37. Dotti G, Fiocchi R, Motta T, Mammana C, Gotti E, Riva S, Cornelli P, Gridelli B, Viero P, Oldani E, Ferrazzi P, Remuzzi G, Barbui T, Rambaldi A. Lymphomas occurring late after solid-organ transplantation: influence of treatment on the clinical outcome. Transplantation. 2002;74:1095–102.
38. Doubrovina E, Oflaz-Sozmen B, Prockop SE, Kernan NA, Abramson S, Teruya-Feldstein J, Hedvat C, Chou JF, Heller G, Barker JN, Boulad F, Castro-Malaspina H, George D, Jakubowski A, Koehne G, Papadopoulos EB, Scaradavou A, Small TN, Khalaf R, Young JW, O'Reilly RJ. Adoptive immunotherapy with unselected or EBV-specific T cells for biopsy-proven EBV+ lymphomas after allogeneic hematopoietic cell transplantation. Blood. 2012;119:2644–56.
39. Dror Y, Greenberg M, Taylor G, Superina R, Hebert D, West L, Connolly B, Sena L, Allen U, Weitzman S. Lymphoproliferative disorders after organ transplantation in children. Transplantation. 1999;67:990–8.
40. Dulek DE, Michaels MG. Into thin air: predicting PTLD in pediatric lung transplant patients. Pediatr Transplant. 2017;21(6), e13029.
41. Dumas PY, Ruggeri A, Robin M, Crotta A, Abraham J, Forcade E, Bay JO, Michallet M, Bertrand Y, Socié G. Incidence and risk factors of EBV reactivation after unrelated cord blood transplantation: a Eurocord and Societe Francaise de Greffe de Moelle-Therapie Cellulaire collaborative study. Bone Marrow Transplant. 2013;48:253.
42. Elstrom RL, Andreadis C, Aqui NA, Ahya VN, Bloom RD, Brozena SC, Olthoff KM, Schuster SJ, Nasta SD, Stadtmauer EA, Tsai DE. Treatment of PTLD with rituximab or chemotherapy. Am J Transplant. 2006;6:569–76.
43. Engels EA, Pfeiffer RM, Fraumeni JF Jr, Kasiske BL, Israni AK, Snyder JJ, Wolfe RA, Goodrich NP, Bayakly AR, Clarke CA, Copeland G, Finch JL, Fleissner ML, Goodman MT, Kahn A, Koch L, Lynch CF, Madeleine MM, Pawlish K, Rao C, Williams MA, Castenson D, Curry M, Parsons R, Fant G, Lin M. Spectrum of cancer risk among US solid organ transplant recipients. JAMA. 2011;306:1891–901.
44. Engels EA, Savoldo B, Pfeiffer RM, Costello R, Zingone A, Heslop HE, Landgren O. Plasma markers of B-cell activation and clonality in pediatric liver and hematopoietic stem cell transplant recipients. Transplantation. 2013;95:519–26.
45. Evens AM, Choquet S, Kroll-Desrosiers AR, Jagadeesh D, Smith SM, Morschhauser F, Leblond V, Roy R, Barton B, Gordon LI, Gandhi MK, Dierickx D, Schiff D, Habermann TM, Trappe R. Primary CNS posttransplant lymphoproliferative disease (PTLD): an international report of 84 cases in the Modern Era. Am J Transplant. 2013: n/a-n/a
46. Faye A, Van Den Abeele T, Peuchmaur M, Mathieu-Boue A, Vilmer E. Anti-CD20 monoclonal antibody for post-transplant lymphoproliferative disorders. Lancet. 1998;352:1285.
47. Ferreiro JF, Morscio J, Dierickx D, Vandenberghe P, Gheysens O, Verhoef G, Zamani M, Tousseyn T, Wlodarska I. EBV-positive and EBV-negative posttransplant diffuse large B cell lymphomas have distinct genomic and transcriptomic features. Am J Transplant. 2016;16:414–25.
48. Francis A, Johnson DW, Craig JC, Wong G. Incidence and predictors of Cancer following kidney transplantation in childhood. Am J Transplant. 2017;17:2650–8.
49. Fryer JF, Heath AB, Wilkinson DE, Minor PD, Group Collaborative Study. A collaborative study to establish the 1st WHO international standard for Epstein-Barr virus for nucleic acid amplification techniques. Biologicals. 2016;44:423–33.
50. Fukushima D, Sato K, Kawagishi N, Ohuchi N, Satomi S. Epstein-Barr virus--associated posttransplantation lymphoproliferative disorder with tacrolimus metabolism deterioration in infants after living-donor liver transplantation. Transplantation. 2015;99:114–9.
51. Gajarski RJ, Blume ED, Urschel S, Schechtman K, Zheng J, West LJ, Altamirano L, Miyamoto S, Naftel DC, Kirklin JK, Zamberlan

MC, Canter CE, Study IPHT. Infection and malignancy after pediatric heart transplantation: the role of induction therapy. J Heart Lung Transplant. 2011;30:299–308.
52. Gallot G, Vollant S, Saïagh S, Clémenceau B, Vivien R, Cerato E, Bignon J-D, Ferrand C, Jaccard A, Vigouroux S. T-cell therapy using a bank of EBV-specific cytotoxic T cells: lessons from a phase I/II feasibility and safety study. J Immunother. 2014;37:170–9.
53. Gandhi MK, Wilkie GM, Dua U, Mollee PN, Grimmett K, Williams T, Whitaker N, Gill D, Crawford DH. Immunity, homing and efficacy of allogeneic adoptive immunotherapy for posttransplant lymphoproliferative disorders. Am J Transplant. 2007;7:1293–9.
54. Gheorghe G, Radu O, Milanovich S, Hamilton RL, Jaffe R, Southern JF, Ozolek JA. Pathology of central nervous system posttransplant lymphoproliferative disorders: lessons from pediatric autopsies. Pediatr Dev Pathol. 2013;16:67–73.
55. Ginat DT, Purakal A, Pytel P. Susceptibility-weighted imaging and diffusion-weighted imaging findings in central nervous system monomorphic B cell post-transplant lymphoproliferative disorder before and after treatment and comparison with primary B cell central nervous system lymphoma. J Neuro-Oncol. 2015;125:297–305.
56. Glotz D, Chapman JR, Dharnidharka VR, Hanto DW, Castro MC, Hirsch HH, Leblond V, Mehta AK, Moulin B, Pagliuca A, Pascual J, Rickinson AB, Russo FP, Trappe RU, Webster AC, Zuckermann AO, Gross TG. The Seville expert workshop for progress in posttransplant lymphoproliferative disorders. Transplantation. 2012;94:784–93.
57. Green M, Bueno J, Rowe D, Mazariegos G, Qu L, Abu-Almagd K, Reyes J. Predictive negative value of persistent low Epstein-Barr virus viral load after intestinal transplantation in children. Transplantation. 2000;70:593–6.
58. Green M, Michaels MG, Katz BZ, Burroughs M, Gerber D, Shneider BL, Newell K, Rowe D, Reyes J. CMV-IVIG for prevention of Epstein Barr virus disease and posttransplant lymphoproliferative disease in pediatric liver transplant recipients. Am J Transplant. 2006;6:1906–12.
59. Green M, Soltys K, Rowe DT, Webber SA, Mazareigos G. Chronic high Epstein-Barr viral load carriage in pediatric liver transplant recipients. Pediatr Transplant. 2009;13:319–23.
60. Green, Michael, and Marian Michaels. 2010. Prevention of Epstein—Barr virus infection and posttransplant lymphoproliferative disease following transplantation. In: Post-transplant lymphoproliferative disorders, Berlin, Heidelberg: Springer.
61. Grinyo J, Charpentier B, Pestana JM, Vanrenterghem Y, Vincenti F, Reyes-Acevedo R, Apanovitch AM, Gujrathi S, Agarwal M, Thomas D, Larsen CP. An integrated safety profile analysis of belatacept in kidney transplant recipients. Transplantation. 2010;90:1521–7.
62. Gross TG, Bucuvalas JC, Park JR, Greiner TC, Hinrich SH, Kaufman SS, Langnas AN, McDonald RA, Ryckman FC, Shaw BW, Sudan DL, Lynch JC. Low-dose chemotherapy for Epstein-Barr virus-positive post-transplantation lymphoproliferative disease in children after solid organ transplantation. J Clin Oncol. 2005;23:6481–8.
63. Gross TG, Orjuela MA, Perkins SL, Park JR, Lynch JC, Cairo MS, Smith LM, Hayashi RJ. Low-dose chemotherapy and rituximab for posttransplant lymphoproliferative disease (PTLD): a Children's Oncology Group report. Am J Transplant. 2012;12:3069–75.
64. Gross TG, Steinbuch M, DeFor T, Shapiro RS, McGlave P, Ramsay NK, Wagner JE, Filipovich AH. B cell lymphoproliferative disorders following hematopoietic stem cell transplantation: risk factors, treatment and outcome. Bone Marrow Transplant. 1999;23:251–8.
65. Gupta S, Fricker FJ, Gonzalez-Peralta RP, Slayton WB, Schuler PM, Dharnidharka VR. Post-transplant lymphoproliferative disorder in children: recent outcomes and response to dual rituximab/low-dose chemotherapy combination. Pediatr Transplant. 2010;14:896–902.
66. Hall EC, Pfeiffer RM, Segev DL, Engels EA. Cumulative incidence of cancer after solid organ transplantation. Cancer. 2013;119:2300–8.
67. Haque T, Chaggar T, Schafers J, Atkinson C, McAulay KA, Crawford DH. Soluble CD30: a serum marker for Epstein-Barr virus-associated lymphoproliferative diseases. J Med Virol. 2011;83:311–6.
68. Haque T, Wilkie GM, Jones MM, Higgins CD, Urquhart G, Wingate P, Burns D, McAulay K, Turner M, Bellamy C, Amlot PL, Kelly D, MacGilchrist A, Gandhi MK, Swerdlow AJ, Crawford DH. Allogeneic cytotoxic T-cell therapy for EBV-positive posttransplantation lymphoproliferative disease: results of a phase 2 multicenter clinical trial. Blood. 2007;110:1123–31.
69. Harjunpää A, Wiklund T, Collan J, Janes R, Rosenberg J, Lee D, Grillo-López A, Meri S. Complement activation in circulation and central nervous system after rituximab (anti-CD20) treatment of B-cell lymphoma. Leuk Lymphoma. 2001;42:731–8.
70. Hart M, Thakral B, Yohe S, Balfour HH Jr, Singh C, Spears M, McKenna RW. EBV-positive mucocutaneous ulcer in organ transplant recipients: a localized indolent posttransplant lymphoproliferative disorder. Am J Surg Pathol. 2014;38:1522–9.
71. Hayashi RJ, Kraus MD, Patel AL, Canter C, Cohen AH, Hmiel P, Howard T, Huddleston C, Lowell JA, Mallory G, Mendeloff E, Molleston J, Sweet S, DeBaun MR. Posttransplant lymphoproliferative disease in children: correlation of histology to clinical behavior. J Pediatr Hematol Oncol. 2001;23:14–8.
72. Hayes D Jr, Breuer CK, Horwitz EM, Yates AR, Tobias JD, Shinoka T. Influence of Posttransplant lymphoproliferative disorder on survival in children after heart transplantation. Pediatr Cardiol. 2015;36:1748–53.
73. Haynes SE, Saini S, Schowengerdt KO. Post-transplant lymphoproliferative disease and other malignancies after pediatric cardiac transplantation: an evolving landscape. Curr Opin Organ Transplant. 2015;20:562–9.
74. Henry DD, Hunger SP, Braylan RC, Dharnidharka VR. Low viral load post-transplant lymphoproliferative disease localized within the tongue. Transpl Infect Dis. 2008;10:426–30.
75. Heslop HE, Brenner MK, Rooney CM. Donor T cells to treat EBV-associated lymphoma. N Engl J Med. 1994;331:679–80.
76. Heslop HE, Slobod KS, Pule MA, Hale GA, Rousseau A, Smith CA, Bollard CM, Liu H, Wu MF, Rochester RJ, Amrolia PJ, Hurwitz JL, Brenner MK, Rooney CM. Long-term outcome of EBV-specific T-cell infusions to prevent or treat EBV-related lymphoproliferative disease in transplant recipients. Blood. 2010;115:925–35.
77. Hinrichs C, Wendland S, Zimmermann H, Eurich D, Neuhaus R, Schlattmann P, Babel N, Riess H, Gartner B, Anagnostopoulos I, Reinke P, Trappe RU. IL-6 and IL-10 in post-transplant lymphoproliferative disorders development and maintenance: a longitudinal study of cytokine plasma levels and T-cell subsets in 38 patients undergoing treatment. Transpl Int. 2011;24:892–903.
78. Hocker B, Fickenscher H, Delecluse HJ, Bohm S, Kusters U, Schnitzler P, Pohl M, John U, Kemper MJ, Fehrenbach H, Wigger M, Holder M, Schroder M, Billing H, Fichtner A, Feneberg R, Sander A, Kopf-Shakib S, Susal C, Tonshoff B. Epidemiology and morbidity of Epstein-Barr virus infection in pediatric renal transplant recipients: a multicenter, prospective study. Clin Infect Dis. 2013;56:84–92.
79. Humar A, Michaels M. American Society of Transplantation recommendations for screening, monitoring and reporting of infec-

tious complications in immunosuppression trials in recipients of organ transplantation. Am J Transplant. 2006;6:262–74.
80. Icheva V, Kayser S, Wolff D, Tuve S, Kyzirakos C, Bethge W, Greil J, Albert MH, Schwinger W, Nathrath M. Adoptive transfer of Epstein-Barr virus (EBV) nuclear antigen 1–specific T cells as treatment for EBV reactivation and lymphoproliferative disorders after allogeneic stem-cell transplantation. J Clin Oncol. 2012;31:39–48.
81. Iu LP, Yeung JC, Loong F, Chiang AK. Successful treatment of intraocular post-transplant lymphoproliferative disorder with intravenous rituximab. Pediatr Blood Cancer. 2015;62:169–72.
82. Kamani NR, Kumar S, Hassebroek A, Eapen M, LeRademacher J, Casper J, Cowan M, Sanchez de Toledo J, Ferster A, Szabolcs P, Wingard JR, Horwitz E, Filipovich AH. Malignancies after hematopoietic cell transplantation for primary immune deficiencies: a report from the Center for International Blood and Marrow Transplant Research. Biol Blood Marrow Transplant. 2011;17:1783–9.
83. Kanakry JA, Kasamon YL, Bolanos-Meade J, Borrello IM, Brodsky RA, Fuchs EJ, Ghosh N, Gladstone DE, Gocke CD, Huff CA, Kanakry CG, Luznik L, Matsui W, Mogri HJ, Swinnen LJ, Symons HJ, Jones RJ, Ambinder RF. Absence of post-transplantation lymphoproliferative disorder after allogeneic blood or marrow transplantation using post-transplantation cyclophosphamide as graft-versus-host disease prophylaxis. Biol Blood Marrow Transplant. 2013;19:1514–7.
84. Kasiske BL, Zeier MG, Chapman JR, Craig JC, Ekberg H, Garvey CA, Green MD, Jha V, Josephson MA, Kiberd BA. KDIGO clinical practice guideline for the care of kidney transplant recipients: a summary. Kidney Int. 2010;77:299–311.
85. Kelly DA, Bucuvalas JC, Alonso EM, Karpen SJ, Allen U, Green M, Farmer D, Shemesh E, McDonald RA. Long-term medical management of the pediatric patient after liver transplantation: 2013 practice guideline by the American Association for the Study of Liver Diseases and the American Society of Transplantation. Liver Transpl. 2013;19:798–825.
86. Kerkar N, Morotti RA, Madan RP, Shneider B, Herold BC, Dugan C, Miloh T, Karabicak I, Strauchen JA, Emre S. The changing face of post-transplant lymphoproliferative disease in the era of molecular EBV monitoring. Pediatr Transplant. 2010;14:504–11.
87. Kinch A, Sundstrom C, Tufveson G, Glimelius I. Association between HLA-A1 and -A2 types and Epstein-Barr virus status of post-transplant lymphoproliferative disorder. Leuk Lymphoma. 2016;57:2351–8.
88. Kirk AD, Cherikh WS, Ring M, Burke G, Kaufman D, Knechtle SJ, Potdar S, Shapiro R, Dharnidharka VR, Kauffman HM. Dissociation of depletional induction and posttransplant lymphoproliferative disease in kidney recipients treated with alemtuzumab. Am J Transplant. 2007;7:2619–25.
89. Koo J, Dawson DW, Dry S, French SW, Naini BV, Wang HL. Allograft biopsy findings in patients with small bowel transplantation. Clin Transpl. 2016;30:1433–9.
90. Krasuska-Sławińska E, Izabela M-C, Pawłowska J, Dembowska-Bagińska B, Pronicki M, Olczak-Kowalczyk D. Post-Transplant lymphoproliferative disorder (PTLD) manifesting in the Oral cavity of a 13-year-old liver Transplant recipient (LTx). Ann Transplant. 2015;20:478–82.
91. L'huillier AG, Dipchand A, Ng V, Hebert D, Avitzur Y, Solomon M, Yeung S, Allen UD. Post-transplant lymphoproliferative disorder in pediatric patients: Clinical sites of occurrence and related survival rates. In: In *Open Forum Infectious Diseases*, 2342. Oxford University: Press; 2016.
92. Laberko A, Bogoyavlenskaya A, Shelikhova L, Shekhovtsova Z, Balashov D, Voronin K, Kurnikova E, Boyakova E, Raykina E, Brilliantova V, Pirumova V, Novichkova G, Maschan A, Maschan M. Risk factors for and the Clinical impact of cytomegalovirus and Epstein-Barr virus infections in pediatric recipients of TCR-alpha/beta- and CD19-depleted grafts. Biol Blood Marrow Transplant. 2017;23:483–90.
93. Lake W, Chang JE, Kennedy T, Morgan A, Salamat S, Baskaya MK. A case series of primary central nervous system posttransplantation lymphoproliferative disorder: imaging and clinical characteristics. Neurosurgery. 2013;72:960–70. discussion 70
94. Landgren O, Gilbert ES, Rizzo JD, Socie G, Banks PM, Sobocinski KA, Horowitz MM, Jaffe ES, Kingma DW, Travis LB, Flowers ME, Martin PJ, Deeg HJ, Curtis RE. Risk factors for lymphoproliferative disorders after allogeneic hematopoietic cell transplantation. Blood. 2009;113:4992–5001.
95. Lazda VA. Evaluation of Epstein-Barr virus (EBV) antibody screening of organ donors for allocation of organs to EBV serostatus matched recipients. Transplant Proc. 2006;38:3404–5.
96. Leblond V, Dhedin N, Bruneel M-FM, Choquet S, Hermine O, Porcher R, Quoc SN, Davi F, Charlotte F, Dorent R. Identification of prognostic factors in 61 patients with posttransplantation lymphoproliferative disorders. J Clin Oncol. 2001;19:772–8.
97. Lee TC, Savoldo B, Barshes NR, Rooney CM, Heslop HE, Gee AP, Caldwell Y, Scott JD, Goss JA. Use of cytokine polymorphisms and Epstein-Barr virus viral load to predict development of post-transplant lymphoproliferative disorder in paediatric liver transplant recipients. Clin Transpl. 2006;20:389–93.
98. Lee TC, Savoldo B, Rooney CM, Heslop HE, Gee AP, Caldwell Y, Barshes NR, Scott JD, Bristow LJ, O'Mahony CA, Goss JA. Quantitative EBV viral loads and immunosuppression alterations can decrease PTLD incidence in pediatric liver transplant recipients. Am J Transplant. 2005;5:2222–8.
99. Lieberman F, Yazbeck V, Raptis A, Felgar R, Boyiadzis M. Primary central nervous system post-transplant lymphoproliferative disorders following allogeneic hematopoietic stem cell transplantation. J Neuro-Oncol. 2012;107:225–32.
100. Luskin MR, Heil DS, Tan KS, Choi S, Stadtmauer EA, Schuster SJ, Porter DL, Vonderheide RH, Bagg A, Heitjan DF. The impact of EBV status on characteristics and outcomes of posttransplantation lymphoproliferative disorder. Am J Transplant. 2015;15:2665–73.
101. Maecker B, Jack T, Zimmermann M, Abdul-Khaliq H, Burdelski M, Fuchs A, Hoyer P, Koepf S, Kraemer U, Laube GF, Muller-Wiefel DE, Netz H, Pohl M, Toenshoff B, Wagner HJ, Wallot M, Welte K, Melter M, Offner G, Klein C. CNS or bone marrow involvement as risk factors for poor survival in post-transplantation lymphoproliferative disorders in children after solid organ transplantation. J Clin Oncol. 2007;25:4902–8.
102. Maecker-Kolhoff B, Beier R, Zimmermann M, Schlegelberger B, Baumann U, Mueller C, Pape L, Reiter A, Rossig C, Schubert S, Toenshoff B, Wingen A, Meissner B, Kebelmann-Betzing C, Henze G, Kreipe HH, Klein C. Response-adapted sequential Immuno-chemotherapy of post-Transplant lymphoproliferative disorders in pediatric solid Organ Transplant recipients: results from the prospective Ped-PTLD 2005 trial. Blood. 2014;124:4468.
103. Marks WH, Ilsley JN, Dharnidharka VR. Posttransplantation lymphoproliferative disorder in kidney and heart transplant recipients receiving thymoglobulin: a systematic review. Transplant Proc. 2011;43:1395–404.
104. McAulay KA, Haque T, Crawford DH. Tumour necrosis factor gene polymorphism: a predictive factor for the development of post-transplant lymphoproliferative disease. Br J Cancer. 2009;101:1019–27.
105. McDiarmid SV, Jordan S, Kim GS, Toyoda M, Goss JA, Vargas JH, Martin MG, Bahar R, Maxfield AL, Ament ME, Busuttil RW. Prevention and preemptive therapy of postransplant lymphoproliferative disease in pediatric liver recipients. Transplantation. 1998;66:1604–11.
106. Messahel B, Taj MM, Hobson R, Hadzic N, Ramsay A, Hann I, Pinkerton R. Single agent efficacy of rituximab in childhood

immunosuppression related lymphoproliferative disease: a United Kingdom Children's Cancer Study Group (UKCCSG) retrospective review. Leuk Lymphoma. 2006;47:2584–9.
107. Montanari F, Radeski D, Seshan V, Alobeid B, Bhagat G, O'connor OA. Recursive partitioning analysis of prognostic factors in post-transplant lymphoproliferative disorders (PTLD): a 120 case single institution series. Br J Haematol. 2015;171:491–500.
108. Moosmann A, Bigalke I, Tischer J, Schirrmann L, Kasten J, Tippmer S, Leeping M, Prevalšek D, Jaeger G, Ledderose G. Effective and long-term control of EBV PTLD after transfer of peptide-selected T cells. Blood. 2010;115:2960–70.
109. Morscio J, Dierickx D, Tousseyn T. Molecular pathogenesis of B-cell posttransplant lymphoproliferative disorder: what do we know so far? Clin Dev Immunol. 2013;2013:150835.
110. Mynarek M, Schober T, Behrends U, Maecker-Kolhoff B. Posttransplant lymphoproliferative disease after pediatric solid organ transplantation. Clin Dev Immunol. 2013;2013:814973.
111. Narkewicz MR, Green M, Dunn S, Millis M, McDiarmid S, Mazariegos G, Anand R, Yin W, Group Studies of Pediatric Liver Transplantation Research. Decreasing incidence of symptomatic Epstein-Barr virus disease and posttransplant lymphoproliferative disorder in pediatric liver transplant recipients: report of the studies of pediatric liver transplantation experience. Liver Transpl. 2013;19:730–40.
112. Nassif S, Kaufman S, Vahdat S, Yazigi N, Kallakury B, Island E, Ozdemirli M. Clinicopathologic features of post-transplant lymphoproliferative disorders arising after pediatric small bowel transplant. Pediatr Transplant. 2013;17:765–73.
113. Nee R, Hurst FP, Dharnidharka VR, Jindal RM, Agodoa LY, Abbott KC. Racial variation in the development of posttransplant lymphoproliferative disorders after renal transplantation. Transplantation. 2011;92:190–5.
114. Opelz G, Daniel V, Naujokat C, Fickenscher H, Döhler B. Effect of cytomegalovirus prophylaxis with immunoglobulin or with antiviral drugs on post-transplant non-Hodgkin lymphoma: a multicentre retrospective analysis. Lancet Oncol. 2007;8:212–8.
115. Opelz G, Döhler B. Lymphomas after solid Organ Transplantation: a collaborative Transplant Study report. Am J Transplant. 2004;4:222–30.
116. Orjuela MA, Alobeid B, Liu X, Siebert AL, Kott ER, Addonizio LJ, Morris E, Garvin JH, Lobritto SJ, Cairo MS. CD20 expression predicts survival in paediatric post-transplant lymphoproliferative disease (PTLD) following solid organ transplantation. Br J Haematol. 2011;152:733–42.
117. Papadopoulos EB, Ladanyi M, Emanuel D, Mackinnon S, Boulad F, Carabasi MH, Castro-Malaspina H, Childs BH, Gillio AP, Small TN, et al. Infusions of donor leukocytes to treat Epstein-Barr virus-associated lymphoproliferative disorders after allogeneic bone marrow transplantation. N Engl J Med. 1994;330:1185–91.
118. Parker A, Kristin B, Andrew Bradley J, Emery V, Featherstone C, Gupte G, Marcus R, Parameshwar J, Ramsay A, Newstead C. Diagnosis of post-transplant lymphoproliferative disorder in solid organ transplant recipients–BCSH and BTS Guidelines. Br J Haematol. 2010;149:675–92.
119. Parrish A, Fenchel M, Storch GA, Buller R, Mason S, Williams N, Ikle D, Conrad C, Faro A, Goldfarb S, Hayes D Jr, Melicoff-Portillo E, Schecter M, Visner G, Sweet S, Danziger-Isakov L, Children Clinical Trials in Organ Transplantation in. Epstein-Barr viral loads do not predict post-transplant lymphoproliferative disorder in pediatric lung transplant recipients: a multicenter prospective cohort study. Pediatr Transplant. 2017;21:e13011.
120. Perrine SP, Hermine O, Small T, Suarez F, O'Reilly R, Boulad F, Fingeroth J, Askin M, Levy A, Mentzer SJ, Di Nicola M, Gianni AM, Klein C, Horwitz S, Faller DV. A phase 1/2 trial of arginine butyrate and ganciclovir in patients with Epstein-Barr virus-associated lymphoid malignancies. Blood. 2007;109:2571–8.
121. Petrara MR, Giunco S, Serraino D, Dolcetti R, De Rossi A. Post-transplant lymphoproliferative disorders: from epidemiology to pathogenesis-driven treatment. Cancer Lett. 2015;369:37–44.
122. Picarsic J, Jaffe R, Mazariegos G, Webber SA, Ellis D, Green MD, Reyes-Mugica M. Post-transplant Burkitt lymphoma is a more aggressive and distinct form of post-transplant lymphoproliferative disorder. Cancer. 2011;117:4540–50.
123. Posfay-Barbe KM, Siegrist C-A. Immunization and transplantation–what is new and what is coming? Pediatr Transplant. 2009;13:404–10.
124. Quinlan SC, Pfeiffer RM, Morton LM, Engels EA. Risk factors for early-onset and late-onset post-transplant lymphoproliferative disorder in kidney recipients in the United States. Am J Hematol. 2011;86:206–9.
125. Ramos E, Hernandez F, Andres A, Martinez-Ojinaga E, Molina M, Sarria J, Lopez-Santamaria M, Prieto G. Post-transplant lymphoproliferative disorders and other malignancies after pediatric intestinal transplantation: incidence, clinical features and outcome. Pediatr Transplant. 2013;17:472–8.
126. Reshef R, Luskin MR, Kamoun M, Vardhanabhuti S, Tomaszewski JE, Stadtmauer EA, Porter DL, Heitjan DF, E. Tsai de. Association of HLA polymorphisms with post-transplant lymphoproliferative disorder in solid-organ transplant recipients. Am J Transplant. 2011a;11:817–25.
127. Reshef R, Vardhanabhuti S, Luskin MR, Heitjan DF, Hadjiliadis D, Goral S, Krok KL, Goldberg LR, Porter DL, Stadtmauer EA, Tsai DE. Reduction of immunosuppression as initial therapy for posttransplantation lymphoproliferative disorder(bigstar). Am J Transplant. 2011b;11:336–47.
128. Riebel T, Kebelmann-Betzing C, Scheer I. Ultrasound in abdominal and soft-tissue childhood PTLD (post-transplant lymphoproliferative disease). Ultraschall Med. 2007;28:201–5.
129. Rodriguez L, Punnett A. Posttransplant lymphoproliferative disorders following solid organ transplantation. In: Dr. Randhawa G, editor. Organ donation and transplantation – public policy and clinical perspectives. 2012. ISBN: 978-953-51-0039-3, InTech. Available from: http://www.intechopen.com/books/organ-donation-and-transplantation-public-policy-and-clinicalperspectives/posttransplant-lymphoproliferative-disorders-following-solid-organ-transplantation
130. Rooney CM, Ng CYC, Loftin S, Smith CA, Li C, Krance RA, Brenner MK, Heslop HE. Use of gene-modified virus-specific T lymphocytes to control Epstein-Barr-virus-related lymphoproliferation. Lancet. 1995;345:9–13.
131. Rooney CM, Smith CA, Ng CY, Loftin SK, Sixbey JW, Gan Y, Srivastava DK, Bowman LC, Krance RA, Brenner MK, Heslop HE. Infusion of cytotoxic T cells for the prevention and treatment of Epstein-Barr virus-induced lymphoma in allogeneic transplant recipients. Blood. 1998;92:1549–55.
132. Rouce RH, Louis CU, Heslop HE. EBV lymphoproliferative disease after hematopoietic stem cell transplant. Curr Opin Hematol. 2014;21:476.
133. Rowe DT, Webber S, Schauer EM, Reyes J, Green M. Epstein–Barr virus load monitoring: its role in the prevention and management of post-transplant lymphoproliferative disease. Transpl Infect Dis. 2000;3:79–87.
134. Ruf S, Behnke-Hall K, Gruhn B, Reiter A, Wagner HJ. EBV load in whole blood correlates with LMP2 gene expression after pediatric heart transplantation or allogeneic hematopoietic stem cell transplantation. Transplantation. 2014;97:958–64.
135. Sampaio MS, Cho YW, Shah T, Bunnapradist S, Hutchinson IV. Association of immunosuppressive maintenance regimens with posttransplant lymphoproliferative disorder in kidney transplant recipients. Transplantation. 2012;93:73–81.
136. Saueressig MG, Boussaud V, Amrein C, Guillemain R, Souilamas J, Souilamas R. Risk factors for post-transplant lymphopro-

liferative disease in patients with cystic fibrosis. Clin Transpl. 2011;25:E430–6.
137. Savoldo B, Goss JA, Hammer MM, Zhang L, Lopez T, Gee AP, Lin YF, Quiros-Tejeira RE, Reinke P, Schubert S, Gottschalk S, Finegold MJ, Brenner MK, Rooney CM, Heslop HE. Treatment of solid organ transplant recipients with autologous Epstein Barr virus-specific cytotoxic T lymphocytes (CTLs). Blood. 2006;108:2942–9.
138. Schachtner T, Reinke P. Pretransplant prophylactic rituximab to prevent Epstein-Barr virus (EBV) viremia in EBV-seronegative kidney transplant recipients from EBV-seropositive donors: results of a pilot study. Transpl Infect Dis. 2016;18:881–8.
139. Schiffer L, Henke-Gendo C, Wilsdorf N, Hussein K, Pape L, Schmitt C, Haller H, Schiffer M, Klein C, Kreipe H, Maecker-Kolhoff B. CXCL13 as a novel marker for diagnosis and disease monitoring in pediatric PTLD. Am J Transplant. 2012;12:1610–7.
140. Schober T, Framke T, Kreipe H, Schulz TF, Grosshennig A, Hussein K, Baumann U, Pape L, Schubert S, Wingen AM, Jack T, Koch A, Klein C, Maecker-Kolhoff B. Characteristics of early and late PTLD development in pediatric solid organ transplant recipients. Transplantation. 2013;95:240–6.
141. Schubert S, Renner C, Hammer M, Abdul-Khaliq H, Lehmkuhl HB, Berger F, Hetzer R, Reinke P. Relationship of immunosuppression to Epstein-Barr viral load and lymphoproliferative disease in pediatric heart transplant patients. J Heart Lung Transplant. 2008;27:100–5.
142. Selvaggi G, Sarkar S, Mittal N, Acar BC, Weppler D, Kato T, Tryphonopoulos P, Tzakis A, Ruiz P. Etiology and management of alimentary tract ulcers in pediatric intestinal transplantation patients. Transplant Proc. 2006;38:1768–9.
143. Serre J-E, Michonneau D, Bachy E, Noël L-H, Dubois V, Suberbielle C, Kreis H, Legendre C, Mamzer-Bruneel M-F, Morelon E. Maintaining calcineurin inhibition after the diagnosis of post-transplant lymphoproliferative disorder improves renal graft survival. Kidney Int. 2014;85:182–90.
144. Shpilberg O, Wilson J, Whiteside TL, Herberman RB. Pretransplant immunological profile and risk factor analysis of posttransplant lymphoproliferative disease development: the results of a nested matched case-control study. The University of Pittsburgh PTLD Study Group. Leuk Lymphoma. 1999;36:109–21.
145. Simard JF, Baecklund E, Kinch A, Brattstrom C, Ingvar A, Molin D, Adami J, Fernberg P, Wilczek H, Ekbom A, Smedby KE. Pediatric organ transplantation and risk of premalignant and malignant tumors in Sweden. Am J Transplant. 2011;11:146–51.
146. Smith JM, Dharnidharka VR. Viral surveillance and subclinical viral infection in pediatric kidney transplantation. Pediatr Nephrol. 2015;30:741–8.
147. Soriano-Lopez DP, Alcantar-Fierros JM, Hernandez-Plata JA, Gonzalez-Jorge AL, Velazquez-Ramos S, Flores-Hernandez MA, Fuentes V, Castaneda P, Nieto J, Sanchez JL, Lopez B, Valencia-Mayoral P, Varela-Fascinetto G. A scheduled program of molecular screening for Epstein-Barr virus decreases the incidence of post-transplantation lymphoproliferative disease in pediatric liver Transplantation. Transplant Proc. 2016;48:654–7.
148. Stevens SJ, Stevens C, Verschuuren EAM, Pronk I, van der Bij W, Harmsen MC, Hauw The T, Meijer CJLM, van den Brule AJC, Middeldorp JM. Frequent monitoring of Epstein-Barr virus DNA load in unfractionated whole blood is essential for early detection of posttransplant lymphoproliferative disease in high-risk patients. Blood. 2001;97:1165–71.
149. Styczynski J, Gil L, Tridello G, Ljungman P, Donnelly JP, van der Velden W, Omar H, Martino R, Halkes C, Faraci M, Theunissen K, Kalwak K, Hubacek P, Sica S, Nozzoli C, Fagioli F, Matthes S, Diaz MA, Migliavacca M, Balduzzi A, Tomaszewska A, Camara Rde L, van Biezen A, Hoek J, Iacobelli S, Einsele H, Cesaro S, Blood Infectious Diseases Working Party of the European Group for, and Transplantation Marrow. Response to rituximab-based therapy and risk factor analysis in Epstein Barr virus-related lymphoproliferative disorder after hematopoietic stem cell transplant in children and adults: a study from the infectious diseases working Party of the European Group for blood and marrow Transplantation. Clin Infect Dis. 2013;57:794–802.
150. Styczynski J, van der Velden W, Fox CP, Engelhard D, de la Camara R, Cordonnier C, Ljungman P. Management of Epstein-Barr Virus infections and post-transplant lymphoproliferative disorders in patients after allogeneic hematopoietic stem cell transplantation: Sixth European Conference on Infections in Leukemia (ECIL-6) guidelines. haematologica. 2016;101:803–11.
151. Sun Q, Burton R, Reddy V. Safety of allogeneic Epstein–Barr virus (EBV)-specific cytotoxic T lymphocytes for patients with refractory EBV-related lymphoma. Br J Haematol. 2002;118:799–808.
152. Svoboda J, Kotloff R, Tsai DE. Management of patients with posttransplant lymphoproliferative disorder: the role of rituximab. Transpl Int. 2006;19:259–69.
153. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, Advani R, Ghielmini M, Salles GA, Zelenetz AD. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127:2375–90.
154. Taylor E, Jones M, Hourigan MJ, Johnson DW, Gill DS, Isbel N, Hawley CM, Marlton P, Gandhi MK, Campbell SB. Cessation of immunosuppression during chemotherapy for post-transplant lymphoproliferative disorders in renal transplant patients. Nephrol Dial Transplant. 2015;30:1774–9.
155. Tischer J, Engel N, Fritsch S, Prevalsek D, Hubmann M, Schulz C, Zoellner AK, Bucklein V, Reibke R, Mumm F, Rieger CT, Hill W, Ledderose G, Stemmler HJ, Kohnke T, Jager G, Kolb HJ, Schmid C, Moosmann A, Hausmann A. Virus infection in HLA-haploidentical hematopoietic stem cell transplantation: incidence in the context of immune recovery in two different transplantation settings. Ann Hematol. 2015;94:1677–88.
156. Trappe R, Riess H, Babel N, Hummel M, Lehmkuhl H, Jonas S, Anagnostopoulos I, Papp-Vary M, Reinke P, Hetzer R, Dorken B, Oertel S. Salvage chemotherapy for refractory and relapsed posttransplant lymphoproliferative disorders (PTLD) after treatment with single-agent rituximab. Transplantation. 2007;83:912–8.
157. Trappe RU, Dierickx D, Zimmermann H, Morschhauser F, Mollee P, Zaucha JM, Dreyling MH, Duhrsen U, Reinke P, Verhoef G, Subklewe M, Huttmann A, Tousseyn T, Salles G, Kliem V, Hauser IA, Tarella C, Van Den Neste E, Gheysens O, Anagnostopoulos I, Leblond V, Riess H, Choquet S. Response to rituximab induction is a predictive marker in B-cell post-transplant lymphoproliferative disorder and allows successful stratification into rituximab or R-CHOP consolidation in an international, prospective, multicenter phase II trial. J Clin Oncol. 2017;35:536–43.
158. Trappe R, Oertel S, Leblond V, Mollee P, Sender M, Reinke P, Neuhaus R, Lehmkuhl H, Horst HA, Salles G. Sequential treatment with rituximab followed by CHOP chemotherapy in adult B-cell post-transplant lymphoproliferative disorder (PTLD): the prospective international multicentre phase 2 PTLD-1 trial. Lancet Oncol. 2012;13:196–206.
159. Trofe J, Buell JF, Beebe TM, Hanaway MJ, First MR, Alloway RR, Gross TG, Succop P, Woodle ES. Analysis of factors that influence survival with post-transplant lymphoproliferative disorder in renal transplant recipients: the Israel Penn International Transplant Tumor Registry experience. Am J Transplant. 2005;5:775–80.
160. Tsai DE, Douglas L, Andreadis C, Vogl DT, Arnoldi S, Kotloff R, Svoboda J, Bloom RD, Olthoff KM, Brozena SC. EBV PCR in the diagnosis and monitoring of posttransplant lymphoproliferative disorder: results of a two-arm prospective trial. Am J Transplant. 2008;8:1016–24.

161. Uhlin M, Wikell H, Sundin M, Blennow O, Maeurer M, Ringden O, Winiarski J, Ljungman P, Remberger M, Mattsson J. Risk factors for Epstein-Barr virus-related post-transplant lymphoproliferative disease after allogeneic hematopoietic stem cell transplantation. Haematologica. 2014;99:346–52.
162. Upton A, Diane H, Martin P, Raymond T, Dat T, Riccardo S, Derek S, Lori W, Samia W, Susan N. Utility of semiquantitative polymerase chain reaction for Epstein-Barr virus to measure virus load in pediatric organ transplant recipients with and without posttransplant lymphoproliferative disease. Clin Infect Dis. 2001;33:145–50.
163. Vali R, Punnett A, Bajno L, Moineddin R, Shammas A. The value of 18F-FDG PET in pediatric patients with post-transplant lymphoproliferative disorder at initial diagnosis. Pediatr Transplant. 2015;19:932–9.
164. van de Glind G, de Graaf S, Klein C, Cornelissen M, Maecker B, Loeffen J. Intrathecal rituximab treatment for pediatric post-transplant lymphoproliferative disorder of the central nervous system. Pediatr Blood Cancer. 2008;50:886–8.
165. Vase MØ, Maksten EF, Bendix K, Hamilton-Dutoit S, Andersen C, Møller MB, Sørensen SS, Jespersen B, Kampmann J, Søndergård E. Occurrence and prognostic relevance of CD30 expression in post-transplant lymphoproliferative disorders. Leuk Lymphoma. 2015;56:1677–85.
166. Vickers MA, Wilkie GM, Robinson N, Rivera N, Haque T, Crawford DH, Barry J, Fraser N, Turner DM, Robertson V. Establishment and operation of a Good Manufacturing Practice-compliant allogeneic Epstein-Barr virus (EBV)-specific cytotoxic cell bank for the treatment of EBV-associated lymphoproliferative disease. Br J Haematol. 2014;167:402–10.
167. Wagner HJ, Cheng YC, Huls MH, Gee AP, Kuehnle I, Krance RA, Brenner MK, Rooney CM, Heslop HE. Prompt versus preemptive intervention for EBV lymphoproliferative disease. Blood. 2004;103:3979–81.
168. Walker RC, Marshall WF, Strickler JG, Wiesner RH, Velosa JA, Habermann TM, McGregor CG, Paya CV. Pretransplantation assessment of the risk of lymphoproliferative disorder. Clin Infect Dis. 1995;20:1346–53.
169. Webber S, Harmon W, Faro A, Green M, Sarwal M, Hayashi R, Canter C, Thomas D, Jaffe R, Fine R. Anti-CD20 monoclonal antibody (rituximab) for refractory PTLD after pediatric solid Organ Transplantation: multicenter experience from a registry and from a prospective Clinical trial. Blood. 2004;104:746.
170. Webber SA, Naftel DC, Jay Fricker F, Olesnevich P, Blume ED, Addonizio L, Kirklin JK, Canter CE. Lymphoproliferative disorders after paediatric heart transplantation: a multi-institutional study. Lancet. 2006;367:233–9.
171. Webster A, Woodroffe RC, Taylor RS, Chapman JR, Craig JC. Tacrolimus versus cyclosporin as primary immunosuppression for kidney transplant recipients. Cochrane Database Syst Rev. 2005;4(4):CD003961.
172. Weintraub L, Weiner C, Miloh T, Tomaino J, Joashi U, Benchimol C, Strauchen J, Roth M, Wistinghausen B. Identifying predictive factors for posttransplant lymphoproliferative disease in pediatric solid organ transplant recipients with Epstein-Barr virus viremia. J Pediatr Hematol Oncol. 2014;36:e481–6.
173. Wiesmayr S, Webber SA, Macedo C, Popescu I, Smith L, Luce J, Metes D. Decreased NKp46 and NKG2D and elevated PD-1 are associated with altered NK-cell function in pediatric transplant patients with PTLD. Eur J Immunol. 2012;42:541–50.
174. Wilkie GM, Taylor C, Jones MM, Burns DM, Turner M, Kilpatrick D, Amlot PL, Crawford DH, Haque T. Establishment and characterization of a bank of cytotoxic T lymphocytes for immunotherapy of epstein-barr virus-associated diseases. J Immunother. 2004;27:309–16.
175. Wu M, Huang F, Jiang X, Fan Z, Zhou H, Liu C, Jiang Q, Zhang Y, Zhao K, Xuan L, Zhai X, Zhang F, Yin C, Sun J, Feng R, Liu Q. Herpesvirus-associated central nervous system diseases after allogeneic hematopoietic stem cell transplantation. PLoS One. 2013;8:e77805.
176. Xu LP, Zhang CL, Mo XD, Zhang XH, Chen H, Han W, Chen YH, Wang Y, Yan CH, Wang JZ, Wang FR, Zhao T, Liu YR, Liu KY, Huang XJ. Epstein-Barr virus-related post-transplantation lymphoproliferative disorder after unmanipulated human leukocyte antigen haploidentical hematopoietic stem cell transplantation: incidence, risk factors, treatment, and clinical outcomes. Biol Blood Marrow Transplant. 2015;21:2185–91.
177. Yamamoto N, Nishimura N, Takeuchi M, Ito T, Yokozaki H, Hirase S, Kubokawa I, Mori T, Yanai T, Hayakawa A. The emergence of CD20−/CD19− tumor cells after rituximab therapy for Epstein–Barr virus-associated post-transplant lymphoproliferative disorder complicated with hemophagocytic lymphohistiocytosis. Eur J Pediatr. 2014;173:1615–8.
178. Yanik EL, Shiels MS, Smith JM, Clarke CA, Lynch CF, Kahn AR, Koch L, Pawlish KS, Engels EA. Contribution of solid organ transplant recipients to the pediatric non-hodgkin lymphoma burden in the United States. Cancer. 2017;123:4663–71.
179. Zimmermann T, Hoppe-Lotichius M, Tripkovic V, Barreiros AP, Wehler TC, Zimmermann A, Schattenberg JM, Heise M, Biesterfeld S, Galle PR, Otto G, Schuchmann M. Liver transplanted patients with preoperative autoimmune hepatitis and immunological disorders are at increased risk for Post-Transplant Lymphoproliferative Disease (PTLD). Eur J Intern Med. 2010;21:208–15.

Pediatric-Type Follicular Lymphoma (PTFL)

16

Andishe Attarbaschi

Introduction

Pediatric-type follicular lymphoma (PTFL) is a rare subtype of Non-Hodgkin's lymphoma (NHL) in childhood and adolescence accounting for less than 2% of all cases [1–7]. However, it also occurs in young adults and may be sporadically seen in older individuals [3, 6, 8]. According to the recently updated 2016 *World Health Organization (WHO) Classification of Tumors of Hematopoietic and Lymphoid Tissues*, PTFL is recognized as a distinct histopathological and clinical entity (Table 16.1) among the group of follicular lymphomas (FL), that is clearly separated from other lymphomas with a follicular growth pattern in childhood, such as large B-cell lymphoma with *IRF4* rearrangement [3, 6]. This chapter will discuss diagnostic criteria required, differential diagnoses, clinical presentation, therapy, prognosis, and outcome of PTFL, which clearly contrast with the usual (non-pediatric-type) FL seen in adulthood representing the second most common subtype of NHL in that age group and usually considered as a chronic (incurable) disorder with an indolent disease course [9]. Nevertheless, optimal therapy of PTFL has not yet been defined, with some study groups still using intensive B-cell NHL-type chemotherapy according to stage of disease, others relying on cyclophosphamide, hydroxy-daunorubicine, vincristine, and prednisone (CHOP)-derived chemotherapy courses, and still others favoring a "watchful waiting" strategy after complete resection [5, 10–17]. Regardless of the type of therapy, cure rates exceed 95% [5, 10–18].

Of note, most reports published to date assessing clinical characteristics and outcome of FL in childhood and adolescence have not used the current definition of the 2016 *WHO Classification of Tumours of Hematopoietic and Lymphoid Tissues* of PTFL for inclusion of patients [3, 6].

Table 16.1 Primary diagnostic criteria for pediatric-type follicular lymphoma (PTFL)

Morphology	At least partial effacement of nodal architecture (required)
	Pure follicular proliferation (required)[a]
	Expansile follicles[b]
	Intermediate-sized, so-called, blastoid cells (not centrocytes)[b]
Immunohistochemistry	BCL6 positivity
	BCL2 negativity or weak positivity
	High proliferative fraction (>30%)
Genomics	No *BCL2*, *BCL6*, *IRF4*, or aberrant *IG* rearrangement
	No *BCL2* amplification
Clinical features	Nodal disease (required)
	Stage I or II disease (required)
	Patient age < 40 years[b]
	Marked male predominance

[a]The presence of any component of diffuse large B-cell lymphoma or advanced-stage disease excludes PTFL
[b]These are common features of PTFL, but not required for diagnosis

Pathology

Morphology

Pediatric-type follicular lymphoma derives from germinal center B-cells, and the lymph node architecture usually is either totally or sub-totally effaced by an entirely follicular growth pattern of expansile and/or serpiginous follicles (Fig. 16.1) [3, 6]. However, there often remains the rim of a normal lymph node seen in the peripheral parts of the resected sample indicating a "node-in-node" appearance. The tumor cells usually form a monotonous population of intermediate-sized blastoid cells, which are distinct from centrocytes and centroblasts. According to the 2016 *WHO*

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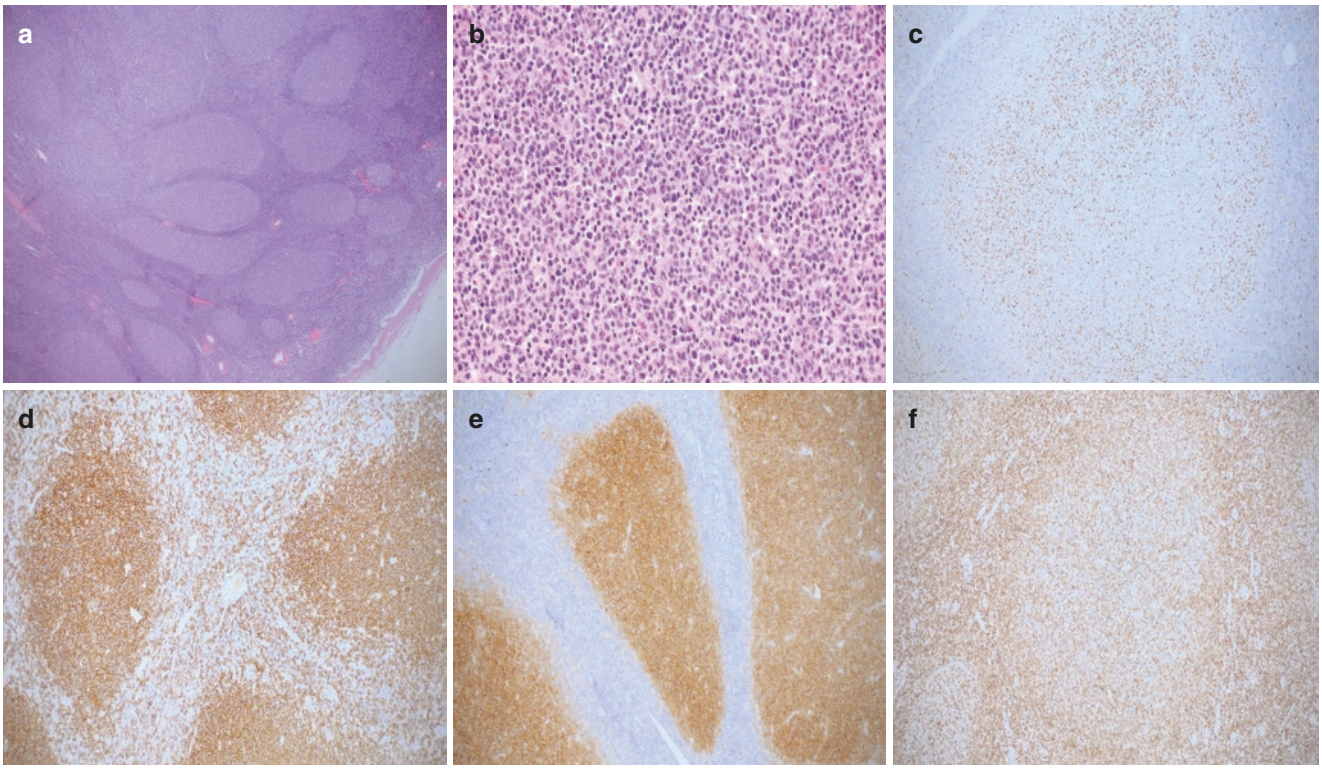

Fig. 16.1 Histopathological features (**a**, **b**), Ki-67 staining (**c**), and immunohistochemistry (**d**–**f**) of pediatric-type follicular lymphoma (PTFL). (**a**) Hematoxylin and eosin stain. (**b**) Hematoxylin and eosin stain. (**c**) Ki-67 staining. (**d**) CD20 expression. (**e**) CD10 expression. (**f**) Lack of BCL2 expression. (With kind permission by Prof. Dr. W. Klapper, Kiel, Germany)

Classification of Tumours of Hematopoietic and Lymphoid Tissues, typical histopathological grading (1, 2, 3a, and 3b) is usually not needed in PTFL if all other diagnostic criteria are met (Table 16.1) [3, 6]. Nevertheless, most cases of PTFL would fall into the group of grade 3 FL. As per definition, components of diffuse large B-cell lymphoma (DLBCL) preclude the diagnosis of PTFL [3, 6]. In addition, testicular FL is also excluded from the category of PTFL as it has several different morphological features [3, 6].

Immunohistochemistry

Pediatric-type follicular lymphoma is a mature B-cell neoplasm with the neoplastic cells consistently positive for CD20, CD79a, and PAX5 (Fig. 16.1) [3, 6]. Moreover, CD10 and BCL6 are usually expressed by the cells, whereas BCL2 is almost always negative (Fig. 16.1) [3, 6]. Expression of MUM/IRF4 should be negative, whereas its expression should rather raise the possibility of large B-cell lymphoma with *IRF4* rearrangement [3, 6, 19]. The neoplastic cells of PTFL usually demonstrate a moderate to high proliferation rate as revealed by Ki-67 staining (>30% of follicular cells).

Genetics

Whereas 85% of cases of usual FL in adulthood are associated with the translocation t(14;18)(q32;q21), resulting in the *IGH/BCL2* gene fusion, this chromosomal translocation is absent in PTFL [3, 6, 9]. According to the 2016 *WHO Classification of Tumours of Hematopoietic and Lymphoid Tissues*, PTFL does not carry rearrangements of *BCL2*, *BCL6*, *IRF4,* and *IG* loci, nor does it show amplifications of *BCL2* (Table 16.1) [3, 6]. Although a particular recurrent genetic aberration seems to be missing in PTFL, abnormalities involving the following chromosomal regions and genes have been observed in a subset of cases: gains or amplifications of 6pter-p24.3, deletions of 1p36, deletions and mutations of *TNFRSF14,* and mutations of *MAP2K1* [3, 6, 20, 21].

Differential Diagnoses

Reactive follicular hyperplasia, large B-cell lymphoma with *IRF4* rearrangement, marginal zone lymphoma as well as nodular lymphocyte-predominant or nodular sclerosis classical Hodgkin's lymphoma represent the most impor-

tant differential diagnoses of FL in childhood and adolescence [3, 6, 22].

According to the 2016 *WHO Classification of Tumours of Hematopoietic and Lymphoid Tissues*, large B-cell lymphoma with *IRF4* rearrangement is a distinct entity of mature large B-cell lymphomas, even when it displays a purely follicular growth pattern [3, 6, 19, 23, 24]. This very rare subtype of large B-cell lymphoma derives from germinal center B-cells with *IRF4* rearrangement, resulting in IRF4/MUM1 expression [24]. The growth pattern of the monotonous population of medium-sized to large neoplastic cells can be entirely diffuse, follicular and diffuse, or entirely follicular. The tumor cells are usually positive for CD20, CD79a, and PAX5 and, as per definition, strongly positive for IRF4/MUM1. Moreover, CD10, BCL6, and BCL2 expression are also seen in large B-cell lymphoma with *IRF4* rearrangement. In rare cases of patients fulfilling the pathological diagnostic criteria required, but lacking an *IRF4* translocation, *BCL6* rearrangements may be detected [3, 6, 19]. Clinical features are very similar to PTFL with most of the patients being children, adolescents, or young adults, showing an equal sex distribution (in contrast to PTFL), mainly presenting with enlarged lymph nodes in the head and neck region or involvement of the Waldeyer's ring (tonsils) and having a good prognosis after chemotherapy [24]. Involvement of the gastrointestinal tract has also been occasionally reported [24].

Etiology

The etiology of PTFL remains obscure. Most importantly, in contrast to other rare subtypes of NHL in childhood and adolescence – such as marginal zone lymphoma, primary central nervous system lymphoma, or peripheral T-cell lymphoma – PTFL is not particularly or more often seen in patients with pre-existing disorders, such as cancer predisposition syndromes or autoimmune conditions [11, 25–27].

Clinical Presentation

Extensive data on clinical features at initial diagnosis of PTFL are scarce. In principle, most cases of PTFL seem to involve the lymph nodes of the head and neck region, to present with stage I disease and to lack B-symptoms, such as fever, night sweats, and/or weight loss [1, 5, 10, 11, 13].

In order to get more insight into this rare disease and develop uniform therapy recommendations, two of the largest consortia in childhood NHL, the European Intergroup for Childhood NHL (EICNHL) and the international Berlin-Frankfurt-Münster (i-BFM) Group, recently performed a retrospective multinational study assessing the clinical characteristics and outcome of 63 pediatric patients with FL, albeit not using the 2016 *WHO Classification of Tumours of Hematopoietic and Lymphoid Tissues* of PTFL [3, 6, 11].

This study showed that FL in childhood and adolescence is usually associated with male gender (3:1), adolescent age (40% 10–15 years, 32% ≥15 years old), low serum LDH levels (<500 U/l in 75%), and limited disease (87% with stage I/II disease), mostly involving peripheral lymph nodes (nodal stage I/II disease being diagnostically required in the current definition of PTFL) [11]. However, as this study also identified stage III/IV patients (excluded from the current definition of PTFL), authors concluded that the initial diagnostic work-up should continue to follow the modified St. Jude staging system [28, 29]. Due to its rarity, only few case reports and series on FL have been published so far, with patient numbers ranging from 4 to 25 and also including patients with testicular FL or advanced-stage disease of whom none would have been classified as PTFL according to the 2016 *WHO Classification of Tumours of Hematopoietic and Lymphoid Tissues* [1, 3, 5, 6, 10, 12, 14, 15, 17]. Nevertheless, most of the reports demonstrated similar findings concerning the initial clinical features of FL as found in the EICNHL/i-BFM study [1, 5, 10–12, 14, 15, 17]. Details on patient characteristics and sites of involvement in the cohort of 63 patients are summarized in Table 16.2, showing that 50/63 patients (79%) had peripheral lymph node involvement only. Histopathological grading was available for 48/63 patients (76%), demonstrating grade 1 or 2 morphology in 6/48 (12.5%) and grade 3 morphology in 42/48 patients (87.5%). Nine/42 patients (21%) with grade 3 FL had areas of DLBCL (excluded from the current definition of PTFL).

Therapy, Prognosis, and Outcome

The prognosis and outcome of PTFL seems to be excellent with survival rates exceeding 95% [5, 10–12, 14, 15, 17, 18]. According to the EICNHL/i-BFM study, 44/63 FL patients (70%) received any polychemotherapy and 1 (2%) rituximab only, while 17 (26%) underwent a "watch-and-wait" strategy (all with initial complete resection) (Table 16.2) [11]. In one patient (2%), type of therapy received could not be ascertained. Of the 39/44 patients with available information, all but three patients received low- or intermediate-risk B-cell NHL-type therapy (Table 16.2). Only one/63 patients (2%) relapsed (after "watch and wait") and none of the patients died from the disease itself or therapy-related toxicity. The 2-year event-free and overall survival rates were 94 ± 5% and 100% (Fig. 16.2), respectively, after a median follow-up of 2.2 years.

Table 16.2 Clinical, laboratory, treatment characteristics, and outcome of the 63 patients with pediatric follicular lymphoma

Variable	No. of pts.	Variable	No. of pts.
Gender		Resection status	
Male	47 (75%)	Incomplete/biopsy	26 (41%)
Female	16 (25%)	Complete	32 (51%)
		n.a.	5 (8%)
Age (y)			
Median	13.0	Treatment	
Range	1.4–17.7	Chemotherapy[e]	44 (70%)
<10	18 (28%)	Rituximab only	1 (2%)
≥10–15	25 (40%)	"watch and wait"	17 (26%)
≥15	20 (32%)	n.a.	1 (2%)
sLDH level (U/l)		Complete resection	
Median	252	"watch and wait"	17 (53%)
Range	93–550	Chemotherapy	15 (47%)
<500	47 (75%)		
≥500	5 (8%)	Resection acc. to stage	
n.a.	11 (17%)	Stage I	36
		Stage I-R	30 (83%)
Stage of disease		Stage I-NR	4 (11%)
Stage I	36 (57%)	Stage I-n.a.	2 (6%)
Stage II	19 (30%)		
Stage III	6 (10%)	Stage II	19
Stage IV	2 (3%)	Stage II-R	2 (10%)
		Stage II-NR	14 (74%)
Histological grading		Stage II-n.a.	3 (16%)
Grade 1	4 (6%)		
Grade 2	1 (2%)	Stage III/IV-NR	8 (100%)
Grade 3[a]	27 (43%)		
Grade 1 + 2	1 (2%)	Radiotherapy	
Grade 1 + 3a	1 (2%)	Yes	1 (2%)
Grade 1 + 2 + 3a + MZL	1 (2%)	No	61 (96%)
Grade 2 + 3a	2 (3%)	n.a.	1 (2%)
Grade 3 + DLBCL[b]	9 (14%)		
Grade 3a + MZL	2 (3%)	Outcome	
n.a.	15 (24%)	Relapse	1 (2%)
		Death	0
Sites of involvement[c]		2-year EFS	94 ± 5%
Peripheral lymph nodes[d]	50 (79%)	2-year OS	100%
Head and neck (extranodal)	1 (2%)		
Tonsils	4 (6%)	Follow-up (y)	
Ear-nose-throat	4 (6%)	Median	2,2
Mediastinum	0	Range	0.2–8.7
Abdomen	9 (14%)		
Bone marrow	2 (3%)	Lost to follow-up	1 (2%)[f]
Central nervous system	0		
Testis	2 (3%)		
Skin	1 (2%)		
Bone	1 (2%)		

Abbreviations: No. of pts. number of patients, *y* years, *sLDH* serum lactate dehydrogenase, *n.a*. not available, *MZL* marginal zone lymphoma, *DLBCL* diffuse large B-cell lymphoma, *acc.* according, *R* complete resection, *NR* no complete resection, *CCR* complete continuous remission, *EFS* event-free survival, *OS* overall survival

[a]13/27 with grade 3a, 10/27 with grade 3b, and 3/27 patients with no information on the 3a/3b variant

[b]3/9 with grade 3a and 6/9 patients with grade 3b morphology

[c]27/63 patients suffered from stage II, III, or IV disease and thus had >1 site of involvement

[d]Corresponding to cervical (submandibular), supra- and infraclavicular, pre- and retro-auricular, nuchal, parotical, axillary and inguinal lymph node regions

[e]According to protocols of the NHL-BFM ($n = 27$), AIEOP ($n = 3$), LMB ($n = 2$), JACLS ($n = 5$) and UKCCSG ($n = 1$) studies; CHOP ($n = 5$), CVP ($n = 1$)

[f]This patient was lost to follow-up immediately after the primary operation

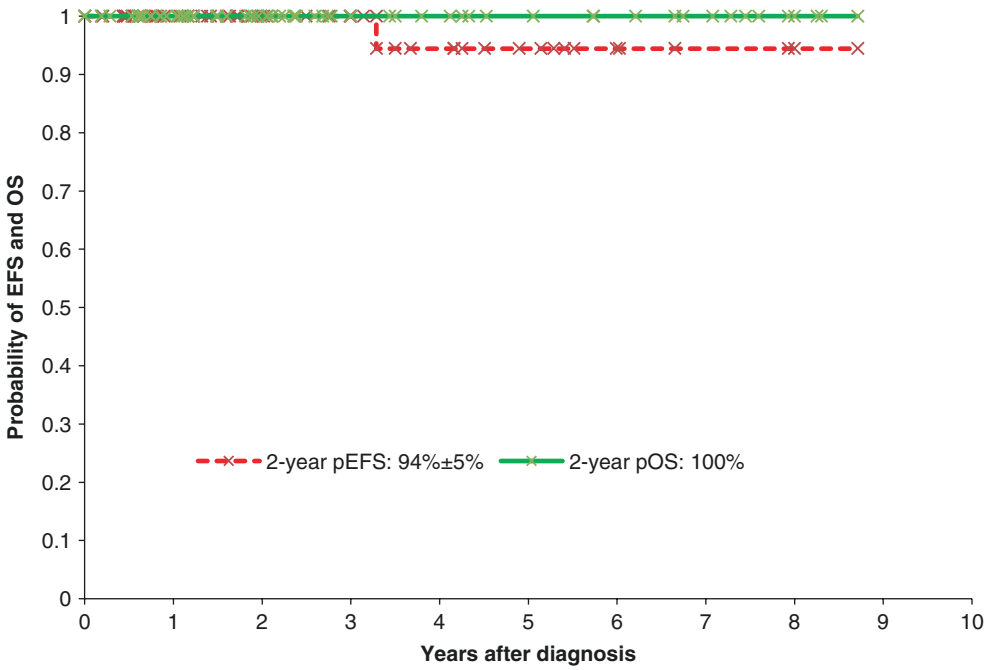

Fig. 16.2 Two-year event-free and overall survival of the 63 patients with pediatric follicular lymphoma. *Abbreviations:* pEFS, probability of event-free survival; pOS, probability of overall survival

In contrast to FL in adults, which is usually of low-grade morphology and an incurable disease albeit diverse treatment approaches, PTFL seems to have a very good outcome after limited chemotherapy or complete resection followed by a "watch-and-wait" strategy [5, 11, 16, 30]. Importantly, neither higher histological grading nor initial components of DLBCL seem to be associated with an unfavorable prognosis [11]. Within the EICNHL/i-BFM study, of the 32 patients with initial complete resection (including 30/36 stage I patients), 17 (53%) children had no further treatment with only 1 relapse (local), suggesting no systemic disease in localized FL [11]. The excellent overall outcome of the EICNHL/i-BFM cohort of FL patients is comparable to the results published in the literature showing that pediatric stage-adapted B-cell NHL-type chemotherapy and CHOP-like cycles ± rituximab are effective in (in)completely resectable disease [1, 5, 11, 12, 14, 15, 17, 31]. However, the exact role of complete resection and observation has not been validated yet. Thus, future clinical trials should aim to establish the least amount of effective (chemo)therapy necessary for the cure of PTFL. As almost all cycles of chemotherapy used for pediatric B-cell NHL still include anthracyclines, alkylating agents, and intrathecal therapy, low-intensity chemotherapy for PTFL should ideally be free of the latter compounds usually carrying the risk for acute- and long-term morbidity [32–35]. A study in pediatric early-stage nodular lymphocyte predominant Hodgkin's lymphoma may serve as a paradigm, as it has shown that low-intensity chemotherapy is successful in non-completely resectable disease, while more toxic therapy courses used for classical Hodgkin's lymphoma can be reserved for relapse [36].

Conclusions

In conclusion, regardless of the therapy the patients with PTFL received, it seems that PTFL does not automatically require chemotherapy due to the excellent outcome. Based on the data gained from the EICNHL/i-BFM study on FL, one might infer that in case of complete resection in carefully evaluated stage I patients a "watchful waiting" strategy is possible [11]. However, it should be emphasized that patients may only be candidates for complete surgical resection if the operation can be performed easily and safely, and most importantly, without any mutilation. In all other patients, initial surgery should include the least invasive procedure to establish the diagnosis, to be then followed by limited chemotherapy. Given the difficulties in differentiating PTFL from follicular hyperplasia, evaluation by an experienced hematopathologist is highly recommended before starting any therapy [3, 6, 22]. As children with non-resectable FL might have an excellent outcome with multidrug chemotherapy, which is, however, associated with acute- and long-term toxicity, multinational controlled trials have yet to be performed. These should take into account the current definition of the 2016 *WHO Classification of Tumours of Hematopoietic and Lymphoid Tissues* of PTFL in order to clearly establish, not only that no chemotherapy is a safe approach in stage I patients with complete resection, but also that low-intensity chemotherapy ± monoclonal antibodies is sufficient for patients with non-completely resectable disease and for pediatric patients with FL, who do not fulfill the diagnostic criteria of PTFL, such as children with rearrangements involving *BCL2*, *BCL6*, *IRF4*, or *IG* loci [11, 13, 31, 33, 37–39].

References

1. Agrawal R, Wang J. Pediatric follicular lymphoma: a rare clinicopathologic entity. Arch Pathol Lab Med. 2009;133(1):142–6.
2. Anderson JR, Armitage JO, Weisenburger DD. Epidemiology of the non-Hodgkin's lymphomas: distributions of the major subtypes differ by geographic locations. Non-Hodgkin's Lymphoma Classification Project. Ann Oncol. 1998;9(7):717–20.
3. Ferry JA, de Leval L, Louissaint A Jr, Harris NL. Follicular lymphoma. In: Jaffe ES, Arber DA, Campo E, Harris NL, Quintanilla-Martinez L, editors. Hematopathology. Philadelphia: Elsevier; 2017. p. p321–52.
4. Kansal R, Singleton TP, Ross CW, Finn WG, Padmore RF, Schnitzer B. Follicular hodgkin lymphoma: a histopathologic study. Am J Clin Pathol. 2002 Jan;117(1):29–35.
5. Oschlies I, Salaverria I, Mahn F, Meinhardt A, Zimmermann M, Woessmann W, et al. Pediatric follicular lymphoma—a clinicopathological study of a population-based series of patients treated within the Non-Hodgkin's Lymphoma—Berlin-Frankfurt-Munster (NHL-BFM) multicenter trials. Haematologica. 2010;95(2):253–9.
6. Jaffe ES, Harris NL, Siebert R. Paediatric-type follicular lymphoma. In: Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, editors. WHO Classification of Tumours of Haemtopoietic and Lymphoid Tissues. Lyon: International Agency for Research on Cancer (IARC); 2017. p. 278–9.
7. Liu Q, Salaverria I, Pittaluga S, Jegalian AG, Xi L, Siebert R, et al. Follicular lymphomas in children and young adults: a comparison of the pediatric variant with usual follicular lymphoma. Am J Surg Pathol. 2013;37(3):333–43.
8. Louissaint A Jr, Ackerman AM, Dias-Santagata D, Ferry JA, Hochberg EP, Huang MS, et al. Pediatric-type nodal follicular lymphoma: an indolent clonal proliferation in children and adults with high proliferation index and no BCL2 rearrangement. Blood. 2012;120(12):2395–404.
9. Rohatiner AZ, Lister TA. The clinical course of follicular lymphoma. Best Pract Res Clin Haematol. 2005;18(1):1–10.
10. Atra A, Meller ST, Stevens RS, Hobson R, Grundy R, Carter RL, et al. Conservative management of follicular non-Hodgkin's lymphoma in childhood. Br J Haematol. 1998;103(1):220–3.
11. Attarbaschi A, Beishuizen A, Mann G, Rosolen A, Mori T, Uyttebroeck A, et al. Children and adolescents with follicular lymphoma have an excellent prognosis with either limited chemotherapy or with a "Watch and wait" strategy after complete resection. Ann Hematol. 2013;92(11):1537–41.
12. Finn LS, Viswanatha DS, Belasco JB, Snyder H, Huebner D, Sorbara L, et al. Primary follicular lymphoma of the testis in childhood. Cancer. 1999;85(7):1626–35.
13. Kumar R, Galardy PJ, Dogan A, Rodriguez V, Khan SP. Rituximab in combination with multiagent chemotherapy for pediatric follicular lymphoma. Pediatr Blood Cancer. 2011;57(2):317–20.
14. Lones MA, Raphael M, McCarthy K, Wotherspoon A, Terrier-Lacombe MJ, Ramsay AD, et al. Primary follicular lymphoma of the testis in children and adolescents. J Pediatr Hematol Oncol. 2012;34(1):68–71.
15. Lorsbach RB, Shay-Seymore D, Moore J, Banks PM, Hasserjian RP, Sandlund JT, et al. Clinicopathologic analysis of follicular lymphoma occurring in children. Blood. 2002;99(6):1959–64.
16. McNamara C, Davies J, Dyer M, Hoskin P, Illidge T, Lyttelton M, et al. Guidelines on the investigation and management of follicular lymphoma. Br J Haematol. 2012;156(4):446–67.
17. Pinto A, Hutchison RE, Grant LH, Trevenen CL, Berard CW. Follicular lymphomas in pediatric patients. Mod Pathol. 1990;3(3):308–13.
18. Sorge C, Costa LJ, Taub JW, Xavier AC. Incidence and outcomes of rare paediatric non-hodgkin lymphomas. Br J Haematol. 2018.
19. Pittaluga S, Harris NL, Siebert R, Salaverria I. Large B-cell lymphoma with *IRF4* rearrangment. In: Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, editors. WHO Classification of Tumours of Haemtopoietic and Lymphoid Tissues. Lyon: International Agency for Research on Cancer (IARC); 2017. p. 280–1.
20. Louissaint A Jr, Schafernak KT, Geyer JT, Kovach AE, Ghandi M, Gratzinger D, et al. Pediatric-type nodal follicular lymphoma: a biologically distinct lymphoma with frequent MAPK pathway mutations. Blood. 2016;128(8):1093–100.
21. Martin-Guerrero I, Salaverria I, Burkhardt B, Szczepanowski M, Baudis M, Bens S, et al. Recurrent loss of heterozygosity in 1p36 associated with TNFRSF14 mutations in IRF4 translocation negative pediatric follicular lymphomas. Haematologica. 2013;98(8):1237–41.
22. Kussick SJ, Kalnoski M, Braziel RM, Wood BL. Prominent clonal B-cell populations identified by flow cytometry in histologically reactive lymphoid proliferations. Am J Clin Pathol. 2004;121(4):464–72.
23. Salaverria I, Philipp C, Oschlies I, Kohler CW, Kreuz M, Szczepanowski M, et al. Translocations activating IRF4 identify a subtype of germinal center-derived B-cell lymphoma affecting predominantly children and young adults. Blood. 2011;118(1):139–47.
24. Quintanilla-Martinez L, Sander B, Chan JK, Xerri L, Ott G, Campo E, et al. Indolent lymphomas in the pediatric population: follicular lymphoma, IRF4/MUM1+ lymphoma, nodal marginal zone lymphoma and chronic lymphocytic leukemia. Virchows Arch. 2016;468(2):141–57.
25. Mellgren K, Attarbaschi A, Abla O, Alexander S, Bomken S, Bubanska E, et al. Non-anaplastic peripheral T cell lymphoma in children and adolescents-an international review of 143 cases. Ann Hematol. 2016;95(8):1295–305.
26. Ronceray L, Abla O, Barzilai-Birenboim S, Bomken S, Chiang AK, Jazbec J, et al. Children and adolescents with marginal zone lymphoma have an excellent prognosis with limited chemotherapy or a watch-and-wait strategy after complete resection. Pediatr Blood Cancer. 2018;65(4).
27. Thorer H, Zimmermann M, Makarova O, Oschlies I, Klapper W, Lang P, et al. Primary central nervous system lymphoma in children and adolescents: low relapse rate after treatment according to Non-Hodgkin-Lymphoma Berlin-Frankfurt-Munster protocols for systemic lymphoma. Haematologica. 2014;99(11):e238–41.
28. Murphy SB, Fairclough DL, Hutchison RE, Berard CW. Non-Hodgkin's lymphomas of childhood: an analysis of the histology, staging, and response to treatment of 338 cases at a single institution. J Clin Oncol. 1989;7(2):186–93.
29. Rosolen A, Perkins SL, Pinkerton CR, Guillerman RP, Sandlund JT, Patte C, et al. Revised International Pediatric Non-Hodgkin Lymphoma Staging System. J Clin Oncol. 2015;33(18):2112–8.
30. Kridel R, Sehn LH, Gascoyne RD. Pathogenesis of follicular lymphoma. J Clin Invest. 2012;122(10):3424–31.
31. Perkins SL, Gross TG. Pediatric indolent lymphoma--would less be better? Pediatr Blood Cancer. 2011;57(2):189–90.
32. Gerrard M, Cairo MS, Weston C, Auperin A, Pinkerton R, Lambilliote A, et al. Excellent survival following two courses of COPAD chemotherapy in children and adolescents with resected localized B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study. Br J Haematol. 2008;141(6):840–7.
33. Goldman S, Smith L, Anderson JR, Perkins S, Harrison L, Geyer MB, et al. Rituximab and FAB/LMB 96 chemotherapy in children with Stage III/IV B-cell non-Hodgkin lymphoma: a Children's Oncology Group report. Leukemia. 2013;27(5):1174–7.
34. Pillon M, Di Tullio MT, Garaventa A, Cesaro S, Putti MC, Favre C, et al. Long-term results of the first Italian Association of Pediatric

Hematology and Oncology protocol for the treatment of pediatric B-cell non-Hodgkin lymphoma (AIEOP LNH92). Cancer. 2004;101(2):385–94.
35. Woessmann W, Seidemann K, Mann G, Zimmermann M, Burkhardt B, Oschlies I, et al. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. Blood. 2005;105(3):948–58.
36. Shankar A, Hall GW, Gorde-Grosjean S, Hasenclever D, Leblanc T, Hayward J, et al. Treatment outcome after low intensity chemotherapy [CVP] in children and adolescents with early stage nodular lymphocyte predominant Hodgkin's lymphoma - an Anglo-French collaborative report. Eur J Cancer. 2012;48(11):1700–6.
37. Marcus R, Imrie K, Belch A, Cunningham D, Flores E, Catalano J, et al. CVP chemotherapy plus rituximab compared with CVP as first-line treatment for advanced follicular lymphoma. Blood. 2005;105(4):1417–23.
38. Marcus R, Imrie K, Solal-Celigny P, Catalano JV, Dmoszynska A, Raposo JC, et al. Phase III study of R-CVP compared with cyclophosphamide, vincristine, and prednisone alone in patients with previously untreated advanced follicular lymphoma. J Clin Oncol. 2008;26(28):4579–86.
39. Meinhardt A, Burkhardt B, Zimmermann M, Borkhardt A, Kontny U, Klingebiel T, et al. Phase II window study on rituximab in newly diagnosed pediatric mature B-cell non-Hodgkin's lymphoma and Burkitt leukemia. J Clin Oncol. 2010;28(19):3115–21.

Marginal Zone Lymphoma

17

Birgit Burkhardt

Introduction

Marginal zone lymphoma (MZL) is a mature B-cell lymphoma. The classification of lymphoid tumors of the World Health Organization (WHO) differentiates nodal MZL (nMZL), extranodal MZL (enMZL), and splenic MZL [1, 2]. Extranodal and nodal marginal zone lymphoma is the third most common type of B-cell lymphoma in adults and accounts for 5–17% of NHL [1, 3]. Conversely, this lymphoma subtype is rare in children and adolescents. Data on clinical features, treatment, and outcome in pediatric MZL are limited with few reports on small patient series and several case reports published to date [4–18]. In contrast to more common aggressive mature B-cell lymphoma e.g. Burkitt lymphoma and diffuse large B-cell lymphoma (DLBCL), MZL present with slow kinetics of disease progression. Individual courses with waxing and waning of symptoms are not uncommon.

Interestingly, in adults extranodal MZL is clearly more common than nodal MZL. For the pediatric and adolescent population, the number of published cases is too small to finally conclude on the distribution of nodal versus extranodal MZL. Splenic MZL are extremely rare in children. Since the introduction of pediatric MZL in the WHO classification the incidence and/or the frequency of MZL diagnosis and reporting in children and adolescents is increasing.

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Epidemiology, Clinical Presentation, and Staging

MZL represent less than 2% of all NHL in children and adolescents with a clear male predominance. MZL are more commonly diagnosed in adolescents than in younger children [4, 5, 18]. Most patients present with limited or no clinical symptoms, normal serum lactate dehydrogenase (LDH) level and limited stages of disease. Nevertheless, individual patients with advanced and disseminated stage disease are reported, emphasizing the need for complete staging.

Nodal MZL

Pediatric nodal MZL commonly present with localized nodal lymphoma. Given their specific clinicopathological feature, "pediatric nodal MZL" were introduced into the WHO classification of lymphoid tumors in 2008 [1, 2]. Nodal MZL predominantly involve lymph nodes of the head and neck region with limited stage I or stage II disease in the vast majority of patients [4, 5, 9]. The diagnosis of bone marrow involvement remains challenging, and robust data on bone marrow involvement of pediatric nMZL are lacking [19]. The median age of the patients is about 15 years and there is a notable male predominance in nMZL exceeding the male predominance in Burkitt lymphoma. The serum LDH level, which represents a widely accepted surrogate for tumor cell load in aggressive B-NHL, is normal or slightly elevated in mainly all nMZL patients.

Extranodal MZL

Extranodal marginal zone lymphoma (enMZL) are predominant among adults and were reported in about 30–60% of pediatric and adolescent MZL cases [4, 5, 13, 18]. Typical sites of enMZL in children and adolescents are the

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ear-nose-throat area, salivary glands, digestive tract, lungs, spleen, and conjunctiva or ocular adnexal [20, 21] often involving sites of acquired mucosa-associated tissues. In addition, manifestations in the skin [6, 14, 22, 23], orbita, breast, kidney, spleen, and central nervous system are described. Occasional involvement of the bone marrow and peripheral blood is reported [24–26]. Since the cells of MZL are difficult to recognize in bone marrow smears by cytomorphology, flow cytometry analysis of peripheral blood and bone marrow is advisable. Furthermore, at suspicion of bone marrow involvement, bone marrow biopsy in addition to bone marrow aspirates for histopathological analysis is indicated. Nevertheless, bone marrow infiltration by MZL in pediatric cases is rare, and only individual cases are reported [4, 5].

The median age of pediatric enMZL is between 10 and 15 years. The male to female ratio is 2:1. An association of extranodal MZL of mucosa-associated lymphoid tissue (MALT lymphoma) with autoimmune system disorders or immune system dysregulation as a result of stimulation by chronic infection is well described [27–30]. For example, the incidence of MZL of salivary glands in patients with Sjögren syndrome is 40-fold higher than in the entire population [7, 18, 28, 31]. Specific microorganisms have been considered to be associated with MALT lymphoma, such as *Helicobacter pylori* in the stomach, *Chlamydia psittaci* in the ocular adnexa, *Campylobacter jejuni* in the small intestine, and *Borrelia burgdorferi* in the skin [18, 29, 30, 32, 33]. In addition, most recently Kluin et al. reported a novel type of nodal marginal zone hyperplasia, which is stimulated by *Haemophilus influenzae* [34]. Other pre-existing diseases described in patients with enMZL are Crigler-Najjar syndrome, Hodgkin lymphoma [5], immunosuppression after organ transplantation [35, 36], common variable immunodeficiency [4, 37], systemic lupus erythematosus [18], squamous papilloma, and hyperandrogenism with hirsutism [4].

Pathology

Pediatric nodal marginal zone lymphoma are characterized by obliteration of the sinuses and partial to total architectural effacement by marginal zone expansion and disruption of reactive follicles by neoplastic cells [18, 38]. The polymorphic population of small- to medium-sized atypical cells shows predominantly interfollicular distribution with marked expansion of the marginal zones (Fig. 17.1). There is no defining immunophenotype. MZL are positive for CD20 and often for CD43 [13, 18]. Germinal center markers including CD10 are often negative. BCL2 is variably expressed. Light chain restriction and evidence of clonality are characteristics for nMZL. Cytogenetic abnormalities are reported in approximately 20% of pediatric nMZL with trisomy 18 being the most frequent one and rare cases with trisomies 3 and 18 [13]. Translocations involving *MYC*, *BCL2*, *BCL6,* and *IRF4* are usually absent. A recent study of six cases analyzed by whole exome sequencing reported a low mutational burden and failed to identify recurrent mutations [10, 13].

Extranodal MZL show a dense lymphoid infiltrate composed of broad sheets of monocytoid cells and or centrocyte-like cells with glandular destruction and architectural distortion [18]. Extension into adjacent soft tissue structures is seen. As in nodal MZL, residual reactive follicles are frequent. For enMZL of the skin dense dermal perivascular and periadnexal lymphoid infiltrates are described with extension into the subcutaneous tissue but sparing of the epidermis. The immunohistochemical findings are similar in nMZL and enMZL. Histologically enMZL in children and adolescents are reported indistinguishable from enMZL in older patients [18]. Genetic abnormalities are identified at a slightly lower incidence in extranodal marginal zone lymphoma in children, adolescents, and young adults compared to nMZL [13]. For adult patient series histological transformation from enMZL to diffuse large B-cell lymphoma is described [39]. Analogue reports for pediatric and adolescent are lacking so far.

Differential Diagnosis

Pediatric as well as adult MZL are characterized by a neoplastic proliferation of B-cells with a marginal zone phenotype. In adults, MZL mainly has to be distinguished from non-neoplastic inflammatory reactions and other subtypes of indolent B-cell lymphoma such as small lymphocytic lymphoma and mantle cell lymphoma. In children, the main differential diagnosis of MZL are pediatric-type follicular lymphoma and reactive inflammatory conditions like progressively transformed germinal centers and reactive expansions of the marginal zone. Reactive but atypical marginal zone hyperplasia with light chain restriction has been described in younger patients arising in mucosa-associated lymphatic tissues and lymph nodes, which comprises a potentially non-neoplastic mimicker of lymphoma [18, 34, 40]. Immunosuppression after solid organ transplantation might predispose pediatric patients to atypical marginal zone hyperplasia [41]. It is reported that lymphoid hyperplasia in those patients can present with a self-limited course in the absence of treatment or reduction of immunosuppression. Non-neoplastic differential diagnosis is distinguishable from MZL by the lack of clonal rearrangement of the immunoglobulin genes in combination with histological and immunophenotypical features. Expansion of the interfollicular region and destruction of follicular structures, at least focally, are features that favor nMZL over a reactive processes.

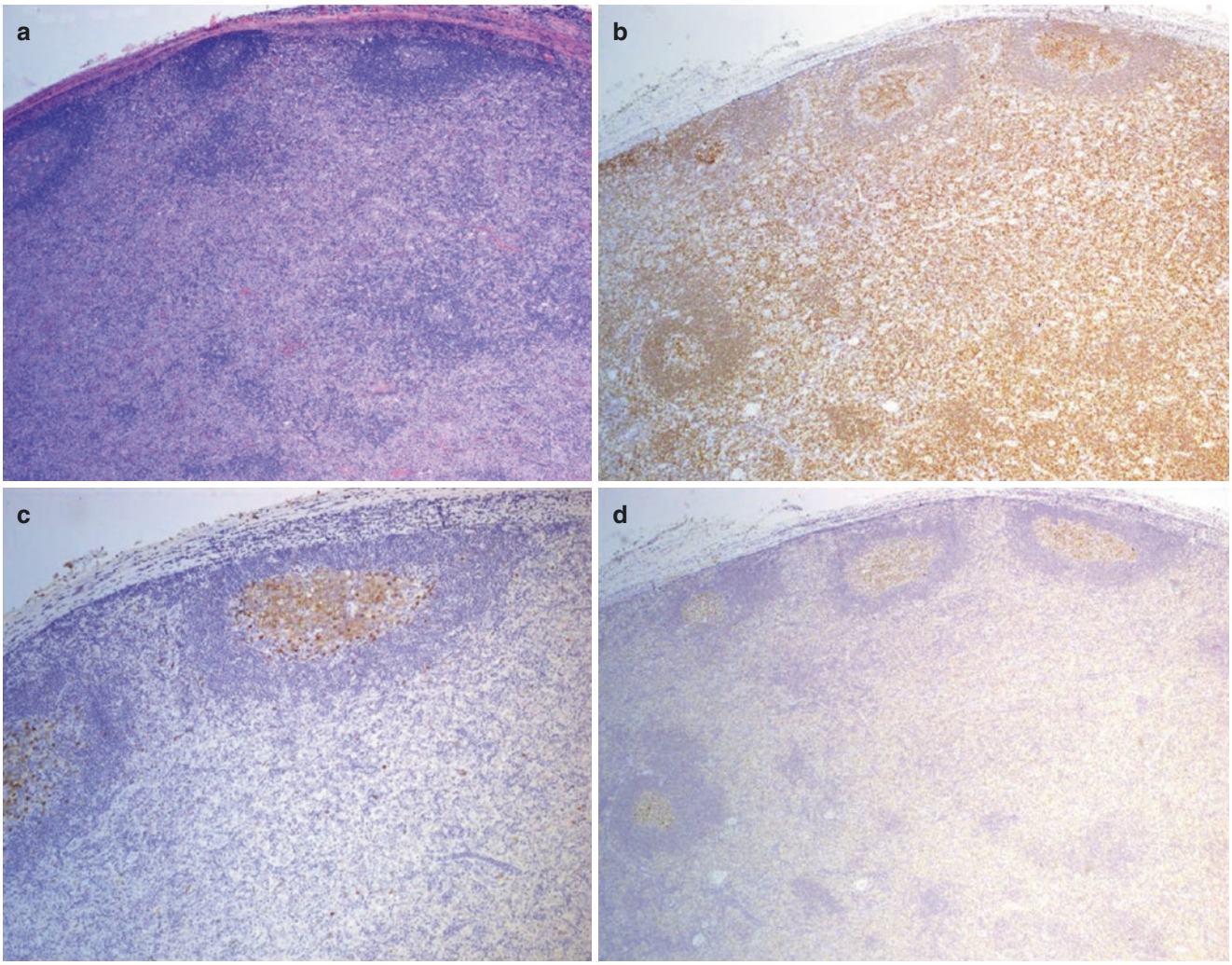

Fig. 17.1 Pediatric nodal marginal zone lymphoma. Lymph node with an interfollicular infiltration with neoplastic cells (**a**, Giemsa) with the expansion of the B-cell areas over the follicles into the interfollicular spaces (**b**, CD20). The infiltrating cells stain negative for germinal center markers CD10 (**c**, CD10) and BCL6 (**d**, BCL6)

Staging

Initial staging for nMZL and enMZL is performed according to the St. Jude staging system and since 2015 according to the Revised International Pediatric Non-Hodgkin Lymphoma Staging System [42, 43]. Current recommendation for staging examinations included medical history, physical assessment, full blood count with peripheral blood smears, total serum lactate dehydrogenase (LDH), bone marrow aspirates and biopsies, cerebrospinal fluid (CSF) cytology, chest x-ray and ultrasonography of lymph nodes, abdomen and testes as well as magnetic resonance imaging (MRI), or computed tomography (CT) scan of involved sites. Advanced disease stages (≥ stage III) are observed in about one third of patients. Bone marrow involvement is rare but reported in adult patients with MZL [24, 25, 44] and pediatric patients [4, 5, 26]. Since the cells of MZL are difficult to recognize in bone marrow smears by cytomorphology, flow cytometry analysis of peripheral blood and bone marrow is advisable. Furthermore, at suspicion of bone marrow involvement, bone marrow biopsy in addition to bone marrow aspirates for histopathological analysis is indicated. So far only one pediatric case of MZL with involvement of the CNS is reported [4]; however, a recent series in adult patients summarized 69 cases of enMZL of the CNS, interestingly with female preponderance, good treatment outcome and prognosis [45]. Therefore current staging recommendations include CNS directed staging with CSF evaluation and MRI in case of clinical symptoms.

Treatment and Outcome

Prospective clinical trials recruiting pediatric and adolescent patients with MZL are lacking. Recent case series reported diagnostic and clinical characteristics [9, 18]. It became obvious that standard intensive polychemotherapy according

to common treatment protocols for aggressive B-NHL [46–49] in children and adolescents results in overtreatment of the majority of pediatric MZL patients.

The non-Hodgkin lymphoma (NHL) Berlin-Frankfurt-Münster (BFM) group reported their results on pediatric MZL [5]. Between March 2004 and April 2015 a total of 33 MZL (20 nMZL and 13 enMZL) with central reference-pathological evaluation were identified among 2032 pediatric and adolescent patients with newly diagnosed NHL registered in the NHL-BFM data center. The NHL-BFM treatment protocol for mature aggressive B-NHL stratifies patients into four risk groups based on the resection status, LDH level, and stage of disease with two to six courses of chemotherapy [49]. Radiotherapy is not part of treatment protocols. Since MZL in pediatric patients is a rare condition, no standard treatment regimen is available and treatment decisions are made individually after consultation of the NHL-BFM study center. Until 2008 patients were treated with B-NHL treatment protocols. NHL-BFM recommendation for MZL patients diagnosed in 2008 or later with completely resected MZL is a watch and wait (w&w) strategy without systemic treatment. After complete (n = 16) and incomplete (n = 5) surgical resection, 21 patients received no systemic treatment but were followed by w&w. Eight patients received NHL-BFM chemotherapy for mature B-NHL [49] including two patients with two courses (risk group R1), five patients (15%) with four courses (R2), and one patient with six courses (R4). One patient (3%) received only systemic rituximab. All four patients with *Helicobacter*-associated MALT-lymphomas received therapy for eradication of *Helicobacter* spp. Afterward, endoscopic control with biopsy showed no lymphoma infiltration in one patient, who received no further treatment. Two patients received four doses rituximab due to persistence of MALT after *Helicobacter* spp. eradication therapy. The fourth patient with stage III disease received chemotherapy according to R2 risk group complemented with rituximab. With a median follow-up of 2.8 years, the probability of event-free for the NHL-BFM cohort is 84 ± 8% and of overall survival 100%. This series represents the largest population-based experience in pediatric MZL and supports the approach of complete staging and a watch & wait (w&w) strategy in standard MZL with close clinical monitoring even for cases without complete surgical resection.

A retrospective cooperative study of the international Berlin-Frankfurt-Münster (I-BFM) Study Group and the European Intergroup for Childhood NHL (EICNHL) analyzed and reported the largest series comprising a total of 66 pediatric and adolescent patients below 18 years of age with MZL including the above-cited series of the NHL-BFM group [4]. Of the 66 MZL, 44 (67%) were diagnosed as enMZL, 21 (32%) as nMZL and one splenic MZL. The latter patient, a 17-year-old female patient, was treated with splenectomy and is in remission for 5 years. Among the 21 nMZL, 17 patients underwent complete resection followed by w&w. One patient suffered relapse and is in remission after relapse treatment (see below). Four patients with incomplete resection of nMZL were followed by w&w in three patients and rituximab combined with chemotherapy in one patient. All four patients are in remission. Among the 44 patients with enMZL, 20 patients received complete surgical resection followed by w&w in 9 patients with continuous remission in 6 patients. Eleven out of the 20 enMZL with complete resection received additional treatment with continuous remission in 4 patients, 1 patient died from allogeneic transplantation and 6 patients suffered relapse. One of those six patients died from toxicity after allogeneic bone marrow transplantation. Twenty-four patients received incomplete resection followed by w&w in 3 patients; all 3 in remission. Twenty-one patients received additional treatment after incomplete resection with remission in 20 patients and relapse in one patient. The treatment and outcome of the 44 enMZL is summarized in Fig. 17.2.

Treatment of Gastric MALT

In the NHL-BFM series, all four patients with MALT lymphoma of the stomach received *Helicobacter* eradication therapy, two of them additionally received four rituximab courses because of persistence of the pathological findings [5]. One patient received systemic chemotherapy because of stage III disease. All four patients are event free. Especially in *Helicobacter* positive MALT lymphomas endoscopic control after eradication of *Helicobacter* with bioptic controls is advisable. Cases without evidence of further disease after eradication might not be in need of any further treatment. Radiotherapy is reported to be an effective treatment modality in gastric enMZL in adult patients [50–53]. Given the available alternative treatment approaches on the one hand and the risk of adverse acute and long-term effects of irradiation in children and adolescents on the other hand, the indication for local irradiation in pediatric enMZL is usually considered carefully.

Treatment of Skin Lesions

Several therapeutic approaches are in use in the treatment of cutaneous MZL in adult patients. Besides surgical resection of single lesions and antibiotics or systemic treatment with rituximab, local radiotherapy, interferon-alpha injections, or intralesional steroids are applied successfully [54–59]. Data on pediatric and adolescent patients are limited. In a small series of three pediatric patients treatment included excision of the MZL lesion or administration of antibiotics with com-

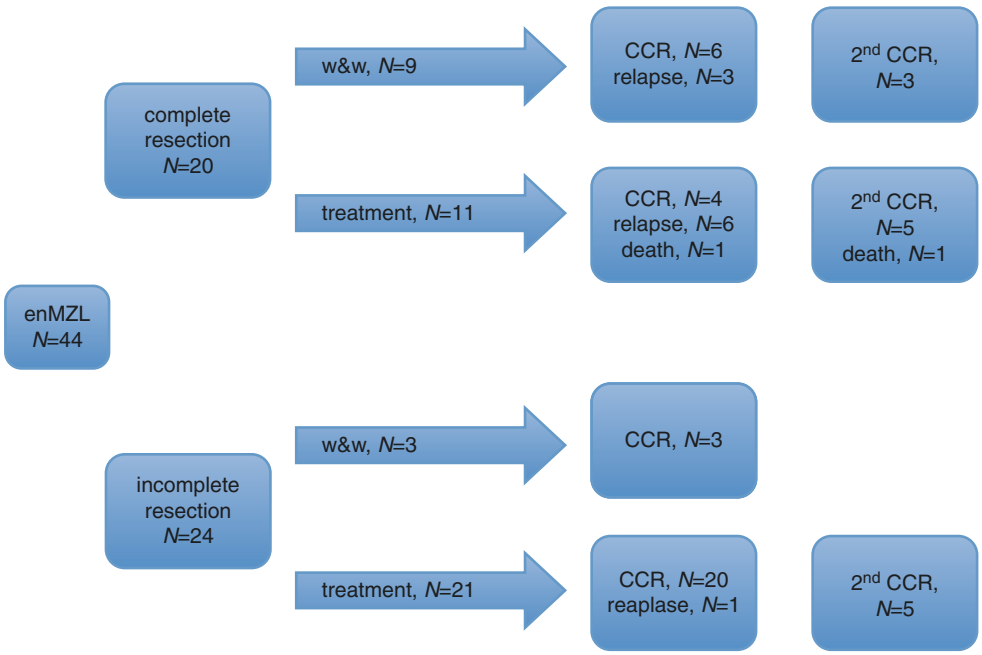

Fig. 17.2 Management of extranodal marginal zone lymphoma in the I-BFM and EICNHL study. Legend: enMZL, extranodal marginal zone lymphoma; w&w, watch and wait; CCR, continuous complete remission

plete remission in all three patients [14]. The cohort of the NHL-BFM group included only one patient with cutaneous MZL. This patient suffered relapse after complete resection of multiple lesions and w&w. At relapse, administration of intralesional rituximab achieved disease control [5]. In the literature intralesional administration of rituximab was reported in an 11-year-old boy [22] as well as in some small series of adult patients with favorable treatment tolerance and outcome [60–62].

Treatment of Ocular and Conjunctival Lymphoma

Adult patients with ocular and conjunctival MZL are treated with local steroids, antibiotic treatment, systemic rituximab, or local irradiation [21, 63]. Recent reports describe the intraocular application of rituximab in combination with systemic rituximab treatment [35, 64].

Treatment Strategies at Relapse

Relapses of MZL in children and adolescents are more frequent in enMZL than nMZL. The interval between initial diagnosis and the occurrence of relapse is variable between weeks and years. Also the site of relapse varies; in some cases relapse occurs at the site of initial diagnosis while in other patients new sites are involved at relapse. Pediatric MZL respond well to relapse treatment. If possible, local treatment approaches seem adequate at relapse. Individual patients require systemic treatment. Among 33 MZL patients of the NHL-BFM series, 4 patients suffered relapses [5]. One patient with completely resected nodal MZL manifestation in a parotid lymph node was diagnosed with relapse in an inguinal lymph node 3 months after initial diagnosis. After complete resection the patient received no further therapy and remained in remission. Another patient with extranodal MZL of the parotid gland with pre-existing Crigler-Najjar syndrome, relapsed 3 years after initial diagnosis and treatment with chemotherapy according to risk group R1. The patient was successfully treated. The third patient suffered relapse 2 months after complete resection of enMZL of the sublingual gland. Relapse was diagnosed in sublingual gland and in cervical and axillary lymph nodes, the patient received modified chemotherapy according to NHL-BFM R2 risk group and rituximab. This patient is alive free of progression. The fourth relapse was reported in a patient with cutaneous MZL 3 months after complete resection of the initial lesion. Retrospectively, this patient had a 4-year long history of recurrent skin lesions at the arm and submandibular area before the diagnosis of a cutaneous MZL was confirmed. At relapse, therapy with intra-lesional rituximab achieved a complete remission. The probability of overall survival for the NHL-BFM cohort is 100%.

The probability of overall survival for the EICNHL and I-BFM series of 66 MZL is excellent with 98 ± 2% [4]. Two MZL patients who underwent allogeneic bone marrow transplantation for their underlying immunodeficiency died of transplant-associated complications. The reported relapse rate is 17%, predominantly in enMZL with a median interval of 2.1 years from initial diagnosis. Six of 10 enMZL relapses occurred at the initial manifestation site while four presented at new sites. First-line management was radiotherapy ($n = 4$),

w&w ($n = 3$), and chemotherapy in two as well as rituximab plus chemotherapy in one patient; all the three latter patients suffered from preexisting disorders. Treatment at relapsed enMZL was chemotherapy ($n = 3$), chemotherapy plus rituximab ($n = 1$), rituximab monotherapy ($n = 2$), radiotherapy ($n = 2$), radiotherapy plus rituximab ($n = 1$), and w&w after complete resection in one patient. Ten of the eleven relapsed patients are in second remission, while one patient died from transplant-associated toxicity (Fig. 17.2).

Conclusion

Pediatric MZL represent a rare subtype of NHL predominantly observed in adolescents. Nodal MZL typically occur in male patients without underlying disease and present with limited disease and excellent prognosis. For extranodal MZL associations with infectious, inflammatory, and autoimmune stimuli are known. Differential diagnosis of MZL includes other B-cell lymphoma, especially pediatric follicular lymphoma, and non-malignant marginal zone hyperplasia underlining the role of experienced pathologists for diagnosis. Light chain restriction and evidence of clonality are characteristics for MZL. Individual patients with advanced and disseminated stage disease are reported, emphasizing the need for complete staging. The available clinical data form international series support the current approach of complete staging followed by watch & wait strategy with close clinical monitoring even in cases with incomplete resection.

References

1. Swerdlow SH, et al. WHO classification of tumors of hematopoietic and lymphoid tissues. 4th ed. Lyon: IARC Press; 2008.
2. Swerdlow SH, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127(20):2375–90.
3. Zinzani PL. The many faces of marginal zone lymphoma. Hematology Am Soc Hematol Educ Program. 2012;2012:426–32.
4. Ronceray L, et al. Children and adolescents with marginal zone lymphoma have an excellent prognosis with limited chemotherapy or a watch-and-wait strategy after complete resection. Pediatr Blood Cancer. 2018;65(4).
5. Makarova O, et al. Excellent outcome with limited treatment in paediatric patients with marginal zone lymphoma. Br J Haematol. 2017;182(5):735–9.
6. Amitay-Laish I, et al. Paediatric primary cutaneous marginal zone B-cell lymphoma: does it differ from its adult counterpart? Br J Dermatol. 2017;176(4):1010–20.
7. Collado P, et al. Extranodal marginal zone B cell lymphoma: an unexpected complication in children with Sjogren's syndrome. Reumatol Clin. 2018;14(4):227–9.
8. Hori D, et al. Pediatric nodal marginal zone lymphoma in a Japanese boy. Pediatr Int. 2017;59(3):358–60.
9. O'Suoji C, et al. Rare pediatric Non-Hodgkin lymphomas: a report from Children's Oncology Group Study ANHL 04B1. Pediatr Blood Cancer. 2016;63(5):794–800.
10. Ozawa MG, et al. A study of the mutational landscape of pediatric-type follicular lymphoma and pediatric nodal marginal zone lymphoma. Mod Pathol. 2016;29(10):1212–20.
11. Lum SH, et al. Successful curative therapy with rituximab and allogeneic haematopoietic stem cell transplantation for MALT lymphoma associated With STK4-mutated CD4+ lymphocytopenia. Pediatr Blood Cancer. 2016;63(9):1657–9.
12. Gabali A, et al. Pediatric extranodal marginal zone B-cell lymphoma presenting as amyloidosis in minor salivary glands: a case report and review of the literature. J Pediatr Hematol Oncol. 2013;35(3):e130–3.
13. Rizzo KA, et al. Marginal zone lymphomas in children and the young adult population; characterization of genetic aberrations by FISH and RT-PCR. Mod Pathol. 2010;23(6):866–73.
14. Kempf W, et al. Primary cutaneous marginal zone lymphoma in children: a report of 3 cases and review of the literature. Am J Dermatopathol. 2014;36(8):661–6.
15. Ryu M, et al. Pediatric mucosa-associated lymphoid tissue (MALT) lymphoma of lip: a case report and literature review. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2009;107(3):393–7.
16. Claviez A, et al. MALT lymphoma in children: a report from the NHL-BFM Study Group. Pediatr Blood Cancer. 2006;47(2):210–4.
17. Mo JQ, et al. MALT lymphoma in children: case report and review of the literature. Pediatr Dev Pathol. 2004;7(4):407–13.
18. Taddesse-Heath L, et al. Marginal zone B-cell lymphoma in children and young adults. Am J Surg Pathol. 2003;27(4):522–31.
19. van den Brand M, van Krieken JH. Recognizing nodal marginal zone lymphoma: recent advances and pitfalls. A systematic review. Haematologica. 2013;98(7):1003–13.
20. Desai A, et al. Long-term course of patients with primary ocular adnexal MALT lymphoma: a large single-institution cohort study. Blood. 2017;129(3):324–32.
21. Hoh H, et al. MALT lymphoma of the conjunctiva in a 13-year old child--5-year relapse-free follow-up following antibiotic treatment. Klin Monatsbl Augenheilkd. 2016;233(1):79–84.
22. Park MY, et al. Pediatric primary cutaneous marginal zone B-cell lymphoma treated with intralesional rituximab. Eur J Dermatol. 2010;20(4):533–4.
23. Ghatalia P, et al. Primary cutaneous marginal zone lymphoma associated with juxta-articular fibrotic nodules in a teenager. J Cutan Pathol. 2013;40(5):477–84.
24. Berger F, et al. Non-MALT marginal zone B-cell lymphomas: a description of clinical presentation and outcome in 124 patients. Blood. 2000;95(6):1950–6.
25. Sovani V, et al. Bone marrow trephine biopsy involvement by lymphoma: review of histopathological features in 511 specimens and correlation with diagnostic biopsy, aspirate and peripheral blood findings. J Clin Pathol. 2014;67(5):389–95.
26. Mittal N, et al. A unique description of stage IV extranodal marginal zone lymphoma (EMZL) in an adolescent associated with gamma heavy chain disease. Pediatr Blood Cancer. 2015;62(5):905–8.
27. Hyjek E, Isaacson PG. Primary B cell lymphoma of the thyroid and its relationship to Hashimoto's thyroiditis. Hum Pathol. 1988;19(11):1315–26.
28. Hyjek E, Smith WJ, Isaacson PG. Primary B-cell lymphoma of salivary glands and its relationship to myoepithelial sialadenitis. Hum Pathol. 1988;19(7):766–76.
29. Suarez F, et al. Infection-associated lymphomas derived from marginal zone B cells: a model of antigen-driven lymphoproliferation. Blood. 2006;107(8):3034–44.
30. Perrone S, et al. Infectious aetiology of marginal zone lymphoma and role of anti-infective therapy. Mediterr J Hematol Infect Dis. 2016;8(1):e2016006.
31. Ekstrom Smedby K, et al. Autoimmune disorders and risk of non-Hodgkin lymphoma subtypes: a pooled analysis within the InterLymph Consortium. Blood. 2008;111(8):4029–38.

32. Ferreri AJ, Govi S, Ponzoni M. Marginal zone lymphomas and infectious agents. Semin Cancer Biol. 2013;23(6):431–40.
33. Morgner A, et al. Malignant tumors of the stomach. Gastric mucosa-associated lymphoid tissue lymphoma and Helicobacter pylori. Gastroenterol Clin N Am. 2000;29(3):593–607.
34. Kluin PM, et al. Paediatric nodal marginal zone B-cell lymphadenopathy of the neck: a Haemophilus influenzae-driven immune disorder? J Pathol. 2015;236(3):302–14.
35. Bata BM, et al. Combined intraocular and systemic rituximab for ocular lymphoproliferative disorder with extranodal marginal zone lymphoma-type morphology after heart transplant. J AAPOS. 2018;22(2):159–61.
36. Gibson SE, et al. EBV-positive extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue in the posttransplant setting: a distinct type of posttransplant lymphoproliferative disorder? Am J Surg Pathol. 2011;35(6):807–15.
37. Aghamohammadi A, et al. Lymphoma of mucosa-associated lymphoid tissue in common variable immunodeficiency. Leuk Lymphoma. 2006;47(2):343–6.
38. Koo M, Ohgami RS. Pediatric-type follicular lymphoma and pediatric nodal marginal zone lymphoma: recent clinical, morphologic, immunophenotypic, and genetic insights. Adv Anat Pathol. 2017;24(3):128–35.
39. Maeshima AM, et al. Clinicopathological features of histological transformation from extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue to diffuse large B-cell lymphoma: an analysis of 467 patients. Br J Haematol. 2016;174(6):923–31.
40. Attygalle AD, et al. Atypical marginal zone hyperplasia of mucosa-associated lymphoid tissue: a reactive condition of childhood showing immunoglobulin lambda light-chain restriction. Blood. 2004;104(10):3343–8.
41. Caltharp SA, Qayed M, Park SI. Atypical marginal zone hyperplasia is a mimic for lymphoma in pediatric transplant recipients: report of two patients. Pediatr Dev Pathol. 2015;18(5):416–21.
42. Murphy SB. Classification, staging and end results of treatment of childhood non-Hodgkin's lymphomas: dissimilarities from lymphomas in adults. Semin Oncol. 1980;7(3):332–9.
43. Rosolen A, et al. Revised international pediatric Non-Hodgkin lymphoma staging system. J Clin Oncol. 2015;33(18):2112–8.
44. Park JY, et al. Bone marrow involvement is rare in superficial gastric mucosa-associated lymphoid tissue lymphoma. Dig Liver Dis. 2016;48(1):81–6.
45. Ayanambakkam A, et al. Extranodal marginal zone lymphoma of the central nervous system. Clin Lymphoma Myeloma Leuk. 2018;18(1):34–37 e8.
46. Fujita N, et al. Results of the Japan Association of Childhood Leukemia Study (JACLS) NHL-98 protocol for the treatment of B-cell non-Hodgkin lymphoma and mature B-cell acute lymphoblastic leukemia in childhood. Leuk Lymphoma. 2011;52(2):223–9.
47. Gerrard M, et al. Excellent survival following two courses of COPAD chemotherapy in children and adolescents with resected localized B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study. Br J Haematol. 2008;141(6):840–7.
48. Goldman S, et al. Rituximab and FAB/LMB 96 chemotherapy in children with Stage III/IV B-cell non-Hodgkin lymphoma: a Children's Oncology Group report. Leukemia. 2013;27(5):1174–7.
49. Woessmann W, et al. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. Blood. 2005;105(3):948–58.
50. Wirth A, et al. Long-term outcome for gastric marginal zone lymphoma treated with radiotherapy: a retrospective, multi-centre, International Extranodal Lymphoma Study Group study. Ann Oncol. 2013;24(5):1344–51.
51. Nam TK, et al. The role of radiotherapy in the treatment of gastric mucosa-associated lymphoid tissue lymphoma. Cancer Res Treat. 2014;46(1):33–40.
52. Olszewski AJ, Desai A. Radiation therapy administration and survival in stage I/II extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue. Int J Radiat Oncol Biol Phys. 2014;88(3):642–9.
53. Ruskone-Fourmestraux A, et al. Exclusive moderate-dose radiotherapy in gastric marginal zone B-cell MALT lymphoma: results of a prospective study with a long term follow-up. Radiother Oncol. 2015;117(1):178–82.
54. Fabbri A, et al. Rituximab plus liposomal pegylated doxorubicin in the treatment of primary cutaneous B-cell lymphomas. Eur J Haematol. 2014;93(2):129–36.
55. Suarez AL, et al. Primary cutaneous B-cell lymphomas: part II. Therapy and future directions. J Am Acad Dermatol. 2013;69(3):343 e1–11; quiz 355-6.
56. Hamilton SN, et al. Treatment and outcomes in patients with primary cutaneous B-cell lymphoma: the BC Cancer Agency experience. Int J Radiat Oncol Biol Phys. 2013;87(4):719–25.
57. Specht L, et al. Modern radiation therapy for primary cutaneous lymphomas: field and dose guidelines from the International Lymphoma Radiation Oncology Group. Int J Radiat Oncol Biol Phys. 2015;92(1):32–9.
58. Vandersee S, et al. Treatment of indolent primary cutaneous B-cell lymphomas with subcutaneous interferon-alfa. J Am Acad Dermatol. 2014;70(4):709–15.
59. Wilcox RA. Cutaneous B-cell lymphomas: 2018 update on diagnosis, risk-stratification, and management. Am J Hematol. 2018;93:1427–30.
60. Penate Y, et al. Intralesional rituximab in the treatment of indolent primary cutaneous B-cell lymphomas: an epidemiological observational multicentre study. The Spanish Working Group on Cutaneous Lymphoma. Br J Dermatol. 2012;167(1):174–9.
61. Kyrtsonis MC, et al. Favorable outcome of primary cutaneous marginal zone lymphoma treated with intralesional rituximab. Eur J Haematol. 2006;77(4):300–3.
62. Kerl K, et al. Intralesional and intravenous treatment of cutaneous B-cell lymphomas with the monoclonal anti-CD20 antibody rituximab: report and follow-up of eight cases. Br J Dermatol. 2006;155(6):1197–200.
63. Park HH, et al. Treatment outcome and risk analysis for cataract after radiotherapy of localized ocular adnexal mucosa-associated lymphoid tissue (MALT) lymphoma. Radiat Oncol J. 2017;35(3):249–56.
64. Ferreri AJ, et al. Intralesional rituximab: a new therapeutic approach for patients with conjunctival lymphomas. Ophthalmology. 2011;118(1):24–8.

Primary Central Nervous System Lymphoma

18

Oussama Abla, Tracy T. Batchelor, and Andishe Attarbaschi

Introduction

Primary central nervous system lymphoma (PCNSL) is an extranodal non-Hodgkin lymphoma (NHL) restricted to the brain, leptomeninges, eyes, and/or spinal cord [1]. An estimated 1425 cases of PCNSL were diagnosed each year in the United States (US) from 2007 to 2011, and since 2000 there has been a further increase in the incidence of PCNSL, especially in the elderly [2]. Between 1970 and 2000, the incidence of PCNSL increased, mainly due to the human immunodeficiency virus (HIV) pandemic. In adults, the median age at diagnosis is 65 years and PCNSL is slightly more common among males. The 5- and 10-year survival rates for adult PCNSL are 29.3% and 21.6%, respectively [2].

PCNSL is extremely rare in childhood, accounting for 1% of all PCNSL cases diagnosed from 1973 to 1998 in the US [3]. Surveillance, Epidemiology, and End Results (SEER) Program Data estimates suggest that around 15–20 cases of childhood PCNSL are diagnosed each year in the US [4]. More than 100 cases have been reported so far with the largest series consisting of 29 cases [3]. Although most reported cases were in immunocompetent patients, both acquired and congenital immunodeficiency syndromes increase the risk of PCNSL in children. The largest series reported a median age at diagnosis of 14 years. Although children with PCNSL appear to have a better prognosis than adults, the small number of cases and the lack of large clinical trials make such a conclusion debatable [3]. In this chapter, we will discuss what we have learned from adult clinical trials, and we will summarize data from recent pediatric case series to guide the treatment of PCNSL in children.

Pathogenesis

Acquired or congenital immunodeficiency syndromes are major risk factors for the development of PCNSL. Infection with HIV increases the risk of PCNSL by 3600 fold, and this is thought to have accounted for the increased incidence from 1970 to 2000. However, the incidence of PCNSL has decreased in the era of highly active anti-retroviral therapy (HAART) [5]. Central nervous system post-transplant lymphoproliferative disorder (PTLD) is the second most common malignancy to be diagnosed in organ transplant recipients after skin cancer [6]. The time of appearance of PCNSL following transplantation ranges from 3 weeks to 21 years, with a mean time of 33 months. Patients with congenital immunodeficiencies have a 4% risk of developing PCNSL. Immunosuppressive conditions such as systemic lupus erythematosus and vasculitis also increase the risk of PCNSL.

Histologic Features

In adults, almost 90% of PCNSL cases are diffuse large B-cell lymphomas (DLBCL), with the rest consisting of T-cell lymphomas, poorly characterized low-grade lymphomas or Burkitt's lymphomas [7]. Histologically, primary CNS DLBCL consists of centroblasts or immunoblasts usually clustered in the perivascular space, with reactive small

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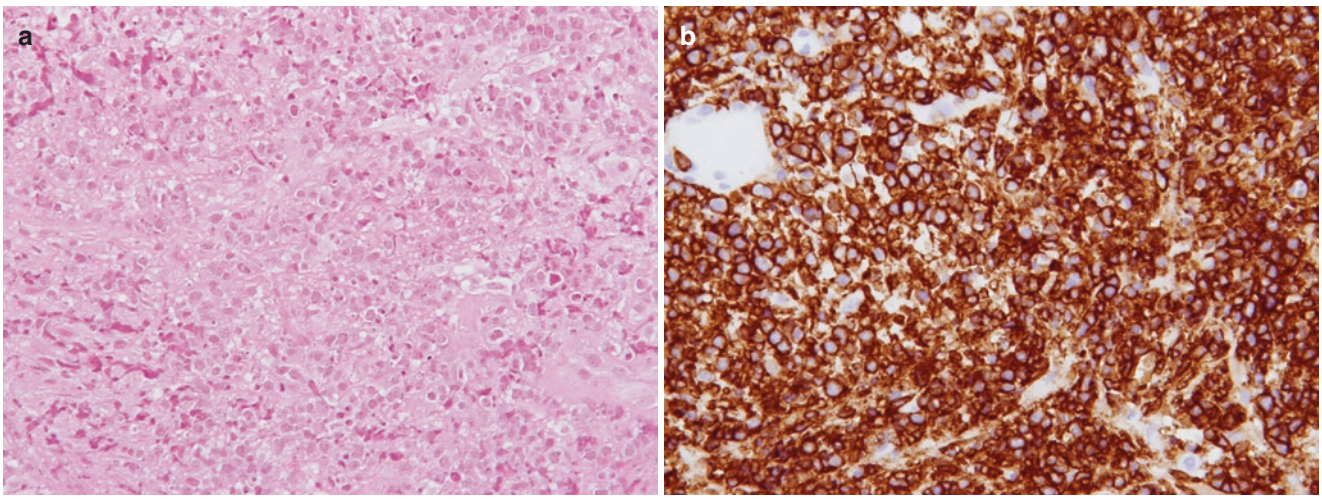

Fig. 18.1 Fourth ventricle biopsy showing diffuse large B-cell PCNSL: (**a**) neoplastic cells arranged in patternless sheets; the nuclei are vesicular with ~20% immunoblast-like nuclei demonstrating prominent nucleoli. Apoptotic bodies are frequent and a small area of necrosis is seen. Some cells have abundant cytoplasm; the tumor cells are diffusely immunopositive for CD45, CD79a, and CD20. (**b**) The MIB-1 proliferative index is almost 100%. (Courtesy of Dr Cynthia Hawkins, Hospital for Sick Children, Toronto)

lymphocytes, macrophages, and activated microglial cells intermixed with the tumor cells (Fig. 18.1a). Most tumors express pan-B-cell markers including CD19, CD20 (Fig. 18.1b), CD22, and CD79a. Similar to systemic DLBCL, PCNSL harbors chromosomal translocations of the *BCL6* gene, deletions in 6q, and aberrant somatic hypermutation in proto-oncogenes including *MYC* and *PAX5*. In addition, PCNSL can be classified into three molecular subclasses by gene expression profiling: type 3 large B-cell lymphoma, germinal center B-cell (GCB) lymphoma, and post-germinal center activated B-cell lymphoma (ABC). In DLBCL cases, the ABC gene expression profile is associated with an inferior prognosis versus the GCB profile. The ABC subclass accounted for >95% of primary CNS DLBCL cases in one series [8]. This higher prevalence of the ABC gene expression profile subtype in PCNSL is probably responsible for the relatively inferior prognosis of this lymphoma compared to systemic DLBCL. Moreover, there are other molecular features that distinguish primary CNS DLBCL from systemic DLBCL. Gene expression profiles demonstrate that PCNSL is characterized by differential expression of genes related to adhesion and extracellular matrix pathways, including *MUM1*, *CXCL13*, and *CHI3L1* [9]. Recent genomic analysis of adults with PCNSL showed a high frequency of oncogenic gain-of-function mutations in MYD88 (MYD88L265P), missense mutations in the immunoreceptor tyrosine-based activation motif domain of CD79B, and missense mutations in the coiled-coil domain of CARD11 [10]. These observations provide insight into potential therapeutic targets for future clinical trials in PCNSL.

DLBCL is also the most common histologic subtype in pediatric PCNSL, with an incidence ranging from 30% to 70% [3, 11–14]. Pediatric systemic DLBCL and CNS DLBCL have moderate to high proliferation index, decreased BCL2 protein expression, and increased frequency of the GCB phenotype (BCL6+) which may contribute partially to better outcomes in children compared to adults [3, 15]. Other common pathological subtypes in children include anaplastic large cell, Burkitt's, and lymphoblastic lymphomas [3].

Clinical Features

The clinical presentation of children with PCNSL is variable. The most common presenting symptoms are those of increased intracranial pressure including headaches and vomiting, followed by ataxia, dysarthria, and hemiparesis. Blurred vision, nystagmus, diplopia, and ptosis have also been reported [3, 12]. Seizures are less common than with other types of brain tumors probably because PCNSL involves mainly the subcortical white matter rather than the epileptogenic gray matter. Other rare presentations include cranial polyneuropathy, Parinaud syndrome due to pineal location of PCNSL, and diabetes insipidus with a thickened pituitary stalk mimicking Langerhans cell histiocytosis [3]. Neuropsychiatric signs can also occur [12] but are more common in adults (43%). Unlike patients with systemic NHL, PCNSL patients rarely manifest B symptoms.

In adults, prognostic scoring systems have been developed specifically for PCNSL [16, 17]. The International Extranodal Lymphoma Study Group (IELSG) identified age >60 years, Eastern Cooperative Oncology Group (ECOG) performance status >1, elevated serum lactate dehydrogenase (LDH) level, elevated cerebrospinal fluid (CSF)

protein concentration, and involvement of deep regions of the brain as independent predictors of poor prognosis. In patients with 0–1 factors (low risk), 2–3 factors (intermediate risk), and 4–5 factors (high risk), the 2-year survival rates were 80%, 48%, and 15%, respectively [16]. In another prognostic model, adult PCNSL patients were divided into three groups based on age and performance status: (1) <50 years old; (2) ≥50 years old with a KPS ≥70; and (3) ≥50 years old with a KPS <70. Based on these three divisions, significant differences in overall and failure-free survival were observed [17].

In the largest pediatric PCNSL series, an ECOG performance status of 0–1 was the only prognostic factor associated with better outcomes with hazard ratios of 0.136 ($p = 0.017$) and 0.073 ($p = 0.033$) for progression-free survival (PFS) and overall survival (OS), respectively; while age, serum LDH, CSF protein, and involvement of deep brain lesions were not significant likely due to the small number of patients [3].

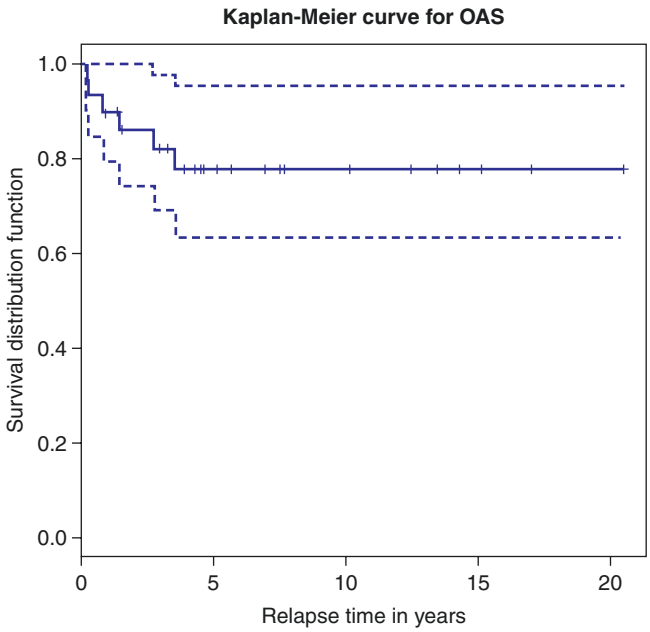

Fig. 18.2 Two-year overall survival curve for 29 children and adolescents with PCNSL [3]

Diagnostic Evaluations

The International PCNSL Collaborative Group (IPCG) has developed guidelines for initial diagnostic assessment of PCNSL patients [18]. A gadolinium-enhanced brain magnetic resonance imaging (MRI) scan is the optimal radiographic study for the detection of PCNSL. If MRI is not possible or is contraindicated, a contrast-enhanced brain computed tomography (CT) scan is recommended. Most children with PCNSL present with a single brain mass, while around 40% present with multiple lesions. The lesions are usually isointense to hypointense on T2-weighted MRI scan sequences (Fig. 18.2). Since PCNSL is characterized by a high nuclear:cytoplasmic ratio and high cell density, there may be regions of restricted diffusion observed on diffusion-weighted MRI sequences, and apparent diffusion coefficient imaging was found to be a useful biomarker of response to chemotherapy in adults [19], but has never been evaluated in children. Involvement of cerebral hemispheres occurs in almost 50% of children, while involvement of deep brain structures (basal ganglia, cerebellum, or brain stem) occurs in at least 40% [3]. The diagnosis of PCNSL is usually made by stereotactic brain biopsy. Occasionally, if a brain biopsy is not safe then a lumbar puncture with CSF analysis (flow cytometry, cytology, and immunoglobulin heavy-chain gene rearrangement), or analysis of vitreous fluid aspirate (with MYD88 PCR) [10] in patients with ocular involvement could be diagnostic. Concurrent leptomeningeal and ocular involvement occurs in approximately 15%–20% and 5%–25% of adult PCNSL patients, respectively. Concurrent leptomeningeal involvement occurred in 17% of patients in the largest pediatric series [3], while isolated leptomeningeal disease was reported in 18% of cases in a previous review [12]. Isolated spinal cord involvement is rare and observed in <1% of cases; therefore spinal MRI is only necessary based on clinical suspicion or to screen for leptomeningeal involvement if a lumbar puncture cannot be safely performed.

A thorough diagnostic work-up is needed to establish the extent of the disease and to confirm isolated CNS involvement. Physical examination should include lymph node examination, a testicular examination in males, and a comprehensive neurological examination. A lumbar puncture should be performed if not contraindicated, and CT/PET scans of the chest, abdomen, and pelvis, and a bone marrow aspirate and biopsy are recommended to exclude occult systemic lymphoma. A slit lamp examination and a comprehensive eye evaluation by an ophthalmologist are required to exclude involvement of the optic nerve, retina, or vitreous humor. Blood tests should include complete blood count, basic metabolic panel, serum LDH, and HIV serology [16].

Treatment

The optimal therapy for children with PCNSL has not yet been determined. Most pediatric centers routinely treat PCNSL patients with either an LMB-96/BFM-based protocols or with regimens derived from the adult experience but with some pediatric adaptation, as shown in the Toronto treatment algorithm (Fig. 18.3).

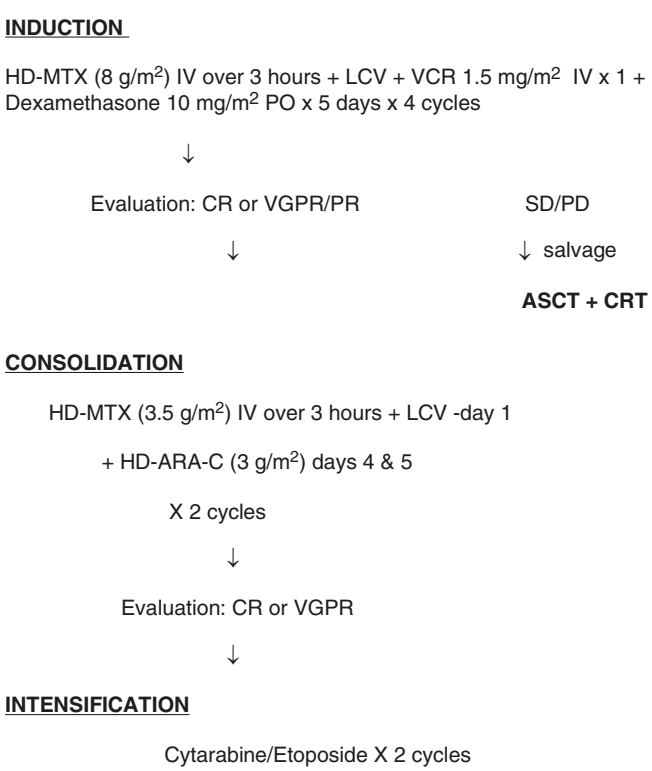

Fig. 18.3 Toronto treatment protocol for pediatric primary CNS lymphoma

Surgery

Due to the multifocal nature of PCNSL, surgical resection is not usually part of the standard treatment [20]. In both adults and children, the role of neurosurgery is to establish a diagnosis through a stereotactic brain biopsy or, in rare cases, to relieve an impending brain herniation via emergent debulking. Although in one report there was a possible benefit of gross total resection in adult PCNSL patients, this was a retrospective study and subset analysis was probably limited by selection bias [21]. Adults with PCNSL have a median survival of 1–4 months following surgery alone [22]. Thus, the recommendation in childhood PCNSL is to restrict surgery to a diagnostic stereotactic biopsy.

Corticosteroids

Corticosteroids should be avoided prior to a stereotactic biopsy if possible, due to the risk of altering cellular morphology which can lead to a non-diagnostic pathology sample. Steroids can decrease tumor-associated edema and may result in partial radiographic regression of PCNSL. However, after an initial response, almost all patients quickly relapse. Nevertheless, one study showed that initial radiographic response to corticosteroids in newly diagnosed adult PCNSL patients was a favorable prognostic marker, with survival of 117 months in responders versus 5.5 months in non-responders [23].

Whole-Brain Radiation Therapy

Historically, PCNSL was treated in adults with whole-brain radiation therapy (WBRT) alone at doses ranging from 36 to 45 Gy which resulted in a high proportion of radiographic responses, but the responses were not durable. In a multicenter phase II trial, 41 adults were treated with WBRT at 40-Gy plus a 20-Gy tumor boost and achieved a median OS of 12 months and a 2-year OS of 25% [24]. A historical pediatric review reported similar outcomes in children treated with WBRT alone or in combination with low-dose chemotherapy with a median OS of 17 months [25]. Due to the early relapses after radiation and the associated risk of neurotoxicity, WBRT alone is not a recommended treatment for children with newly diagnosed PCNSL, except in the palliative care setting.

Chemo-Radiotherapy Combinations

Prior to the establishment of methotrexate as the foundation of chemotherapy for PCNSL, regimens that were standard of care for systemic NHL were adopted. A randomized trial of WBRT versus WBRT and cyclophosphamide, doxorubicin, vincristine, and prednisone (CHOP) in adults was terminated early due to poor accrual, although results suggested that WBRT and CHOP was not superior to WBRT alone [26]. Given that the agents in the CHOP regimen poorly penetrate the blood-brain barrier, and can be quite toxic if administered at high doses, this treatment regimen was abandoned for patients with PCNSL. One adult study reported improved outcomes when WBRT was combined with high-dose methotrexate (HD-MTX)-based chemotherapy regimens [27]. A more recent randomized adult study evaluated HD-MTX with or without WBRT in patients who achieve a complete response after chemotherapy. There was improved PFS among patients treated with chemotherapy plus WBRT, although there was no difference in OS [28].

In childhood PCNSL, WBRT has not been evaluated in a randomized fashion, but there are retrospective reports of patients having favorable outcomes with chemotherapy alone [3, 11, 13]. A small retrospective series of 12 patients from 2006 reported a 5-year event-free survival (EFS) of 70% among the 10 patients who were treated with chemotherapy alone [11]. The largest childhood PCNSL series of 29 patients failed to show a benefit from WBRT, and response rates were higher in patients who received frontline chemotherapy alone possibly because of the increased doses of che-

motherapy given in the absence of WBRT [3]. A more recent pediatric series from the Berlin-Frankfurt-Munster (BFM) group suggested that most children with PCNSL can potentially be cured without WBRT [13]. Considering these results together with the adult data, and due to the potential for serious neurocognitive dysfunction from WBRT in children [29], this modality should be ideally reserved for patients with refractory or relapsed disease.

Systemic Chemotherapy

Given the treatment-related neurotoxicity of WBRT, systemic chemotherapy is the most important part in the treatment of PCNSL. Chemotherapeutic agents such as anthracyclines and cyclophosphamide with poor penetration into the blood-brain barrier (BBB), and with potential toxicity when given at high doses, are not as effective as in systemic lymphomas. CHOP regimens, which are highly active in systemic DLBCL, have indeed shown little activity in adult PCNSL [30]. Methotrexate is the most active agent in the treatment of PCNSL. In adults, HD-MTX has been given at variable doses and schedules but in general, doses ≥3 g/m^2 delivered as an initial bolus followed by an infusion over 3 hours, and administered every 21 days is recommended for optimal outcomes and adequate CSF concentrations [31]. Multiple, phase 2 studies have demonstrated the safety, efficacy, and relatively preserved cognition of HD-MTX-based chemotherapy regimens [32, 33]. Moreover, longer duration of induction chemotherapy with HD-MTX (>6 cycles) has been associated with higher complete response (CR) rates [32, 34]. Prospective randomized trials in adults have shown better response rates and PFS when HD-MTX was combined with thiotepa and HD-Ara-C, both of which have high CNS penetration [35, 36].

Although large trials for PCNSL are lacking in children, small case series and case reports demonstrated that multiagent chemotherapeutic regimens including HD-MTX and HD-Ara-C are effective in most patients. The first large series of pediatric PCNSL consisted of 12 children diagnosed between 1995 and 2003; 10 were treated with chemotherapy alone and 90% of them had HD-MTX and/or HD-Ara-C as part of their treatment, and their 5-year EFS was at 70% [11]. The largest pediatric series included 29 patients treated by either adult or pediatric oncologists, and 93% of them were treated with MTX-containing regimens and mostly with HD-MTX and HD-Ara-C combinations. FAB/LMB 96 protocol was the most common regimen used in this series, and consisted of HD-MTX (3–8 g/m^2), HD-Ara-C (3 g/m^2), and triple intrathecal chemotherapy. The overall response rate was 86%, while the 2-year OS (Fig. 18.2) and PFS (Fig. 18.4) rates were 86% and 61%, respectively; the 3-year OS was 82%. Primary treatment

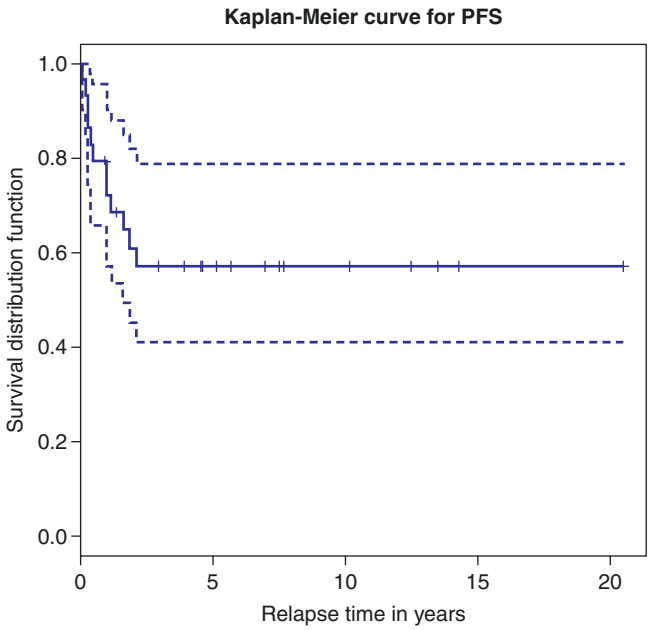

Fig. 18.4 Two-year progression-free survival for pediatric PCNSL [3]

with chemotherapy alone was associated with better overall response rates, and there was a marginally significant relationship between higher doses of MTX and response (p = 0.06) [3]. We have had an anecdotal experience with HD-MTX at 8 g/m^2 (for 4 cycles) inducing a rapid CR in a teenage boy with multifocal PCNSL (Fig. 18.5a, b) [37]. In the largest pediatric series, an ECOG performance score of 0–1 was the only factor associated with better PFS and OS. Five of the six patients who relapsed after chemotherapy alone regimens responded to salvage therapies and were in CR at the time of report [3]. Similarly, a smaller series of six patients treated with LMB96-like regimens showed favorable outcomes with a 5-year OS of 83% [14]. More recently, the BFM group published a series of 17 children (both immunocompetent and immunocompromised) with PCNSL treated according to three consecutive BFM-NHL protocols between 1990 and 2011. Treatment included six cycles of HD-MTX, HD-Ara-C and triple, intrathecal chemotherapy but also with anthracyclines and alkylating agents. Only ALCL patients (n = 5) received WBRT (24 Gy) as an additional part of their therapy. After a median follow-up of 7.5 years, the 3-year OS of the entire cohort was 63%, while the 3-year OS among immunocompetent patients was 92% [13]. In addition, the Children's Oncology Group (COG) published data from their rare NHL registry, which had 5 patients with PCNSL. There were no treatment details on this small cohort of PCNSL patients, but their PFS and OS rates were 100% at a median follow-up of 2.1 years [38]. A summary of all pediatric series and treatment results is shown in Table 18.1.

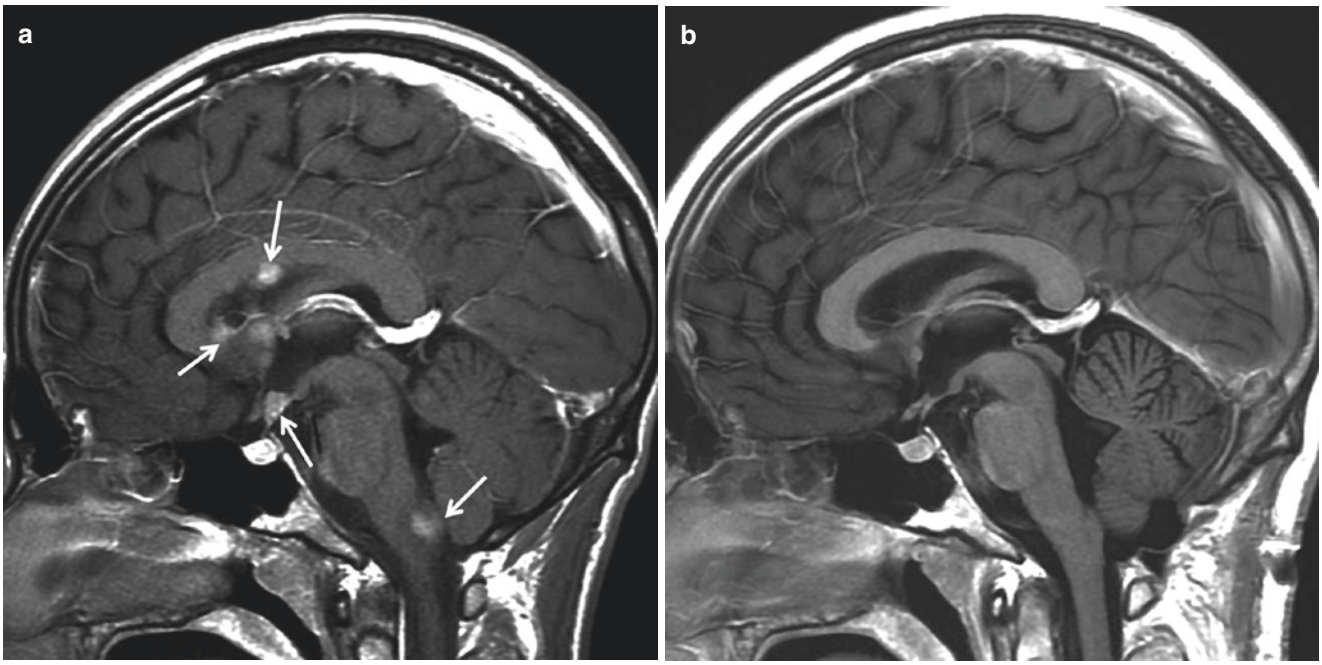

Fig. 18.5 Brain MRI of a 17–year-old immunocompetent boy with PCNSL: (**a**) sagittal T1 weighted post-gadolinium MRI demonstrates multiple enhancing nodules in the medulla, pituitary recess, and along the corpus callosum (*white arrows*); (**b**) follow-up MRI after four cycles of HD-MTX (8 g/m^2) demonstrates complete resolution of the nodules. (Courtesy of Dr Helen Branson, Hospital for Sick Children, Toronto)

Intrathecal Chemotherapy

The role of intrathecal (IT) chemotherapy in pediatric PCNSL is not well defined, although it has been used in most pediatric multi-agent chemotherapeutic regimens. In adults, two retrospective non-randomized studies failed to show any added benefit from IT chemotherapy in the setting of HD-MTX-containing regimens [39, 40]. Consequently, due to the ability of HD-MTX of achieving micromolar concentrations in the CSF, most adult trials do not include IT chemotherapy. There has been no comparison between the outcomes of children treated with high-dose chemotherapy with or without IT chemotherapy.

Immunotherapy

Rituximab is a chimeric monoclonal antibody targeting the CD20 antigen that has been incorporated into induction chemotherapy regimens in adult PCNSL. At an IV dose of 375–800 mg/m^2 of rituximab, CSF levels from 0.1% to 4.4% of serum levels are usually achieved. Despite the limited CSF penetration, relapsed adult PCNSL patients have achieved radiographic responses when treated with rituximab monotherapy [41]. In addition, historically, radiographic CR rates are higher with induction protocols that include rituximab vs those without rituximab [42]. An adult phase II randomized trial showed a superior response of the combination of HD-MTX/Ara-C/thiotepa/rituximab (MATRix) compared with HD-MTX/Ara-C alone and compared with HD-MTX/Ara-C and rituximab [35].

In children, the Inter-B-NHL Ritux 2010 international trial combines a randomized phase III study testing the impact of adding rituximab to the LMB regimen for advanced-stage B-cell lymphoma. The randomization was terminated early due to the high probability of superiority in the rituximab arm. The 1-year EFS was 94.2% vs 81.5% in the rituximab vs no rituximab arms, respectively, and the hazard ratio was 0.33 (90% confidence interval, 0.16–0.69; $P = 0.006$) [43]. In pediatric patients with CD20+ PCNSL, rituximab has been given alone or in combination with chemotherapy to a small number of patients [3] but the data is insufficient to make any definite conclusions.

Autologous Stem Cell Transplantation (ASCT)

Given the success of high-dose chemotherapy (HDT) followed by autologous stem cell transplantation (ASCT) in relapsed or refractory systemic NHL and PCNSL, this approach has been studied as consolidation for newly diagnosed adults with PCNSL [44]. Conditioning regimens including thiotepa have shown the most promising results. In a multicenter, phase II trial, 79 patients were treated with induction HD-MTX, cytarabine, rituximab, and thiotepa, followed by carmustine and thiotepa conditioning prior to

Table 18.1 Summary of the pediatric PCNSL case series reported from 2006 until 2016

Histology								Treatment					
Reference	*N*	Median age (range)	Percentage immuno-compromised	DLBCL (%)	ALCL (%)	BL (%)	Other (%)	Chemo alone (%)	Chemo + RT (%)	Resection before treatment (%)	Other (%)	Chemotherapy regimen	Outcome
Abla et al. [11]	12	7.5y (4–17y)	33	42	33	8	17	83	17	8	0	Ara-C and/or MTX based: 92% -Palliative hydroxyurea: 8%	9/12 patients alive
Abla et al. [3]	29	14y (2–21y)	10	69	17	7	7	62	31	31	7	MTX-based: 93% Non-MTX based: 7%	5-y EFS 70%[a]; 3-y OS 82%
Yoon et al. [14]	6	10y (23mo–13y)	0	50	0	33	17	100	0	0	0	LMB96: 83% CCG106B: 17%	5-y OS 83%
Thorer et al. [13]	17	13y (1–17y)	30	41	29	6	24	71	29	0	0	NHL-BFM90, NHL-BFM95, or B-NHL BFM04	3-y OS 63% (whole cohort) 3-y OS 92% (immuno competent)
O'Suoji et al. [38]	5	13y (6–16y)	20	40	40	0	20	NR	NR	NR	NR	NR	EFS and OS 100% with median follow-up 2.1 y

NR not reported

[a]Among patients treated with chemotherapy alone

ASCT. The overall response rate was 91%, and the 2-year OS was 87% and treatment-related deaths occurred in <10% of patients [44]. More recently, Ferreri et al. reported the results of a randomized study that addressed the efficacy of myeloablative chemotherapy supported by ASCT, as an alternative to WBRT, as consolidation after high-dose-methotrexate-based chemoimmunotherapy. WBRT and ASCT were both equally feasible and effective as consolidation therapies after high-dose methotrexate-based chemoimmunotherapy in patients aged 70 years or younger with PCNSL. Therefore, the risks and implications of cognitive impairment after WBRT should be considered at the time of therapeutic decision [45]. There are three ongoing, multicenter, randomized trials in adults comparing the efficacy of consolidative HDT/ASCT versus chemotherapy or WBRT for newly diagnosed PCNSL (NCT01011920, NCT00863460, NCT01511562). In the largest pediatric PCNSL series, ASCT was used as consolidative therapy in 2 of 29 newly diagnosed PCNSL; 10 of the 29 cases relapsed and 4 of these underwent salvage therapy followed by ASCT, with all 4 patients being alive at the time of report [3].

Future Perspectives and Unanswered Questions

Novel therapeutic agents currently under study for adult primary CNS DLBCL include ibrutinib, lenalidomide, pomalidomide, buparlisib, pemetrexed, everolimus, and bendamustine [9]. In a phase I study of dose-escalating lenalidomide, 9 patients with relapsed or refractory CNS lymphoma were treated and 8/8 evaluable subjects achieved objective responses (4 CR, 4 PR) after 1 month of lenalidomide monotherapy. In a separate cohort of ten patients from the same study with relapsed or refractory CNS lymphoma, lenalidomide was administered as maintenance therapy after first-line salvage treatment, and five patients maintained durable responses ≥2 years suggesting a role for this drug in consolidation or maintenance therapy [46]. Clinical trials of lenalidomide plus rituximab for relapsed or refractory PCNSL are ongoing in adults. Unanswered questions remain in PCNSL, such as the role of PET scans in the initial evaluation of disease response, a clear definition of a complete remission, role of the frontline addition of rituximab, and the optimal treatment of refractory and relapsed patients.

Understanding the molecular biology of pediatric PCNSL is challenging due to the rarity of the disease and the scarcity of pathology samples on brain biopsies. In addition, due to the rarity of PCNSL in children and the need for many years of follow-up to detect late relapses, it is obvious that no prospective phase III trials are feasible through any of the single pediatric cooperative groups. It is also questionable whether children with PCNSL benefit from the addition of drugs with little penetration through the BBB, such as anthracyclines and alkylating agents. An international collaboration between the IPCG members and pediatric international groups (COG-NHL committee, German NHL-BFM and French-LMB groups) might lead to the development of longitudinal data registries and ultimately better treatment strategies for children and adolescents with PCNSL.

Conclusions

Evidence-based treatment recommendations for childhood and adolescent PCNSL are not well established, particularly in the absence of large clinical trials. Combination chemotherapy regimens, largely dependent on the histopathological subtype, are reasonable choices for most patients. Since higher doses of MTX have been correlated with long-term remissions in PCNSL and because children can tolerate higher doses of MTX versus adults, it is appropriate to start with induction therapy consisting of three or four cycles of MTX at 5–8 g/m^2 with folinic acid rescue and, in case of a good early response, to give consolidation with HD-MTX and HD-Ara-C. However, BFM and FAB/LMB96 protocols have also yielded favorable outcomes and could be considered as reasonable treatment options as well. The role of IT chemotherapy is not established especially in the context of HD-MTX-containing regimens. Rituximab may be of benefit in mature B-cell PCNSL based on data from pediatric systemic B-NHL, as well as from the adult PCNSL literature. In pediatric PCNSL, autologous stem cell transplant and WBRT are not recommended during frontline therapy and are routinely advised for refractory or relapsed cases. Of note, however, ASCT is now listed as a frontline consolidation option for adults with PCNSL in the most recent NCCN guidelines. An international case registry between the International BFM study group and the COG is currently enrolling children and adolescents with PCNSL, which could provide better therapeutic insights into this rare pediatric lymphoma. Lastly, a better understanding of the molecular signature of pediatric PCNSL is warranted as this might lead to novel, more effective, and possibly less toxic targeted therapies in the future.

References

1. Batchelor TT, DeAngelis LM, editors. Lymphoma and leukemia of the nervous system. 2nd ed. New York: Springer; 2013.
2. Ostrom QT, Gittleman H, Liao P, Rouse C, Chen Y, Dowling J, et al. CBTRUS statistical report: primary brain and central nervous system tumors diagnosed in the United States in 2007-2011. Neuro-Oncology. 2014;16(Suppl 4):iv1–63.
3. Abla O, Weitzman S, Blay JY, O'Neill BP, Abrey LE, Neuwelt E, et al. Primary CNS lymphoma in children and adolescents: a descriptive analysis from the International Primary CNS Lymphoma Collaborative Group (IPCG). Clin Cancer Res. 2011;17(2):346–52.

4. Hochberg J, El-Mallawany NK, Abla O. Adolescent and young adult non-Hodgkin lymphoma. Br J Haematol. 2016;173(4):637–50.
5. Matinella A, Lanzafame M, Bonometti MA, Gajofatto A, Concia E, Vento S, et al. Neurological complications of HIV infection in pre-HAART and HAART era: a retrospective study. J Neurol. 2015;262(5):1317–27.
6. Cavaliere R, Petroni G, Lopes MB, Schiff D, International Primary Central Nervous System Lymphoma Collaborative Group. Primary central nervous system post-transplantation lymphoproliferative disorder: an International Primary Central Nervous System Lymphoma Collaborative Group Report. Cancer. 2010;116(4):863–70.
7. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, Vardiman JW, editors. WHO classification of tumours of haematopoietic and lymphoid tissues. Lyon: IARC; 2008.
8. Camilleri-Broet S, Criniere E, Broet P, Delwail V, Mokhtari K, Moreau A, et al. A uniform activated B-cell-like immunophenotype might explain the poor prognosis of primary central nervous system lymphomas: analysis of 83 cases. Blood. 2006;107(1):190–6.
9. Ponzoni M, Issa S, Batchelor TT, Rubenstein JL. Beyond high-dose methotrexate and brain radiotherapy: novel targets and agents for primary CNS lymphoma. Ann Oncol. 2014;25(2):316–22.
10. Chapuy B, Roemer MGM, Stewart C, Tan Y, Abo RP, Zhang L, et al. Targetable genetic features of primary testicular and primary central nervous system lymphomas. Blood. 2016;127(7):869–81.
11. Abla O, Sandlund JT, Sung L, et al. A case series of pediatric primary central nervous system lymphoma: favorable outcome without cranial irradiation. Pediatr Blood Cancer. 2006;47:880–5.
12. Abla O, Weitzman S. Primary CNS lymphoma in children. Neurosurg Focus. 2006;21:E8.
13. Thorer H, Zimmermann M, Makarova O, et al. Primary central nervous system lymphoma in children and adolescents: low relapse rate after treatment according to non-Hodgkin-lymphoma Berlin-Frankfurt-Munster protocols for systemic lymphoma. Haematologica. 2014;99:e238–41.
14. Yoon JH, Kang HJ, Kim H, et al. Successful treatment of primary central nervous system lymphoma without irradiation in children: single center experience. J Korean Med Sci. 2012;27:1378–84.
15. Miles R, Raphael M, McCarthy K, et al. Pediatric diffuse large B-cell lymphoma demonstrates a high proliferation index, frequent c-Myc protein expression, and a high incidence of germinal center subtype: report of the French-American-British (FAB) International Study Group. Pediatr Blood Cancer. 2008;51:369–74.
16. Ferreri AJ, Blay JY, Reni M, Pasini F, Spina M, Ambrosetti A, et al. Prognostic scoring system for primary CNS lymphomas: the International Extranodal Lymphoma Study Group experience. J Clin Oncol. 2003;21(2):266–72.
17. Abrey LE, Ben-Porat L, Panageas KS, Yahalom J, Berkey B, Curran W, et al. Primary central nervous system lymphoma: the Memorial Sloan-Kettering Cancer Center prognostic model. J Clin Oncol. 2006;24(36):5711–5.
18. Abrey LE, Batchelor TT, Ferreri AJ, Gospodarowicz M, Pulczynski EJ, Zucca E, et al. Report of an international workshop to standardize baseline evaluation and response criteria for primary CNS lymphoma. J Clin Oncol. 2005;23(22):5034–43.
19. Barajas RF Jr, Rubenstein JL, Chang JS, Hwang J, Cha S. Diffusion-weighted MR imaging derived apparent diffusion coefficient is predictive of clinical outcome in primary central nervous system lymphoma. AJNR Am J Neuroradiol. 2010;31(1):60–6.
20. Bellinzona M, Roser F, Ostertag H, Gaab RM, Saini M. Surgical removal of primary central nervous system lymphomas (PCNSL) presenting as space occupying lesions: a series of 33 cases. Eur J Surg Oncol. 2005;31(1):100–5.
21. Weller M, Martus P, Roth P, Thiel E, Korfel A, German PCNSL Study Group. Surgery for primary CNS lymphoma? Challenging a paradigm. Neuro-Oncology. 2012;14(12):1481–4.
22. Batchelor T, Loeffler JS. Primary CNS lymphoma. J Clin Oncol. 2006;24(8):1281–8.
23. Mathew BS, Carson KA, Grossman SA. Initial response to glucocorticoids. Cancer. 2006;106(2):383–7.
24. Nelson DF, Martz KL, Bonner H, Nelson JS, Newall J, Kerman HD, et al. Non-Hodgkin's lymphoma of the brain: can high dose, large volume radiation therapy improve survival? Report on a prospective trial by the Radiation Therapy Oncology Group (RTOG): RTOG 8315. Int J Radiat Oncol Biol Phys. 1992;23(1):9–17.
25. Kai Y, Kuratsu J, Ushio Y. Primary malignant lymphoma of the brain in childhood. Neurol Med Chir. 1998;38:232–7.
26. Mead GM, Bleehen NM, Gregor A, Bullimore J, Shirley D, Rampling RP, et al. A medical research council randomized trial in patients with primary cerebral non-Hodgkin lymphoma: cerebral radiotherapy with and without cyclophosphamide, doxorubicin, vincristine, and prednisone chemotherapy. Cancer. 2000;89(6):1359–70.
27. O'Brien P, Roos D, Pratt G, et al. Phase II multicenter study of brief single-agent methotrexate followed by irradiation in primary CNS lymphoma. J Clin Oncol. 2000;18:519–26.
28. Thiel E, Korfel A, Martus P, et al. High-dose methotrexate with or without whole brain radiotherapy for primary CNS lymphoma (G-PCNSL-SG-1): a phase 3, randomised, non-inferiority trial. Lancet Oncol. 2010;11:1036–47.
29. Said JA, Waters BG, Cousens P, Stevens MM. Neuropsychological sequelae of central nervous system prophylaxis in survivors of childhood acute lymphoblastic leukemia. J Consult Clin Psychol. 1989;57:251–6.
30. Schultz C, Scott C, Sherman W, et al. Preirradiation chemotherapy with cyclophosphamide, doxorubicin, vincristine, and dexamethasone for primary CNS lymphomas: initial report of radiation therapy oncology group protocol 88-06. J Clin Oncol. 1996;14:556–64.
31. Ferreri AJ, Guerra E, Regazzi M, Pasini F, Ambrosetti A, Pivnik A, et al. Area under the curve of methotrexate and creatinine clearance are outcome-determining factors in primary CNS lymphomas. Br J Cancer. 2004;90(2):353–8.
32. Batchelor T, Carson K, O'Neill A, Grossman SA, Alavi J, New P, et al. Treatment of primary CNS lymphoma with methotrexate and deferred radiotherapy: a report of NABTT 96-07. J Clin Oncol. 2003;21(6):1044–9.
33. Rubenstein JL, Hsi ED, Johnson JL, Jung SH, Nakashima MO, Grant B, et al. Intensive chemotherapy and immunotherapy in patients with newly diagnosed primary CNS lymphoma: CALGB 50202 (Alliance 50202). J Clin Oncol. 2013;31(25):3061–8.
34. Morris PG, Correa DD, Yahalom J, Raizer JJ, Schiff D, Grant B, et al. Rituximab, methotrexate, procarbazine, and vincristine followed by consolidation reduced-dose whole-brain radiotherapy and cytarabine in newly diagnosed primary CNS lymphoma: final results and long-term outcome. J Clin Oncol. 2013;31(31):3971–9.
35. Ferreri AJ, Cwynarski K, Pulczynski E, et al. Chemoimmunotherapy with methotrexate, cytarabine, thiotepa, and rituximab (MATRix regimen) in patients with primary CNS lymphoma: results of the first randomisation of the International Extranodal Lymphoma Study Group-32 (IELSG32) phase 2 trial. Lancet Haematol. 2016;3:e217–27.
36. Ferreri AJ, Reni M, Foppoli M, et al. High-dose cytarabine plus high-dose methotrexate versus high-dose methotrexate alone in patients with primary CNS lymphoma: a randomised phase 2 trial. Lancet. 2009;374:1512–20.
37. Abla O, Branson H, Hawkins C, Weitzman S. Rapid early response to high-dose methotrexate in an adolescent with multifocal primary CNS lymphoma. 12th international conference on malignant lymphoma – Lugano, Switzerland, June 18–21, 2013.
38. O'Suoji C, Welch JJ, Perkins SL, et al. Rare pediatric non-Hodgkin lymphomas: a report from Children's oncology group study ANHL 04B1. Pediatr Blood Cancer. 2016;63:794–800.

39. Khan RB, Shi W, Thaler HT, DeAngelis LM, Abrey LE. Is intrathecal methotrexate necessary in the treatment of primary CNS lymphoma? J Neuro-Oncol. 2002;58(2):175–8.
40. Sierra Del Rio M, Ricard D, Houillier C, Navarro S, Gonzalez-Aguilar A, Idbaih A, et al. Prophylactic intrathecal chemotherapy in primary CNS lymphoma. J Neuro-Oncol. 2012;106(1):143–6.
41. Batchelor TT, Grossman SA, Mikkelsen T, Ye X, Desideri S, Lesser GJ. Rituximab monotherapy for patients with recurrent primary CNS lymphoma. Neurology. 2011;76(10):929–30.
42. Holdhoff M, Ambady P, Abdelaziz A, Sarai G, Bonekamp D, Blakeley J, et al. High-dose methotrexate with or without rituximab in newly diagnosed primary CNS lymphoma. Neurology. 2014;83(3):235–9.
43. Minard-Colin V, Auperin A, Pillon M, et al. Results of the randomized Intergroup trial Inter-B-NHL Ritux 2010 for children and adolescents with high-risk B-cell non-Hodgkin lymphoma (B-NHL) and mature acute leukemia (B-AL): evaluation of Rituximab (R) efficacy in addition to standard LMB chemotherapy (CT) regimen. J Clin Oncol. 2016;34. Abstract 10507.
44. Illerhaus G, Fritsch K, Egerer G, et al. Sequential high dose immuno-chemotherapy followed by autologous peripheral blood stem cell transplantation for patients wtih untreated primary central nervous system lymphoma – a Multicentre Study by the Collaborative PCNSL Study Group Freiburg. Blood 2012, 120. Presented at the American Society of Hematology annual meeting, Atlanta, 2012.
45. Ferreri AJM, Cwynarski K, Pulczynski E, Fox CP, Schorb E, La Rosée P, et al. Whole-brain radiotherapy or autologous stem-cell transplantation as consolidation strategies after high-dose methotrexate-based chemoimmunotherapy in patients with primary CNS lymphoma: results of the second randomisation of the International Extranodal Lymphoma Study Group-32 phase 2 trial. Lancet Haematol. 2017;4(11):e510–23.
46. Rubenstein JL, Formaker P, Wang X, et al. Lenalidomide is highly active in recurrent CNS lymphomas: phase I investigation of lenalidomide plus rituximab and outcomes of lenalidomide as maintenance monotherapy. Hematol Oncol. 2015;33:175. (Abstr 137).

Rare B-Cell Non-Hodgkin's Lymphomas in Childhood and Adolescence

19

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Introduction

Non-Hodgkin's lymphoma (NHL) may arise in tissues with no concomitant infiltration of the lymph nodes, spleen, or thymus; these cases are referred to as primary extra-nodal NHL. It has been reported that 30–50% of B-cell NHL (B-NHL) cases have extra-nodal manifestations in various organs, such as the gastric tract, skin, bone, central nervous system, breast, heart, liver, kidneys, or adrenal glands [1]. Primary extra-nodal B-NHL are very rare. Below, primary pediatric B-NHL in different organs, as well as rare adult-type manifestations, will be described.

Primary Renal Non-Hodgkin's Lymphoma

Epidemiology

Renal involvement is a common finding in patients with advanced stage NHL; however, primary renal NHL is a rare entity, with as few as 70 cases reported in the literature. Primary renal NHL is very uncommon in the pediatric population [2].

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Etiology

The precise etiology of primary renal NHL is unknown, however, there are suggestions that it originates in the lymph nodes of the renal sinus or in the lymphatic network of the renal capsule [3]. In adult patients, primary renal NHL often involves one kidney, whereas in pediatric patients cases can be bilateral [4–7].

Clinical Manifestations

Malbrain et al. [8] proposed criteria for primary renal lymphoma: (a) renal failure as initial presentation, (b) bilateral enlargement of kidneys without obstruction and/or other organ or nodal involvement, (c) diagnosis made by renal biopsy, (d) absence of other causes of renal failure, and (e) rapid improvement of renal function after therapy. More recently, Stallone et al. [9] proposed revised criteria: primary renal NHL is considered if (a) there is lymphomatous renal infiltration, (b) there is non-obstructive uni- or bilateral renal enlargement, and (c) there is no extra-renal localization of lymphoma at the time of diagnosis. Patients commonly present with abdominal mass, gross hematuria, high blood pressure, renal failure, flank pain, and weight loss. Flank pain has been described as one of the most common symptoms of primary renal NHL, at least in adults [10]. In the pediatric cases reported, renal failure is a very common feature [11].

Diagnosis

Complete staging is essential to exclude more disseminated disease. [18F] fluorodeoxyglucose (FDG) positron emission tomography (PET) scan (FDG-PET) has been used in staging for adult and pediatric cases [6, 12]. In adults with Burkitt's lymphoma, FDG-PET has been shown to increase the accuracy of staging [13]. Pediatric studies on the role of

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FDG-PET in staging of B-NHL overall are lacking. The associated finding of thrombus in the inferior vena cava has been reported and careful evaluation is required to differentiate primary renal lymphoma from other more common renal tumors in childhood leading to thrombosis, as well [14]. Renal biopsy is required to confirm the diagnosis of primary renal NHL. Reported histologies are, as expected, Burkitt's lymphoma and diffuse large B-cell NHL (DLBCL). Even rarer, T-cell NHL as the cause of primary renal lymphoma of childhood has been reported [15].

Management

Treatment appears unsatisfactory with the majority of adult patients experiencing extra-renal progression despite standard B-NHL therapy and where reported, death within 1 year of diagnosis is seen in three-quarters of the patients [16]. Optimal therapy in childhood primary renal lymphoma is unknown, though treatment-related mortality due to renal dysfunction is reported [11].

Primary Effusion Lymphoma

Epidemiology

Primary effusion lymphoma (PEL) is a rare type of NHL that often presents with malignant effusions without bulky tumor disease. PEL is the only effusion-based lymphoma listed in the 2016 World Health Organization classification of tumors of hematopoietic tissues [17]. Most patients affected are human immunodeficiency virus (HIV)-positive young adults [18]. The presence of human herpes virus 8 (HHV8) infection is crucial for the diagnosis of PEL [18]. PELs are exceptionally rare in children, with very few reported cases of pediatric "primary pleural lymphoma" in the literature [19, 20].

Diagnosis

The diagnosis of PEL and PEL-like lymphomas is confirmed following examination of the effusion fluid for cytology and immunophenotyping. PEL cells commonly express CD45, CD138, and MUM-1 but usually lack B-cell markers (CD19, CD20, CD79a, surface and cytoplasmic immunoglobulin) and T-cell antigens (CD2, CD3, CD4, CD5, CD7, and CD8). Diagnosis of pediatric PEL-like lymphoma often poses diagnostic challenges. Differential diagnoses in children presenting with lymphomatous effusion include DLBCL, Burkitt's lymphoma, anaplastic large cell lymphoma (ALCL), and pyothorax-associated lymphoma. Increased diagnostic awareness, better understanding of molecular biology, and more effective treatment strategies are needed for these rare lymphomas.

Management

Given the rarity of these lymphomas, there is no specific treatment recommendation for children. The most common chemotherapy regimen used in adult patients with PEL is CHOP-based therapy (cyclophosphamide, doxorubicin, vincristine, and prednisolone) with adult data of only 6 months median survival and overall survival (OS) of 40% [21]. Use of novel therapies such as interferon-α [22], cidofovir [23], and bortezomib [24] has been supported by case reports and small case series. Reports in the literature suggest that disease control could be achieved without any systemic chemotherapy, by drainage of the effusion alone [24, 25].

Primary Cutaneous B-Cell Lymphomas

Primary cutaneous lymphomas are exceptionally rare in children. The T-cell subtypes have similar clinical and pathologic features as their adult counterparts. The outcome of cutaneous B-cell lymphomas depends on subtype; however, there are insufficient data on the respective entities. Prospective clinical studies done on the pediatric population with these rare lymphomas are needed to better understand their biological and clinical behavior and to ultimately discover the best therapeutic strategies. Herein we will focus on pediatric cutaneous B-cell lymphomas only.

Primary cutaneous marginal zone B-cell lymphoma (*PCMZL*) is an indolent B-cell lymphoma which often clinically presents with solitary or multifocal skin lesions, which include violaceous papules, plaques, or nodules, involving the trunk or extremities [26]. Pediatric PCMZL is commonly seen in teenagers, frequently with lesions involving more than one anatomical site, often associated with immunodeficiency [27]. Association with *Borrelia burgdorferi* is rare, but seen.

Pediatric PCMZL is morphologically and immunophenotypically similar to adult cases, with the marginal zone B-cells expressing CD20, CD79a, and bcl-2, but negative for CD10 and bcl-6. This phenotype is useful in distinguishing PCMZL from primary cutaneous follicular center lymphoma (PCFCL). Treatment options vary depending on the extent of the disease. For patients presenting with one or few small skin lesions treatment could be a wait-and-watch approach, surgery, or local radiotherapy. The use of intralesional rituximab in both, adults and children, has shown very promising results [28]. In patients with associated *B. burgdorferi* infection, systemic or oral antibiotics (doxycycline) can be tried first [29]. In patients showing frequent skin relapses, topical or intralesional steroids may be considered. Overall, pediatric PCMZL, similar to its adult counterpart, usually follows an indolent course and has excellent prognosis.

Primary cutaneous follicle center lymphoma (PCFCL) has a distinctive clinical presentation with solitary or grouped

plaques and tumors, mainly involving the scalp, forehead, or the trunk [30, 31]. The tumor cells express the B-cell markers bcl-6 and CD10 (variable) but are negative for MUM-1/IRF-4, consistent with its origin from germinal center B-cells. A common feature is the absence of t(14;18) [32]. PCFCL in children is exceptionally rare, with only few pediatric cases reported in the literature [33, 34]. Due to its rarity, it is unclear if pediatric PCFCL has a worse prognosis compared to its adult counterpart. The current recommended treatment approach is complete excision alone, deferring radiation for relapsed disease.

Primary cutaneous DLBCL, leg type, is a rare and aggressive variant of DLBCLs, presenting with skin lesions on the legs in elderly patients [35]. To date, there are no reported pediatric cases of primary cutaneous DLBCL, leg type.

Plasmablastic lymphoma is a rare subtype of DLBCL commonly affecting the oral cavity of HIV-positive or post-transplant patients. The skin is rarely involved [36]. There is an established association with EBV. In adult patients, the disease is characterized by an aggressive course, with a mean survival of 6 months. Reports of pediatric cases of plasmablastic lymphoma are rare [37, 38].

Lymphomatoid Granulomatosis

A rare angiocentric and angioinvasive EBV-associated B-cell lymphoproliferative disorder, lymphomatoid granulomatosis, was first described in 40 patients by Liebow in 1972 [39]. The disease predominantly involved the lungs with 20% of patients also having central nervous system (CNS) involvement. Among the 40 patients, there was one 8.5-year-old female child who presented with neurological symptoms of ataxia and unilateral loss of vision and who was found to have bilateral lung lesions and subsequent biopsy confirmed lymphomatoid granulomatosis. The neurological symptoms spontaneously improved, but the lung lesions were treated with prednisolone and proved responsive, but she remained on daily treatment at the time of the report [39]. The association of lymphomatoid granulomatosis with EBV and classification as a B-cell lymphoproliferative disorder took a further two decades following the first description [40].

There have been few cases of childhood lymphomatoid granulomatosis reported since 1972. A case report and review in 2014 described only 49 cases [41]. Subsequently, only a handful of additional cases have been reported [42, 43]. A median age of 12 years was found and in keeping with the first case reported, pulmonary and CNS manifestations are predominating. One-third of the patients were immunocompromised. Interleukin-2-inducible T-cell kinase (ITK) deficiency has been found in one 6-year-old girl who developed lymphomatoid granulomatosis and died after hematopoietic stem cell transplantation [44]. The prognosis appears to be poor and in the absence of trial evidence, aggressive treatment with B-NHL therapy appears warranted [41]. The role of rituximab remains unclear [45].

Primary Lymphomas of the Eye in Children

Primary lymphomas of the eye in children consist of the primary ocular adnexal lymphomas (POAL) and primary intraocular lymphomas (PIOL), affecting the ocular adnexa and the vitreous/retina, respectively.

Epidemiology

POAL

Primary ocular adnexal lymphoma (POAL) is the most common type of cancer affecting the ocular adnexa, including the orbit, lacrimal gland/sac, conjunctiva, and eyelids, accounting for 55% of all orbital tumors. POAL accounts for 1–2% of NHL and 5–15% of extra-nodal lymphoma [46, 47].

Extra-nodal marginal zone lymphoma of mucosa-associated lymphoid tissue (EMZL-MALT) is the most common histological subtype, accounting for approximately 57% of POAL cases followed by follicular lymphoma (15%), DLBCL (13%), and mantle cell lymphoma (4%) [47]. Regarding EMZL-MALT, the best-studied subtype, there is a slight female predominance and the median age at diagnosis is nearly 65 years. Rare cases of POAL in children have been described, however, the exact incidence in childhood is not known. Interestingly, the EMZL-MALT subtype is strongly associated with *Chlamydophila psittaci* infection in certain geographic areas as well as with co-existing autoimmune diseases, implying the involvement of chronic antigenic stimulation and chronic inflammation in the pathogenesis of these tumors.

PIOL

Primary intraocular lymphoma (PIOL), the majority of which is DLBCL, is a rare entity in both the adult and childhood population [48, 49]. The limited data on pediatric patients reported are usually part of adult cohort studies [48].

Clinical Manifestations

POAL

The clinical manifestations of POALs vary depending on the anatomical site of tumor origin and the extension of the disease. The most common clinical features of POALs are listed in the Table 19.1. Bilateral involvement ranges from 7% to 24% [46].

Table 19.1 Clinical manifestations of POALs

Site of involvement	Relative Incidence [50, 51]	Most common clinical manifestations
Orbit	37–40%	Proptosis, periorbital edema, ptosis, exophthalmos, diplopia, visual loss, eye movement disorder
Conjunctiva	29–40%	"Salmon patch"-like conjunctival lesions
Lacrimal apparatus	10–20%	Painless palpable mass, periorbital edema, gritty sensation, epiphora
Eyelids	10–14%	Mass/swelling of the eyelid

PIOL

Clinical presentation of primary intraocular DLBCL poses diagnostic challenges. Commonly, the initial symptoms are benign and nonspecific, such as floaters or blurred vision, often mistaken and treated as chronic uveitis, resulting in late diagnosis [52, 53]. Patients can have symptoms of CNS involvement, which can develop at any stage during disease progression. Neurological findings include hemiparesis and cerebellar signs [54].

Diagnosis

Ophthalmological examination and imaging studies (orbit ultrasonography and contrast MRI) are required in order to delineate the extent of ocular disease, but they are unable to define the exact pathology of a mass. Therefore, surgical biopsy of the suspicious lesion for morphological, immunophenotypical, and molecular studies is the gold standard for the diagnosis. Further radiological investigation and additional examinations (BM and CSF examination, total body imaging, PET scans) are required as indicated for the staging [47].

Management

The therapeutic approach of primary lymphomas of the eye includes chemotherapy, radiotherapy, complete excision of the lesion, and/or immunotherapy (anti-CD20 monoclonal antibody intravenously or intralesionally), depending on the sites of involvement, the histology, and the stage of disease.

POAL

Radiation therapy (RT) is currently the primary treatment modality for EMZL-MALT, achieving local control in 86–100% of Ann Arbor stage I cases. Additionally, it is shown in several studies that an antibiotic treatment with doxycycline in cases of *C. psittaci*-positive extra-nodal marginal zone lymphomas of MALT is able to achieve an overall response rate in 45–65% cases without additional therapy [46].

PIOL

Systemic chemotherapy is the treatment of choice in PIOLs, however, many drugs do not penetrate the eye well, and they need to be augmented with intravitreal chemotherapy or external beam radiation. Studies have proven that anti-CD20 monoclonal antibodies are efficacious in the treatment of ocular lymphoma, and the addition of such agents has become standard of care, at least in adults.

Prognosis

POAL

Histological subtype and stage are the most important prognostic factors for POALs. Although the overall response rate of early-stage POALs with current therapy is above 80%, local and systemic relapses occur continuously over time. Desai et al. reported an estimated cumulative incidence of relapse or progression of stage I EMZL-MALT of 5.1% at 1 year, 17.5% at 5 years, and 31% at 10 years [46]. Similarly, distant relapse occurs in 6–50% of follicular lymphoma at a median follow-up of 7 years, while the 10-year OS rate is 83% [50]. In conclusion, surveillance for local or systemic lymphoma is required for a long period of time.

PIOL

Primary intraocular DLBCL is a high-grade, aggressive lymphoma, but is potentially curable [49]. In adult patients, the degree of p53 expression and Ki-67 antigen expression has been shown to correlate significantly with clinical outcome [55]; however, no studies have been done to confirm if this is also true for the pediatric population. Survival in pediatric patients depends on whether the disease is initially localized or metastatic; however, survival is largely unknown due to the limited data on these rare pediatric lymphomas.

Primary Non-Hodgkin's Lymphoma of the Bone in Children

Epidemiology

Primary NHL of the bone is a rare malignancy which comprises 2–3% (5–7% Borst et al. 2013) of primary bone tumors and 5% of extra-nodal NHL in adults and children. The median age at presentation lies in the fifth decade; however, rare cases of primary NHL of the bone in children are also reported, usually presenting in adolescence and accounting for 2–9% of all pediatric NHL. It has a predominance in males, both in adults and children [56].

Histology

Most cases are mature B-NHLs. Although histology is more heterogeneous in children than in adults, DLBCL remains the most common pediatric histological subtype. Precursor B-cell lymphoblastic lymphoma, ALCL, follicular lymphoma, and Burkitt's lymphoma have also been described [56, 57].

Clinical Manifestations

Clinical features include localized bone pain without antecedent trauma, swelling or palpable mass, tenderness, nocturnal pain, limp, pathologic fracture, and constitutional symptoms (fever, weight loss, anorexia, etc.). The femur is the most frequently involved site followed by the pelvis, humerus, and tibia. Patients may present with single or multiple bone involvement or disseminated disease. Moreover, a median delay of several months from the onset of symptoms until the final diagnosis has been reported, which is attributed to the nonspecific symptoms/signs and radiological findings at presentation [56].

Diagnosis

Bone biopsy remains the gold standard in diagnosis. Hypercalcemia is often associated with primary NHL of the bone [56]. Radiographic findings of plain radiographs, CT or MRI are generally nonspecific and are often unable to differentiate between this rare lymphoma subtype and other benign or malignant entities. The most common radiological appearance is a permeable or moth-eaten lytic destructive pattern. Cortical destruction, pathologic fractures, soft-tissue masses, and periosteal reaction may also be present. MRI is the most sensitive modality for detecting a soft-tissue mass [58]. In addition, based on the current literature, PET/CT is suggested for pre-treatment staging and monitoring, since it is more sensitive and specific than CT alone for the detection of extranodal spread of lymphoma [59]. Radiological assessment of the involved sites during and after therapy remains challenging in patients with primary NHL of the bone due to the difficulty in distinguishing between residual lymphoma and osseous remodeling, fibrosis, or necrotic tumor. All imaging modalities may show bone abnormalities even for years after clinical remission resulting in high false-positive rate [60].

Management

Chemotherapy alone or chemotherapy combined with radiotherapy is the treatment of choice [56]. The overall response rate to initial treatment in adults and children ranges from 56% to 95%.

Prognosis

Female gender, age < 9 years, non-large cell histology, BM involvement, and elevated calcium levels appear to have poor prognostic impact on survival. Overall, the prognosis of primary NHL of the bone in children is favorable. Previous studies have shown a survival rate of 40–100%. Moreover, the event-free survival rate for localized disease is 75–100% and for disseminated disease is 25–71% [56].

Primary Gastric Lymphoma (PGL) in Children

Epidemiology

Primary gastric lymphomas (PGL) are very rare in children. They account for less than 2% of NHL in childhood, as shown in a single center study [61], and comprise 2–9% of all primary gastrointestinal lymphomas (PGITLs) [62, 63]. Based on the SEER database, gastric lymphomas occur more commonly in children older than 10 years than in the younger age group (14.1% versus 3.4%). The overwhelming majority of PGLs reported occurred in males, in accordance with the overall male predominance described in pediatric PGITL [62].

Histology

The majority of PGLs are high-grade NHLs of B-cell origin, with Burkitt's lymphoma being the predominant variant. Other histological entities that have been described in literature are DLBCL and mucosa-associated lymphoid tissue (MALT) lymphomas. Although not yet established in pediatric cases, the association between MALT lymphomas and *H. pylori* infection is well documented in adults, suggesting that specific *H. pylori* antigens stimulate lymphomagenesis [64].

Clinical Manifestations

The clinical manifestations of PGL in childhood based on cases reported in literature are listed in Table 19.2. The symptoms usually precede the diagnosis several weeks or months. However, because of the low suspicion index, the diagnosis could be significantly delayed.

Diagnosis

Ultrasound, CT/MRI imaging, or upper gastrointestinal series are commonly used for the initial investigation of the

Table 19.2 Clinical manifestations of PGL in childhood

Epigastric or abdominal pain
Nausea, vomiting
Hematemesis, hematochezia, melena
Dyspepsia
Palpable mass in epigastrium or upper abdomen
Distended abdomen
Poor appetite
Malaise, fatigue
Weight loss
Iron deficiency anemia
Gastrointestinal obstruction (constipation, jaundice)

presenting symptoms, but the mainstay of diagnosis of PGLs is the upper gastrointestinal endoscopy combined with biopsy sampling. Further investigation for *H. pylori* detection and staging is required.

Management

The management of PGLs includes surgery, chemotherapy, and radiotherapy depending mainly on the presenting symptoms, the stage and histological type of the lymphoma [62]. Moreover, *H. pylori* eradication therapy alone has been shown to achieve 60–100% histological remission rates in *H. pylori*-positive early-stage gastric MALT lymphoma in adults, as well as in other histological types [64]. Similarly, *H. pylori* eradication therapy has been used alone or combined with other therapeutic modalities in pediatric cases, too.

Prognosis

Prognostic factors include the histological type and stage of the disease. Gastric lymphomas have the worst prognosis among pediatric PGITL, based on the SEER database, with a 10-year overall and disease-specific survival of 64%. Prognosis seems to be more favorable in older children (>10 years) [62].

Rare Adult-Type Malignant B-Cell Manifestations in Children

Leukemic mature B-cell malignancies, such as chronic lymphocytic leukemia (CLL), multiple myeloma (MM), and prolymphocytic leukemia (PLL), are diseases of the elderly. Pediatric cases are extremely rare. Herein we describe epidemiology, clinical manifestations, and management of the three types and what is known in children.

Chronic Lymphocytic Leukemia in Children

Epidemiology

Although CLL is the most common leukemia in adults in Western countries with a median age at diagnosis of 70 years and an incidence rate of 4.7 cases per 100,000 per year [65], pediatric cases of CLLs are extremely rare. According to the SEER database from 1974 to 2004, 21,058 cases with CLL and small lymphocytic lymphoma (SLL) are recorded, with only 20 patients registered below the age of 25 years [66].

Diagnosis

The diagnosis of CLL is based on detection of elevated monoclonal mature-appearing malignant B lymphocytes (>5.000/μl in peripheral blood), expressing CD19, CD20, CD5, and CD23.

Clinical Manifestations

The clinical presentation and course of CLL varies widely. Patients could be asymptomatic at diagnosis or present with active disease including constitutional or B-symptoms (fatigue, night sweat, fever, weight loss), progressive lymphocytosis, autoimmune cytopenias (anemia and/or thrombocytopenia), lymphadenopathy, hepatosplenomegaly, and recurrent infections. Small lymphocytic lymphoma is considered to be a less frequent variation of the same disease and resembles a low- grade NHL. Patients with SLL exhibit predominately lymph node disease, but lack a leukemic phase, despite BM involvement [65, 67]. Interestingly, in the study of Dores et al., cases with SLL seemed to be more common than CLL under the age of 35 years. In the age group <25 years 16 patients were diagnosed with SLL and only 4 with CLL [66].

Management

Since pediatric cases of CLL are rare, the management of the disease is based on the therapeutic strategies applied in adults (Table 19.3) [65]. The prognosis is overall favorable.

Table 19.3 Therapeutic approaches in CLL

Watch-and-wait (in asymptomatic cases)
Chemo-immunotherapy (FCR, fludarabine, cyclophosphamide, anti-CD20 antibody rituximab, or BR, bendamustine, anti-CD20 rituximab)
Low-intensity regimens (for older patients with poor performance status)
Novel targeted agents (BTK inhibitor ibrutinib, PI3Kδ inhibitor idelalisib, apoptosis regulator BCL-2 antagonist venetoclax)
Allo-HSCT
CAR-T cell therapy

Multiple Myeloma in Childhood

Epidemiology

Multiple myeloma (MM), also known as plasma cell myeloma, is a B-cell-based malignancy characterized by the clonal proliferation of plasma cells in BM, producing a monoclonal immunoglobulin. It is the second most common hematological malignancy, with an incidence of 6 cases per 100.000 per year [68]. The median age at diagnosis is approximately 70 years, while only 0.3% of the cases are younger than 30 years. Pediatric MM is extremely rare [69]. Approximately 30 cases of MM have been reported in literature in patients younger than 15 years of age, although some of these diagnoses were based on older technologies and diagnostic criteria [70].

Clinical Manifestations

The clinical manifestations in patients under 30 years are similar to those of adults and include anemia, infections, osteolytic lesions, bone pain, osteopenia bone disease, pathologic fractures, renal insufficiency, or hypercalcemia [71].

Management

Since cases of MM in childhood are rare, the management of the disease is based on current therapeutic strategies of adults. However, pediatric patients are more likely to be eligible to high-dose chemotherapies or stem cell transplantation due to their better performance status than the majority of adult patients. Novel agents, such as bortezomib have already been used combined with other agents as first-line therapy in pediatric MM [70].

Prognosis

The survival of patients (<30 years) appears to be better than that of patients of all ages with MM [71].

B-Cell Prolymphocytic Leukemia

B-cell prolymphocytic leukemia (PLL) is a rare mature B-cell malignancy in adults which typically present with excessive lymphocytosis, splenomegaly, and B-symptoms. Only two cases of T-cell PLL in childhood have been described, no pediatric B-cell PLL patients are reported in literature yet [72].

References

1. Airaghi L, et al. Unusual presentation of large B cell lymphoma: a case report and review of literature. Clin Lab Haematol. 2006;28:338–42. https://doi.org/10.1111/j.1365-2257.2006.00816.x.
2. Tefekli A, Baykal M, Binbay M, Barut M, Muslumanoglu AY. Lymphoma of the kidney: primary or initial manifestation of rapidly progressive systemic disease? Int Urol Nephrol. 2006;38:775–8. https://doi.org/10.1007/s11255-005-4030-7.
3. Torrecilla Garcia-Ripoll JR, et al. Primary renal lymphoma. Actas Urol Esp. 2003;27:555–8.
4. Ahmad AH, Maclennan GT, Listinsky C. Primary renal lymphoma: a rare neoplasm that may present as a primary renal mass. J Urol. 2005;173:239. https://doi.org/10.1097/01.ju.0000148570.46368.c0.
5. Baran A, Küpeli S, Doğru Ö. A pediatric renal lymphoma case presenting with central nervous system findings. Turkish Journal of Hematology. 2013;30:191–3. https://doi.org/10.4274/Tjh.03164.
6. Dhull VS, et al. Bilateral primary renal lymphoma in a pediatric patient: staging and response evaluation with (1)(8)F-FDG PET/CT. Rev Esp Med Nucl Imagen Mol. 2015;34:49–52. https://doi.org/10.1016/j.remn.2014.05.004.
7. Karadeniz C, et al. Primary renal lymphoma and xanthogranulomatous pyelonephritis in childhood. J Nephrol. 2002;15:597–600.
8. Malbrain ML, et al. Acute renal failure due to bilateral lymphomatous infiltrates. Primary extranodal non-Hodgkin's lymphoma (p-EN-NHL) of the kidneys: does it really exist? Clin Nephrol. 1994;42:163–9.
9. Stallone G, et al. Primary renal lymphoma does exist: case report and review of the literature. J Nephrol. 2000;13:367–72.
10. Okuno SH, Hoyer JD, Ristow K, Witzig TE. Primary renal non-Hodgkin's lymphoma. An unusual extranodal site. Cancer. 1995;75:2258–61.
11. Sieniawska M, Bialasik D, Jedrzejowski A, Sopylo B, Maldyk J. Bilateral primary renal Burkitt lymphoma in a child presenting with acute renal failure. Nephrol Dial Transplant. 1997;12:1490–2. https://doi.org/10.1093/ndt/12.7.1490.
12. Hagihara M, Hua J, Iwaki Y, Inoue M, Sato T. Primary renal lymphoma: a case report and literature review. Intern Med. 2015;54:2655–9. https://doi.org/10.2169/internalmedicine.54.3368.
13. Estrella C-C, et al. Role of 18F-FDG-PET/CT in the management of Burkitt lymphoma. Eur J Haematol. 2015;94:23–30. https://doi.org/10.1111/ejh.12284.
14. Kumar D, et al. Pediatric renal non-hodgkin lymphoma with inferior vena cava thrombosis. J Pediatr Hematol Oncol. 2010;32:147–9. https://doi.org/10.1097/MPH.0b013e3181bdbc78.
15. Paladugu S, Garro R, Schrijver I, Kambham N, Higgins JPT. A 30-month-old child with acute renal failure due to primary renal cytotoxic T-cell lymphoma. Am J Surg Pathol. 2010;34:1066–70. https://doi.org/10.1097/PAS.0b013e3181de693c.
16. Shetty S, Singh AC, Babu V. Primary renal lymphoma - a case report and review of literature. J Clin Diagn Res. 2016;10:XD05–7. https://doi.org/10.7860/JCDR/2016/20901.8577.
17. Swerdlow SH, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127:2375–90.
18. Simonelli C, et al. Clinical features and outcome of primary effusion lymphoma in HIV-infected patients: a single-institution study. J Clin Oncol. 2003;21:3948–54. https://doi.org/10.1200/jco.2003.06.013.
19. Karadeniz C, et al. Primary pleural lymphoma: an unusual presentation of childhood non-Hodgkin lymphoma. Pediatr Hematol Oncol. 2000;17:695–9. https://doi.org/10.1080/08880010050211411.
20. Ravikumar G, Tirumalae R, Das K. Primary pleural non-Hodgkin lymphoma in a child--an exceedingly rare disease.

J Pediatr Surg. 2012;47:e29–31. https://doi.org/10.1016/j.jpedsurg.2011.11.042.
21. Okada S, Goto H, Yotsumoto M. Current status of treatment for primary effusion lymphoma. Intractable Rare Dis Res. 2014;3:65–74.
22. Boulanger E, et al. Prognostic factors and outcome of human herpesvirus 8-associated primary effusion lymphoma in patients with AIDS. J Clin Oncol. 2005;23:4372–80. https://doi.org/10.1200/jco.2005.07.084.
23. Luppi M, et al. Treatment of herpesvirus associated primary effusion lymphoma with intracavity cidofovir. Leukemia. 2005;19:473–6. https://doi.org/10.1038/sj.leu.2403646.
24. Siddiqi T, Joyce RM. A case of HIV-negative primary effusion lymphoma treated with bortezomib, pegylated liposomal doxorubicin, and rituximab. Clin Lymphoma Myeloma. 2008;8:300–4. https://doi.org/10.3816/CLM.2008.n.042.
25. Terasaki Y, et al. Disappearance of malignant cells by effusion drainage alone in two patients with HHV-8-unrelated HIV-negative primary effusion lymphoma-like lymphoma. Int J Hematol. 2011;94:279–84. https://doi.org/10.1007/s12185-011-0906-8.
26. Servitje O, et al. Primary cutaneous marginal zone B-cell lymphoma: a clinical, histopathological, immunophenotypic and molecular genetic study of 22 cases. Br J Dermatol. 2002;147:1147–58.
27. Kimura H, et al. EBV-associated T/NK-cell lymphoproliferative diseases in nonimmunocompromised hosts: prospective analysis of 108 cases. Blood. 2012;119:673–86. https://doi.org/10.1182/blood-2011-10-381921.
28. Soda R, et al. Systemic therapy of primary cutaneous B-cell lymphoma, marginal zone type, with rituximab, a chimeric anti-CD20 monoclonal antibody. Acta Derm Venereol. 2001;81:207–8.
29. Senff NJ, et al. European Organization for Research and Treatment of Cancer and International Society for Cutaneous Lymphoma consensus recommendations for the management of cutaneous B-cell lymphomas. Blood. 2008;112:1600–9. https://doi.org/10.1182/blood-2008-04-152850.
30. Kempf W, Denisjuk N, Kerl K, Cozzio A, Sander C. Primary cutaneous B-cell lymphomas. J Dtsch Dermatol Ges. 2012;10:12–22; quiz 23. https://doi.org/10.1111/j.1610-0387.2011.07852.x.
31. Kempf W, Sander CA. Classification of cutaneous lymphomas - an update. Histopathology. 2010;56:57–70. https://doi.org/10.1111/j.1365-2559.2009.03455.x.
32. Abdul-Wahab A, et al. Chromosomal anomalies in primary cutaneous follicle center cell lymphoma do not portend a poor prognosis. J Am Acad Dermatol. 2014;70:1010–20. https://doi.org/10.1016/j.jaad.2014.01.862.
33. Ghislanzoni M, Gambini D, Perrone T, Alessi E, Berti E. Primary cutaneous follicular center cell lymphoma of the nose with maxillary sinus involvement in a pediatric patient. J Am Acad Dermatol. 2005;52:S73–5. https://doi.org/10.1016/j.jaad.2004.05.025.
34. Amitay-Laish I, et al. Juvenile onset of primary low-grade cutaneous B-cell lymphoma. Br J Dermatol. 2009;161:140–7. https://doi.org/10.1111/j.1365-2133.2009.09121.x.
35. Willemze R. Primary cutaneous B-cell lymphoma: classification and treatment. Curr Opin Oncol. 2006;18:425–31. https://doi.org/10.1097/01.cco.0000239879.31463.42.
36. Black CL, Foster-Smith E, Lewis ID, Faull RJ, Sidhu SK. Post-transplant plasmablastic lymphoma of the skin. Australas J Dermatol. 2013;54:277–82. https://doi.org/10.1111/j.1440-0960.2012.00939.x.
37. Hernandez C, Cetner AS, Wiley EL. Cutaneous presentation of plasmablastic post-transplant lymphoproliferative disorder in a 14-month-old. Pediatr Dermatol. 2009;26:713–6. https://doi.org/10.1111/j.1525-1470.2009.01019.x.
38. Apichai S, et al. Multifocal cutaneous and systemic plasmablastic lymphoma in an infant with combined living donor small bowel and liver transplant. Pediatr Transplant. 2009;13:628–31. https://doi.org/10.1111/j.1399-3046.2008.01026.x.
39. Liebow AA, Carrington CRB, Friedman PJ. Lymphomatoid granulomatosis. Hum Pathol. 1972;3:457–558. https://doi.org/10.1016/S0046-8177(72)80005-4.
40. Guinee D Jr, et al. Pulmonary lymphomatoid granulomatosis. Evidence for a proliferation of Epstein-Barr virus infected B-lymphocytes with a prominent T-cell component and vasculitis. Am J Surg Pathol. 1994;18(8):753.
41. Tacke ZC, et al. Childhood lymphomatoid granulomatosis: a report of 2 cases and review of the literature. J Pediatr Hematol Oncol. 2014; https://doi.org/10.1097/MPH.0000000000000090.
42. Lee S, Kang M-J, Kim H-J, Kim H-B, Lee J. Pulmonary lymphomatoid granulomatosis in a 4-year-old girl. Pediatr Radiol. 2015;45:1082–5. https://doi.org/10.1007/s00247-014-3233-4.
43. Aboobaker S, Tinley C. Primary orbital lymphomatoid granulomatosis in a 1-year-old child. Middle East Afr J Ophthalmol. 2015;22:517–9.
44. Ghosh S, Bienemann K, Boztug K, Borkhardt A. Interleukin-2-inducible T-Cell Kinase (ITK) deficiency - clinical and molecular aspects. J Clin Immunol. 2014;34:892–9. https://doi.org/10.1007/s10875-014-0110-8.
45. Hernandez-Marques C, et al. Rituximab in lymphomatoid granulomatosis. J Pediatr Hematol Oncol. 2014;36:e69–74. https://doi.org/10.1097/MPH.0b013e31827e63a6.
46. Desai A, et al. Long-term course of patients with primary ocular adnexal MALT lymphoma: a large single-institution cohort study. Blood. 2017;129:324–32.
47. Ponzoni M, et al. A reappraisal of the diagnostic and therapeutic management of uncommon histologies of primary ocular adnexal lymphoma. Oncologist. 2013;18:876–84. https://doi.org/10.1634/theoncologist.2012-0425.
48. Ferry JA, et al. Lymphoma of the ocular adnexa: a study of 353 cases. Am J Surg Pathol. 2007;31:170–84. https://doi.org/10.1097/01.pas.0000213350.49767.46.
49. Sjö LD, et al. Increasing incidence of ophthalmic lymphoma in Denmark from 1980 to 2005. Invest Ophthalmol Vis Sci. 2008;49:3283–8. https://doi.org/10.1167/iovs.08-1768.
50. Verdijk RM. Lymphoproliferative tumors of the ocular adnexa. Asia Pac J Ophthalmol (Phila). 2017;6:132–42. https://doi.org/10.22608/apo.2016209.
51. Kalogeropoulos D, Papoudou-Bai A, Kanavaros P, Kalogeropoulos C. Ocular adnexal marginal zone lymphoma of mucosa-associated lymphoid tissue. Clin Exp Med. 2018;18:151–63. https://doi.org/10.1007/s10238-017-0474-1.
52. Schalenbourg A, Mantel I. The eye and cancer. Rev Med Suisse. 2015;11:2395–8.
53. Eckardt AM, Lemound J, Rana M, Gellrich N-C. Orbital lymphoma: diagnostic approach and treatment outcome. World J Surg Oncol. 2013;11:73. https://doi.org/10.1186/1477-7819-11-73.
54. Hochberg FH, Miller DC. Primary central nervous system lymphoma. J Neurosurg. 1988;68:835–53. https://doi.org/10.3171/jns.1988.68.6.0835.
55. Auw-Haedrich C, et al. Long term outcome of ocular adnexal lymphoma subtyped according to the REAL classification. Br J Ophthalmol. 2001;85:63–9.
56. Glotzbecker MP, et al. Primary non-Hodgkin's lymphoma of bone in children. J Bone Joint Surg Am. 2006;88:583–94.
57. Maruyama D, et al. Primary bone lymphoma: a new and detailed characterization of 28 patients in a single-institution study. Jpn J Clin Oncol. 2007;37:216–23.
58. Krishnan A, et al. Primary bone lymphoma: radiographic-MR imaging correlation. Radiographics. 2003;23:1371–83. ; discussion 1384-1377. https://doi.org/10.1148/rg.236025056.
59. Milks KS, McLean TW, Anthony EY. Imaging of primary pediatric lymphoma of bone. Pediatr Radiol. 2016;46:1150–7.

60. Borst AJ, States LJ, Reilly AF, Rheingold SR. Determining response and recurrence in pediatric B-cell lymphomas of the bone. Pediatr Blood Cancer. 2013;60:1281–6.
61. Moschovi M, Menegas D, Stefanaki K, Constantinidou CV, Tzortzatou-Stathopoulou F. Primary gastric Burkitt lymphoma in childhood: associated with Helicobacter pylori? Med Pediatr Oncol. 2003;41:444–7.
62. Kassira N, Pedroso FE, Cheung MC, Koniaris LG, Sola JE. Primary gastrointestinal tract lymphoma in the pediatric patient: review of 265 patients from the SEER registry. J Pediatr Surg. 2011;46:1956–64.
63. Bethel CA, Bhattacharyya N, Hutchinson C, Ruymann F, Cooney DR. Alimentary tract malignancies in children. J Pediatr Surg. 1997;32:1004–8. ; discussion 1008-1009.
64. Kuo SH, Cheng AL. Helicobacter pylori and mucosa-associated lymphoid tissue: what's new. Hematology Am Soc Hematol Educ Program. 2013;2013:109–17. https://doi.org/10.1182/asheducation-2013.1.109.
65. Burger JA, O'Brien S. Evolution of CLL treatment - from chemoimmunotherapy to targeted and individualized therapy. Nat Rev Clin Oncol. 2018; https://doi.org/10.1038/s41571-018-0037-8.
66. Dores GM, et al. Chronic lymphocytic leukaemia and small lymphocytic lymphoma: overview of the descriptive epidemiology. Br J Haematol. 2007;139:809–19. https://doi.org/10.1111/j.1365-2141.2007.06856.x.
67. Demir HA, et al. Chronic lymphocytic leukemia in a child: a challenging diagnosis in pediatric oncology practice. Pediatr Blood Cancer. 2014;61:933–5.
68. Rollig C, Knop S, Bornhauser M. Multiple myeloma. Lancet. 2015;385:2197–208. https://doi.org/10.1016/s0140-6736(14)60493-1.
69. Ludwig H, et al. Myeloma in patients younger than age 50 years presents with more favorable features and shows better survival: an analysis of 10 549 patients from the International Myeloma Working Group. Blood. 2008;111:4039–47. https://doi.org/10.1182/blood-2007-03-081018.
70. Crusoe EDQ, et al. Multiple myeloma: a rare case in an 8-year-old child. Clin Lymphoma Myeloma Leuk. 2015;15:e31–3. https://doi.org/10.1016/j.clml.2014.08.004.
71. Blade J, Kyle RA, Greipp PR. Multiple myeloma in patients younger than 30 years. Report of 10 cases and review of the literature. Arch Intern Med. 1996;156:1463–8.
72. Dearden C. Management of prolymphocytic leukemia. Hematology Am Soc Hematol Educ Program. 2015;2015:361–7.

Part VI

Mature T- Non-Hodgkin's Lymphomas

Anaplastic Large Cell Lymphoma in Children and Adolescents

20

Eric J. Lowe and Laurence Brugieres

Introduction

Anaplastic large cell lymphoma (ALCL) is a distinct form of Non-Hodgkin lymphoma (NHL) which accounts for 10–15% of all childhood lymphomas [1–3]. First described in 1985 by Stein et al., ALCL is characterized by the malignant cell expression of CD30 [4]. In the decade following the initial description, researchers identified a translocation involving chromosome 2p in a number of cases [5–7]. This translocation led to the discovery of *anaplastic large cell lymphoma kinase* (*ALK*) oncogene which now defines certain types of ALCL [8]. In fact, the World Health Organization (WHO) now divides ALCL into three separate entities based on this distinct molecular pathway: ALK-positive ALCL (ALK+), ALK-negative ALCL (ALK-), and primary cutaneous ALCL [9]. Overexpression of the anaplastic large cell lymphoma kinase (ALK) protein is the main cause of oncogenesis for the overwhelming majority of pediatric ALCL cases [10, 11]. Hence, any discussion of ALCL in children by definition focuses mainly on the ALK+ subtype of ALCL.

Pediatric ALCL is characterized by advanced disease at presentation with a high incidence of extra-nodal involvement. Current treatment strategies use multi-agent chemotherapy and achieve ~70% event-free survival (EFS) [12–23]. However, biological discoveries and pharmaceutical advances are likely to provide major advances in the next few years. This chapter will review the biology, presentation, and treatment of ALCL in children with a heavy focus on ALK+ ALCL as that is the most common type found in children.

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Pathology

General

ALCL is a peripheral T- or null-cell lymphoma characterized by strong expression of CD30 [24]. No B-cell cases of ALCL exist as currently defined by the WHO [9]. While most ALCLs are positive for CD2 and CD4, other T-cell markers such as CD3, CD5, CD7, and CD8 are commonly negative. The null-cell phenotype is likely composed of T-cells that have lost most of their antigens but still possess similar genetic features of a peripheral T-cell [25]. Most cases of ALCL are positive for epithelial membrane antigen and negative for EBV. Although the definition and description of ALCL has evolved over that past 20 years, the malignant "hallmark cells" that are identified by the eccentric, horseshoe-shaped nuclei have defined the disease [26]. While the "hallmark cells" define ALCL, it is the background cells and ALK staining which define the histological variants.

ALCL can be divided into two entities based on ALK staining: ALK+ ALCL and ALK- ALCL [9, 27]. The ALK- cases of ALCL are more of a diagnostic challenge, and pathologists must rely on the combination of morphology, immunostaining, genetics, and clinical information in order to correctly make a diagnosis. In contrast, when ALK staining is combined with CD30 staining, the diagnosis of ALK+ ALCL can usually be made. In both cases, the "hallmark cells" are key in making the correct diagnosis. In ALK+ ALCL, the ALK staining may be cytoplasmic and/or nuclear based on the translocation associated with the *ALK* gene [28]. For example, the nucleophosmin (*NPM1*) gene on chromosome 2 encodes a nucleolar domain which explains why ALK staining associated with the classic t(2;5) occurs in the nucleus and cytoplasm, while other cases only stain positive in the cytoplasm.

There are five histological subtypes recognized within ALK+ ALCL: common, lymphohistiocytic, small cell,

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Hodgkin-like, and composite [9, 26]. Histological subtype has been shown to have prognostic implications [29–31]. However, it is important to note that concordance in identifying the specific subtype between pathologists is much lower than with other lymphomas.

As implied by the name, the majority of ALCL cases (60–70%) are "common pattern." In the common pattern, the neoplastic cells are large with bizarre horseshoe-shaped nuclei (hallmark cells) [4, 26]. Occasionally, there are also multinucleated tumor cells that resemble Reed-Sternberg cells. The malignant cells also tend to have large amounts of cytoplasm. Because the lymphoma cells commonly involve the sinuses of the lymph node, pathologists occasionally misdiagnose ALCL as a metastatic solid tumor on first glance.

The "lymphohistiocytic" variant represents approximately 10% of cases and is occasionally confused with reactive lymphadenopathy. As implied by its name, the lymphohistiocytic pattern is characterized by malignant cells mixed in a background of numerous reactive histiocytes. Because of the number of histiocytes, these cases were historically classified as a lymphohistiocytic lymphoma [32]. It wasn't until 1998 when Ott et al. described a case with the classic t(2;5) of ALCL which led to the re-classification of this disease to a subtype of ALCL [33].

The "small cell" pattern represents approximately 10% of ALCL cases. In the small cell variant, the large "hallmark cells" are rare with the majority of malignant cells smaller than seen in the other subtypes. These smaller malignant cells are often clustered in a perivascular pattern providing an important location for the pathologist to concentrate on. Similar to the lymphohistiocytic subtype, it wasn't until the description of this disease in combination with the t(2;5) that this variant was accepted as an ALCL [24].

The rarest subtype (<5%) is the "Hodgkin-like pattern" where the malignant cells resemble Reed-Sternberg cells with the surrounding cells mimicking the appearance of classic nodular sclerosing Hodgkin lymphoma. Prior to the use of ALK staining, it is very likely that these tumors were misclassified as Hodgkin lymphoma as separating the two entities on the basis of morphology is extremely difficult [34].

The last variant is the "composite pattern" representing about 30% of cases. The composite pattern exists when the common pattern and one of the other variants are present in the same specimen. These morphologically different appearing patterns within the same specimen exist because they are linked by a common genetic driver in *ALK* translocation.

Anaplastic Lymphoma Kinase

The translocation t(2;5) was identified soon after the description of ALCL as a distinct lymphoma [5–7]. This unique translocation is present in the majority of pediatric ALCL cases [21, 28]. The two genes responsible for the chromosomal translocation are the *NPM1* gene at 5q35 and *ALK* gene at 2p23. Investigation by Morris et al. in 1994 demonstrated *ALK* to be an oncogene that encodes the protein receptor tyrosine kinase ALK [8]. Similar to other receptor tyrosine kinases, ALK possesses an extracellular ligand-binding region, a transmembrane region, and a cytoplasmic kinase region. Normal ALK signaling occurs when extracellular ligand activation activates the intracellular tyrosine kinase resulting in downstream activation of other intracellular pathways. In contrast to regulated ALK expression, the fusion gene involving *ALK* in ALCL results in the expression of a ligand-independent ALK tyrosine kinase [10]. All ALK+ ALCLs result from a chromosomal translocation involving *ALK* and a partner gene. These translocations generate a fusion protein where the partner protein to ALK promotes dimerization creating a constitutively activated kinase. The unregulated ALK tyrosine kinase activity is the primary mechanism for oncogenesis in ALK+ ALCL [11, 35–37]. While the *NPM1-ALK* translocation is the most common translocation (85–95% of cases) associated with unregulated ALK activity, numerous other translocations involving *ALK* have been implicated in the development of ALCL [38, 39]. In order to detect *ALK* translocations, fluorescent in situ hybridization (FISH) is commonly used. The most commonly used assay is a break-apart assay where the 5' and 3' ends of the *ALK* gene are coded in different colors in order to detect all translocations involving *ALK* regardless of partner gene.

Although the downstream signaling pathways caused by ALK activation have not been completely characterized, it appears that *ALK* translocations activate a number of pathways such as the JAK3-STAT3 pathway, the PI3K-AKT pathway, and the Ras-extracellular signal-regulated kinase pathway [36, 40–43]. As many of these pathways are involved in cell cycle progression, cell survival, and cell proliferation, it is not surprising that unregulated ALK kinase activation is oncogenic. That said, the ubiquitous nature of ALK activation in ALK+ ALCL also provides a potential therapeutic target.

It is well known that patients with ALK+ ALCL display immune responses to ALK. This is best exemplified by anti-ALK antibodies found in a number of patients with varying degrees of activity [44, 45]. In addition to antibody responses, patients have also been shown to have a cellular response to ALK with NPM1-ALK inducing expression of PD-L1 via the STAT3 pathway [46]. Lastly, most ALCLs express PD-L1 and lack CD48 expression which may explain why an activated immune system is ineffective in controlling and preventing ALCL [47, 48]. All of these demonstrate that while ALCLs do indeed induce immune responses, they also express pathways which enable the tumor cells to escape the immune system.

Non-anaplastic Lymphoma Kinase

While ALK+ ALCLs share a common oncogenic pathway, multiple studies have shown that ALK- ALCL is a much more heterogeneous disease [49]. Recent studies have dis-

covered a number of recurrent genetic alterations. Rearrangement of DUSP22 involving t(6;7) has been found in 30% of cases in one series and was associated with an excellent prognosis [50]. This is contrasted by cases with TP63 rearrangements marked by inv [3] which are associated with a poor prognosis. Even more interesting is that Crescenzo et al. have shown that recurrent alterations in ALK- ALCL lead to activation of the JAK-STAT3 pathway [51]. Activation of the JAK-STAT3 pathway is a known consequence of ALK activation suggesting that ALK- ALCL and ALK+ ALCL share some overlap in pathogenesis and also provide for potential therapies which could be used in both entities. However, caution must be used as all of these findings are in adults patients with ALK- ALCL as there have been no studies in children with ALK- ALCL.

Clinical Features

Presentation

ALK+ ALCL occurs most frequently in the first three decades of life with a slight male predominance, while ALK- ALCL is relatively rare in children and young adults affecting mainly older patients. The median age at diagnosis for children and adolescents with ALCL is approximately 12 years of age with rare cases below 1 year of age. Since almost all pediatric cases are ALK+, there are not enough ALK- cases to accurately determine a median age range for ALK- disease. Pediatric patients with ALCL frequently present with B symptoms (50–75%) as well as advanced disease with the majority of cases having stage III–IV disease. There is a high incidence of nodal involvement (>90%) occurring throughout the body both above and below the diaphragm. Even though ALCL is a peripheral T-cell lymphoma, patients often have mediastinal involvement. In addition to nodal involvement, extra-nodal involvement of the skin (25%), lung (10%), bone (17%), and/or liver (8%) is common in pediatric ALCL [2, 52, 53]. Bone marrow involvement varies depending on the mode of detection utilized. Basic morphology has a low level of detection (<10%) which increases to 15–30% when the bone marrow is stained for CD30 and ALK. Molecular studies demonstrate even higher bone marrow involvement with ~50% having detectable disease by polymerase chain reaction (PCR) [29, 54, 55]. While rare, ALCL can present in a leukemic phase with leukocytosis and circulating lymphoma cells [56–59]. Central nervous system involvement is rare occurring in less than 5% of cases [60]. No correlation between ALCL and immune deficiencies has been reported.

In addition to frequently presenting with B symptoms, children with ALCL can present with hemophagocytic lymphohistiocytosis (HLH) or HLH-like symptoms. HLH seems to be more commonly associated with ALCL than other forms of lymphoma [61]. A retrospective review by Pasqualini et al. of 50 children with ALCL found that 6% of cases were associated with HLH at diagnosis [62]. While an important report demonstrating the association of HLH with ALCL, it is still very likely an underestimate of the number of cases. The overlap between symptoms of ALCL and HLH, common extra-nodal involvement of ALCL mimicking HLH, and the known predilection for patients with ALCL to demonstrate systemic immune responses suggests a biological correlation between the two entities. Of note, patients with HLH-associated ALCL did not require specific HLH therapy in this study and responded to standard chemotherapy regimen. This response is not entirely surprising as ALCL and HLH both occur in the setting of immune activation and can be targeted by similar agents.

Staging

One of the original staging classifications for lymphomas was developed by Dr. Murphy while at St. Jude's hospital and either goes by the "Murphy Staging system" or the "St. Jude's classification" [63]. This classification system has been used for decades and is the system utilized in the vast majority of studies noted in this chapter. However, this system relies heavily on nodal disease. As discussed earlier, ALCL commonly presents with extra-nodal disease that does not always fit into this classification system. Thankfully in 2015, a multidisciplinary international task force developed an enhanced staging classification called the International Pediatric NHL Staging System (IPNHLSS) [64]. The new system increased concordance between disease burden and stage by allowing for more definitive organ involvement, utilizing newer imaging techniques to identify areas of involvement, and documenting each site of disease in both nodal and extra-nodal areas. While making historical comparisons more difficult, the newer staging system will hopefully lead to improved description of disease burden and the clinical presentation of ALCL.

Treatment

First-Line Treatment

Varying strategies for treating children with ALCL have emerged over the past two decades with similar success rates (Table 20.1). The reason for the variation in treatment strategies is multifactorial. Initially, the lack of clear diagnostic criteria to differentiate ALCL from other lymphomas led to ALCL being treated on other lymphoma studies. As diagnostic criteria became more standard, clinical trials specifically for ALCL utilized different treatment strategies as there was no standard of care. Some trials used the large hallmark cells

Table 20.1 Frontline treatment results for pediatric ALCL

Treatment Strategy	Randomized or Stratified	Number of Patients	Included Stage I and/or II	Median Age in years (Range)	ALK Status POS/N (%)	Treatment Duration (months)	EFS	EFS for Specific Subsets	Reference (year published)
B-cell strategy with COPADM + maintenance		82	Yes	10 (1.4–17)	74/80 (93%)	8	66%		Brugieres et al. [13]
B-cell strategy with BFM-90	Stratified	89	Yes	10.5 (0.8–17.3)	35/43 (81%)	5	76%		Seidemann et al. [3]
B-cell strategy with LMB	Stratified	72	Yes	11.8 (1.1–16.4)	NR	7–8	59%		Williams [22]
T-cell leukemia therapy		34		11.6 (4.2–14.9)	12/13 (92%)	24	65%		Rosolen et al. [20]
APO		86	No	NR	NR (89%)	12	72%		Laver et al. [16]
Intensive T-cell strategy		86	No	NR	64/71 (90%)	12	68%		Lowe et al. [18]
ALCL99 (B-cell strategy)	Randomized	352	Yes	11 (0.3–19.5)	337/352 (96%)	5	73%		Brugieres et al. [14]
ALCL99 (B-cell strategy)	Randomized	217		NR	NR	12 (VBL) 5 (No VBL)		VBL 72% No VBL 70%	Le Deley et al. [17]
B-cell strategy BFM95	Stratified	32	Yes	10 (0.6–17.6)	28/30 (93%)	5	68%		Pillon et al. [19]
APO with randomization of vinblastine	Randomized	125	No	11.9 (0.7–20)	101/121 (90%)	12	76%	APO 74% APV 79%	Alexander et al. [12]

NR not recorded, *POS* positive, *N* number, *EFS* event-free survival, *VBL* vinblastine, *APV* APO substituting VBL for vincristine

to design treatment strategy similar to other large cell lymphomas, while others used the T-cell immunophenotype as guidance for treatment strategy. This unique history led to widely varying strategies with the largest prospective studies discussed below.

The Pediatric Oncology Group (POG) study 9315 examined the impact of adding intermediate-dose methotrexate and high-dose cytarabine to a 12-month-long APO regimen. The study enrolled 86 patients with ALCL (89% of patients were ALK+) with either stage III or IV disease. After a 4-week induction phase (doxorubicin, vincristine, prednisone, and intrathecal methotrexate), patients were treated every 21 days with APO maintenance (doxorubicin and vincristine on day 1, prednisone and 6-mercaptopurine on days 1–5) or APO maintenance alternating with intermediate-dose methotrexate and higher-dose cytarabine every 21 days. Both maintenance arms included intrathecal methotrexate. Methotrexate was substituted for doxorubicin after a cumulative doxorubicin dose of 300 mg/m^2 was reached. Finding no benefit to the additional agents, Laver et al. reported a combined 4-year EFS of 72%. Of note, three patients were CNS-positive and received cranial radiation [16].

The Children's Oncology Group study ANHL0131 tested the substitution of vinblastine for vincristine in maintenance for children with ALCL when given with the APO regimen. The study enrolled 125 patients with stage III or IV disease (90% of patients were ALK+) with 64 in the standard APO arm and 61 in the APV (vinblastine) arm. The study initially used a dose of 6 mg/m^2 for vinblastine but targeted toxicity rules reduced the dose to 5 mg/m^2 then to 4 mg/m^2. The study closed to accrual when a scheduled interim analysis demonstrated the dose of 4 mg/m^2 of vinblastine would likely meet toxicity stopping rules and would likely not find a difference in EFS between the two arms. The 3-year EFS for all patients was 76% with no benefit to the patients receiving the additional vinblastine (p = 0.73). Of note, four patients were CNS-positive and received cranial radiation [12].

The Children's Cancer Group (CCG) 5941 study used chemotherapy based on that used for T-cell lymphoblastic lymphoma intensified and shortened to 11 months in duration. Chemotherapy was broken down into induction, consolidation, followed by maintenance. The trial enrolled 86 patients with ALCL who had non-localized disease (stage was not reported). Sixty-four of 71 (90%) patients tested were ALK+ and one patient had CNS disease who received cranial radiation. The study reported a 5-year EFS of 68% [18].

An Italian study also used a modified acute leukemia regimen with induction, consolidation followed by maintenance but with total treatment duration of 2 years. The study enrolled 34 patients with ALCL (12 of 13 tested were ALK+). Of the 34 patients, 10 were stage II, 17 stage III, and 7 stage

IV. No patient was CNS-positive. Even with 2 years of treatment, the reported EFS of 65% for all patients is similar to other treatments of less duration [20].

The French Society of Pediatric Oncology (SFOP) utilized B-cell NHL therapy over two consecutive studies. Treatment consisted of a COP reduction followed by two courses of COPADM followed by four courses of multi-agent maintenance. The trial treated all clinical stages uniformly and lasted for 5–7 months. Of note, patients with CNS disease were excluded from the study. The trial accrued 82 patients with ALCL (93% were ALK+). Overall, Brugieres et al. reported a 3-year EFS of 66% [13].

A Berlin-Frankfurt-Munster study (BFM-NHL-90) utilized short (5–7 months) intense chemotherapy resembling that used to treat mature B-cell NHL. Following a short prophase, the NHL-BFM 90 protocol stratified patients into three arms: K1 (stage I and II resected) received three 5-day courses of chemotherapy (methotrexate, dexamethasone, oxazaphorins, etoposide, cytarabine, doxorubicin, and intrathecal therapy); K2 (stage II nonresected and stage III) received six 5-day courses; and K3 (stage IV or multifocal bone disease) received six intensified courses including higher doses of methotrexate, cytarabine, and etoposide. A total of 89 patients were enrolled (35 of 43 patients tested were ALK+). Seidemann et al. reported a 5-year EFS of 76% for all patients and 100%, 73%, and 79% for K1, K2, and K3, respectively. Of note, one patient presented with CNS disease and received radiation [3].

The United Kingdom Children's Cancer Study Group (UKCCSG) utilized similar chemotherapy to that used for Burkitt lymphoma at the time (duration of 7–8 months). The protocol stratified patients into three arms: stage I received eight courses of multi-agent chemotherapy; stage II, III, and IV CNS-negative received five courses of chemotherapy with COPADM and CYM; and stage IV with CNS disease received five intensified courses. Overall, the study reported 5-year EFS of 59% for 72 patients. Of note, three patients had CNS disease [22].

The international ALCL99 trial is the largest published clinical trial for pediatric ALCL and has become the standard of treatment in most of the world. The study used chemotherapy based on BFM-NHL-90 (duration of 4–5 months) with two randomizations. The first randomization compared methotrexate 1 g/m^2 administered over 24 hours with intrathecal chemotherapy throughout therapy versus methotrexate 3 g/m^2 administered over 3 hours with a single dose of intrathecal chemotherapy. With 352 patients randomized, the 2-year EFS (73% versus 75%) did not differ between the two arms but the toxicity due to the methotrexate administered over 3 hours was significantly less [14]. The second randomization was for patients with "high-risk features" defined as involvement of skin, mediastinum, liver, lung, and/or spleen. These patients were randomized to no maintenance chemotherapy or weekly vinblastine as a maintenance therapy for a total duration of 1 year. While vinblastine delayed relapses, there was no difference in the 2-year EFS (73% versus 70%) in the 217 randomized high-risk patients [17].

Although included in a number of the clinical trials above, there are two categories which have not been studied specifically: patients with CNS disease and patients with ALK- ALCL.

Patients presenting with CNS disease are unusual accounting for less than 5% of patients at diagnosis [12, 21, 22, 60, 65]. As noted in the study summaries above, most patients with CNS disease were treated with cranial radiation and similar chemotherapy to other patients. Williams et al. published the largest case series of 12 patients with ALCL and CNS disease at presentation [60]. Of the 12, 5 patients presented with lymphoma cells in their cerebral spinal fluid, 5 presented with an intracranial mass, and 2 presented with both. Although small numbers, there was a higher proportion (36%) of lymphohistiocytic subtype than found in CNS-negative ALCL. All 12 patients received chemotherapy with 9 achieving a CR. Of these nine, four received cranial radiation and none recurred, while three of the five patients who did not receive cranial radiation did relapse. Due to the low numbers, it is not clear if cranial radiation is necessary but their results would suggest that it might be needed to prevent CNS relapse. This is in stark contrast to patients who are CNS-negative who may receive relatively little intrathecal chemotherapy and yet do not have a high relapse rate in the CNS. Because of the rarity of CNS disease, some protocols have excluded patients with CNS ALCL leaving a void in attempting to establish a standard of care.

Another subset of ALCL that does not have a proven standard frontline therapy is children with ALK- ALCL. In adult studies, patients with ALK- ALCL have lower EFS with a higher relapse rate compared to patients with ALK+ ALCL [50, 66–68]. The assumption in children and adolescents is that patients with ALK- also have poorer outcomes, but this has not been studied due to the small number of patients. Many of the pediatric studies included patients who were ALK-, but testing for ALK was not available for all patients in many of the earlier studies, and the more recent studies have demonstrated that the majority of patients (>90%) are ALK+ making any specific determination of the EFS for ALK- ALCL in children extremely difficult. At the current time, most ALK- patients are treated similarly to ALK+ patients. However, this is likely to change as there are a number of drugs which specifically inhibit the ALK kinase making them extremely useful in ALK+ but not ALK- ALCL. These drugs have shown incredible promise in relapsed ALK+ ALCL and likely will be used routinely for frontline treatment of ALK+ ALCL in the future.

In summary, while studies have used a wide range of chemotherapy strategies with varying inclusion criteria and

stratifications, no intervention has been able to improve on the failure rate of 25–30% that exists regardless of treatment strategy. Future interventions need to incorporate novel treatment modalities to have the greatest impact on overall survival and toxicity.

Prognostic Factors

Several prognostic factors have been identified in studies performed in children and adolescents with ALCL (Table 20.2):

- The presence of mediastinal involvement, visceral involvement (defined as lung, liver, or spleen involvement), and skin lesions [52].
- Pathological characteristics such as the presence of a small cell or lymphohistiocytic component [30].
- Detection of minimal disseminated disease (MDD) and minimum residual disease (MRD) which have been extensively studied by the German and Italian study groups [29, 31, 55, 69]. MDD can be detected by qualitative PCR for NPM1-ALK in peripheral blood or bone marrow in 50% of the patients at diagnosis and has been shown in two independent studies to be associated with an increased risk of failure [55, 69]. There is good correlation between MDD in peripheral blood and bone marrow. In addition, the quantification of MDD by real-time PCR allows the identification of a very poor prognosis group of patients characterized by the detection of >10 copies of NPM1-ALK in the peripheral blood or bone marrow at diagnosis. In this group of patients, progression-free survival is under 30% [69]. A persistent MRD after the first course of chemotherapy has also been shown to be associated with a high risk of failure. The positivity of MDD at diagnosis as well as persistent minimal residual disease (MRD) is highly correlated with other risk factors such as mediastinal, skin, or visceral involvement, or histologic subtype including a small cell or lymphohistiocytic component [29, 55, 69].
- Serum anti-ALK antibody level at diagnosis: the NPM1-ALK fusion protein induces the production of anti-ALK antibodies in most patients. The level of the serum anti-ALK antibodies at diagnosis has been shown to be inversely correlated with the risk of failure [44, 70]. The level of anti-ALK antibody during treatment also has a prognostic impact since the risk of failure is very low in patients who show significant anti-ALK antibody titers at the end of treatment [71].
- Finally, the combination of anti-ALK antibody titers and MDD at diagnosis allows a stratification of patients into three biological risk groups: a low-risk group with negative MDD and anti-ALK antibody level > 1/750 (31% of patients with a 5-year PFS of 93%), an intermediate-risk group with either low antibody level or a positive MDD (48% of the patients with an PFS of 68%), and a high-risk group (20% of the patients) defined by positive MDD and low anti-ALK antibody titer associated with a 5-year PFS of 28% [31].

Most of these prognostic factors have not been studied in adults with ALCL. Rather, the International Prognostic index (IPI) that has been developed for the prognostication of B-cell lymphoma has also been shown to be associated with prognosis in adult ALCL [72, 73]. The absence of ALK rearrangement is classically associated with a poor prognosis, but results obtained in a retrospective analysis suggested that in patients under 40 year of age, the prognosis for ALK - ALCL was similar to ALK+ ALCL [73].

Treatment of Relapse

Even before the availability of targeted agents, survival rates over 70% following relapse have been reported in several pediatric studies (Table 20.3). Fortunately, most patients are still sensitive to chemotherapy at relapse. One treatment approach for patients with relapsed ALCL is to give second-line chemotherapy followed by high-dose chemotherapy and autologous or allogeneic hematopoietic stem cell transplantation (HSCT) [74–80]. Vinblastine monotherapy has also been shown to be effective leading to a high response rate (>80%) and long-lasting remission in a high proportion of cases [81]. Despite these reports, the optimal treatment of relapsed/refractory ALCL has not yet been defined. As several new targeted agents have been shown to be effective in relapsed disease, the optimal therapy may depend on risk stratification of relapsed disease.

Most relapses occur shortly after the end of chemotherapy with a median interval between relapse and the end of chemotherapy of 1.7 months in patients treated with ALCL99 chemotherapy [17]. Interestingly, relapses seem to occur shortly after the completion of chemotherapy regardless of the length of initial treatment. The main prognostic factor for patients who relapse is time to relapse with a worse outcome for patients with an early relapse especially relapse/progression during frontline treatment [74, 76, 80]. The association of the expression of CD3 on tumor specimen at initial diagnosis with a poor prognosis at relapse has also been suggested [80]. In children, the efficacy of a risk-adapted strategy for ALCL relapse has been evaluated in a trial run by the European Intergroup for Childhood Non-Hodgkin Lymphoma (EICNHL) between 2004 and 2013 for first relapse in patients treated with ALCL99 as frontline treatment. The 3-year EFS and OS of the 118 patients registered between 2004 and 2013 were 58% and 76%, respectively, at first interim analysis [82]. The treatment strategy was as follows:

Table 20.2 Selected prognostic factors for pediatric ALCL

Risk factor	Number of patients	% of patients with risk factor	5-y Progression-free (or event-free) survival	Reference
Clinical features Skin, visceral, or mediastinal involvement	225	64% HR	89% [82–96%] (LR) vs,61% [53–69%] (HR)	Le Deley et al. [52]
Histology LH or SC component	375	30% HR (LH or SC)	79% (74_83%) (LR) vs 51% (42_60%)	Lamant et al. [30]
MDD at diagnosis HR: MDD + in blood and/ or bone marrow	41 180 59	61% MRD+ 57% MRD+ 50% MRD+	100% (LR) vs 41% ± 11 (HR) 83% (± 5%) vs 51% (±5%) 81% (s.e. 6%) vs 70% (s.e.12%)	Mussolin et al. [55], Damm Welk et al. [29]
Quantitative PCR for NPM ALK HR > 10 copies NPM ALK at diagnosis in BM	74 59	22% (> 10 c) 37% (> 10 c)	78% (s.e. 6%) (LR) vs 23% (s.e. 11%) HR 85% (s.e. 6%)(LR) vs 59% (s.e.12%)	Damm Welk et al. [29]
MRD-positive after course A1 LR MDD – IR MRD+/ MDD–; HR MRD + in BM or blood after course A1	180	MRD– 59%% MRD+/MDD– 20% MRD+/MDD + 20%	82% ± 5% (LR) vs 69 ± 9%(IR) vs 19 ± 8% (HR)	Damm Welk et al. [69]
Anti-ALK antibody titer < 1/750 LR AB titer>1/60750 IR AB titer >1/750 and < 1/60750 HR AB titer ≤1/ 750	87 128 34	31%LR,28%IR,39%HR 70% LR + IR vs 30HR 41% low AB titer	89% ± 6% vs 61% ± 8% vs 33% ±9% 79% (s.e.4%) (LR + IR) vs 42% (s.e. 8%) (HR) 98% (s.e. 5%) (LR) vs 58% (s.e. 15%) (HR)	Ait-Tahar et al. [44], Mussolin [31]
MDD and anti-ALK antibody titers Low risk: MRD- AB titer >1/750 Intermediate MRD + or AB titer <1/750 High risk: MRD+ and AB titer >1/750	128	LR 10% IR 48% HR 31%	LR 93% (s.e. 4%) IR 68% (s.e. 6%) HR 28% (s.e., 9%)	Mussolin [31]
Anti-ALK antibody titers at the end of treatment LR > AB titer 1/750 and HR AB titer ≤1/750 LR less than 2 steps decrease of AB titer, IR > 2 steps decrease and AB titer >1/250, HR AB titer ≤1/250	122 122	HR: < 1/750: 22% LR 30%, IR 43% HR 27%	93% ± 5% (LR) vs 66% ±5% (HR) 91% ±5% (LR) vs 53% ± 6% (IR) vs 52% ±9%	Mussolin et al. [71]

HR high risk, *IR* intermediate risk, *LR* low risk, *LH* lymphohistiocytic, *SC* small cell variant, *MDD* minimal disseminated disease, *MRD* minimal residual disease, *BM* bone marrow, *AB* antibody

Table 20.3 Relapsed treatment results for pediatric ALCL

Study group	Period of treatment	Number of patients	Number of patients in CCR after relapse according to therapeutic strategy	3-year EFS	3-year OS	Prognostic factors	Reference
SFOP	1975–1997	41	Chemotherapy alone 11/20 Autologous HSCT 9/15 Allogeneic SCT 0/1	44%	69%	Time to relapse <1 year (EFS: 28%)	Brugieres et al. [74]
BFM	1990–2003		Chemotherapy alone 1/6 Autologous HSTC 21/39 Allogeneic HSCT 11/16		57%	Progression during treatment CD3 positivity	Woessmann et al. [80]
Japan	1989–2003	26	Chemotherapy alone 6/10 Autologous HSTC 3/8 Allogeneic HSCT 6/6	51%	61%	None	Mori et al. [76]
EICNHL	2004–2013		Risk-adapted strategy Vinblastine 21 Autologous HSTC 31 Allogeneic HSCT 45	59%	78%		Ruf et al. [82] (abstract)

- High-risk relapses defined as relapses during frontline treatment or that have CD3-positive disease accounted for 40% of the cases and were treated with allogeneic HSCT after re-induction with multi-agent chemotherapy. This strategy resulted in a 3-year EFS of 64%.
- Intermediate-risk relapses defined as relapses of a CD3-negative ALCL occurring within 12 months of initial diagnosis accounted for 27% of the cases. Patients were treated with autologous HSCT after re-induction with multi-agent chemotherapy. This strategy had disappointing results with a 3-year EFS of 35%.
- Low-risk relapses defined as CD3-negative and >12 months after diagnosis accounted for 18% of the cases and were treated with weekly vinblastine. These patients achieved a 3-year EFS of 85%.

These results have led the EINCHL to recommend allogeneic HSCT for all high-risk relapses and vinblastine monotherapy for low-risk relapses. Of note, there is still no consensus for the treatment of second or further relapses. Several new drugs have shown efficacy in ALCL and may replace all of the above strategies soon.

In adults, auto-HSCT or allo-HSCT following second-line therapy is the standard of care for relapsed/progressive ALCL in patients without age or comorbidity contraindications [72, 83]. However, before the era of novel targeted agents, most patients did not achieve complete remission and were not eligible to HSCT [84]. Analysis of the outcome of adult patients treated with allogeneic or autologous HSCT for ALCL is limited by the fact that most series included only a small proportion of ALK+ALCL among series of peripheral T-cell lymphoma treated with HSCT. Compared results of autologous versus allogeneic transplant are quite different in adults versus children. In the report from the Center for International Blood and Bone Marrow Transplant Research, ALCL patients undergoing autologous HSCT ($n = 61$) had superior 3-year progression-free survival (PFS) than patients treated with allo-HSCT ($n = 51$) (55 vs 35%, $p = 0.03$) [77].

Novel Therapy in ALCL

Brentuximab Vedotin

Brentuximab vedotin, an anti-CD30 monoclonal antibody conjugated with the microtubule-disrupting agent monomethyl auristatin E (MMAE), is now approved both by FDA and EMA for relapsed ALCL in adults. It is administered by intravenous injection once every 3 weeks at a dose of 1.8 mg/kg with a max of 180 mg. In a phase II study performed in adults with relapsed/refractory ALCL (NCT00866047), including 16 ALK+ ALCL and 42 ALK- ALCL, overall response and complete response (CR) rates were 81% and 69%, respectively, in ALK+ ALCL and 88% and 52% in ALK- ALCL [85]. In patients who achieved a complete response, the median duration of response was 13 months with either autologous or allogeneic HSCT after CR, or prolongation of treatment with brentuximab vedotin for 16 injections. In an update of this study with longer follow-up (median 6 years), the median PFS was 25.5 months for the 16 patients with ALK+ALCL and their 5-year overall survival was 56%. The median PFS of 22 patients who achieved CR (including ALK+ and ALK- ALCL) but did not proceed to HSCT was 39.4 months [86]. Brentuximab vedotin's main side effect is peripheral neuropathy as described in 33 of 58 patients in the trial. Among these patients, 67% had a complete resolution of their symptoms at follow-up. A pediatric phase I/II trial (NCT01492088) evaluated pharmacokinetics, safety, and efficacy of brentuximab vedotin in relapsed/refractory ALCL and Hodgkin lymphoma [87]. In patients with ALCL treated at a dose of 1.8 mg/kg every 21 days, the overall response rate was 53% and the median time to progression 6.2 months. Overall, 33% of the patients experienced neuropathy, mostly grade 1 with 83% improving/resolving by the end of treatment.

To date, brentuximab vedotin has been used as a bridge to transplant in relapsed patients. Multiple ongoing trials are testing the drug in combination with chemotherapy or immunotherapy in ALCL at relapse and in frontline treatment.

ALK Inhibitors

There are now small molecule drugs which directly inhibit activated ALK kinase. Most of these medications have been used in the setting of ALK+ non-small cell lung cancer but several have demonstrated efficacy in ALK+ ALCL. Crizotinib, a competitive inhibitor of ALK and MET kinase activity, is an oral medication which was the first ALK inhibitor entered into clinical trials. Crizotinib was shown to induce a high response rate in ALK+ ALCL in the pediatric trial performed by the Children's Oncology Group [88, 89]. Among 26 relapsed/refractory ALK+ ALCL patients included in this phase I/II study, the complete response rates were 83% for the 6 patients treated at a dose of 165 mg/m^2 twice daily and 80% in the 20 patients treated at 280 mg/m^2 twice daily with a median time to first CR/PR of 27 days. In this trial, the median duration of therapy was 2.8 years in patients treated at 165 mg/m^2 and 0.4 years in patients treated at 280 mg/m^2, with 12 patients ceasing protocol therapy to proceed to transplantation. Only two patients suffered from progressive disease during crizotinib treatment after having achieved complete response. In adults, Gambacorti-Passerini reported a complete response in all nine patients treated with compassionate use crizotinib for relapsed/refractory ALCL [90]. Crizotinib seems to be well tolerated as a single agent with neutropenia being the most common drug-related adverse event [89]. Gastrointestinal side effects, transient

visual disturbances, and prolonged QT have also been described [91]. While crizotinib induces responses in majority of patients, the optimal duration of crizotinib treatment has not been assessed. Most progressions described so far occurred within 2 months of treatment initiation [89, 90] or shortly after cessation of crizotinib [92]. Due to the unknown appropriate length of treatment, crizotinib is currently used to induce second remission in relapsed/refractory ALK+ ALCL patients before allogeneic or autologous HSCT or as lifelong therapy. There are several trials nearing activation which will test the efficacy of combination therapy and/or determine the length of treatment needed to induce lasting remission.

Ceritinib, a 2nd-generation ALK inhibitor, has also been tested in ALCL. In the pediatric phase I trial, two of two patients with ALK+ALCL showed a complete response [93]. Similar to crizotinib, no successful discontinuation has been reported, and thus ceritinib is also mostly used to induce remission in relapsed/refractory ALK+ ALCL patients before allogeneic HSCT. In adult patients, ceritinib has demonstrated lasting responses in all three ALK+ ALCL patients included in the phase I trial ASCEND-1 [94]. Most of the adverse events related to ceritinib treatment that lead to dose reduction were gastrointestinal toxicity and hepatic toxicity [91].

Several next-generation ALK inhibitors such as alectinib, brigatinib, and lorlatinib have been developed in non-small cell lung cancer. The efficacy of these agents in ALCL is unknown, but their efficacy in non-small cell lung cancer progressing after first-generation ALK inhibitors or with CNS involvement suggests that may be of help in patients with progressive disease after first- or second-generation ALK inhibitors or in the rare case of an ALCL with CNS involvement at initial presentation or at relapse.

Anti-PD-1 Immunotherapy

Due to the role of immune system in the control of the disease [46, 95], PD-1 inhibitors are very attractive drugs in ALCL. The strong expression of PD-L1 by tumor cells [36] and the success of anti-PD-1 therapies in other immunogenic diseases such as Hodgkin lymphoma suggest that these drugs may be effective in ALCL. Three cases of dramatic and durable responses with PD-1 inhibitors in patients with refractory ALCL have been reported [96–98]. Several phase II trials are ongoing or in preparation for relapsed/refractory peripheral T-cell lymphomas including ALCL or as a specific trial for relapsed/refractory ALCL. For example, a phase II trial using nivolumab in pediatric and adult relapsed/refractory ALK+ ALCL patients aims to evaluate the response rate to nivolumab for patients with progressive disease despite an ALK inhibitor or brentuximab vedotin as well as test the efficacy of nivolumab as consolidation therapy after CR as a replacement to HSCT.

Other Targets

Beside these main groups of targeted therapies, multiple other therapeutic options including inhibitors of PDGFR [99], mTOR and PI3K inhibitors, vaccination against ALK [100], as well as CD30 targeting CAR-T cells [101] have been described.

The availability of such a large number of new therapeutic agents should lead to improvements in the treatment of ALCL aiming to spare low-risk patients from acute and long-term side effects of chemotherapy and to reduce the failure rate in high-risk patients. Given the rarity of this lymphoma and the multiple therapeutic options, only prospective international therapeutic trials including both children and adults with ALCL will allow the evaluation of the role of these different options in frontline as well as at relapse within a reasonable period of time.

Conclusion

Over the last 25 years, ALCL in children has progressed from an unknown entity to a distinct lymphoma. The discovery and subsequent elucidation of the ALK oncogenic pathway has provided a target for novel therapy and increased our understanding of ALCL biology. We are entering an exciting time in the treatment of pediatric ALCL as novel therapies will soon radically change the treatment paradigm for this disease and lead to less toxicity and improved outcomes.

References

1. Prokoph N, Larose H, Lim MS, Burke GAA, Turner SD. Treatment options for paediatric anaplastic large cell lymphoma (ALCL): current standard and beyond. Cancers (Basel). 2018;10(4)
2. Sandlund JT, Downing JR, Crist WM. Non-Hodgkin's lymphoma in childhood. N Engl J Med. 1996;334(19):1238–48.
3. Seidemann K, Tiemann M, Schrappe M, Yakisan E, Simonitsch I, Janka-Schaub G, et al. Short-pulse B-non-Hodgkin lymphoma-type chemotherapy is efficacious treatment for pediatric anaplastic large cell lymphoma: a report of the Berlin-Frankfurt-Munster Group Trial NHL-BFM 90. Blood. 2001;97(12):3699–706.
4. Stein H, Mason DY, Gerdes J, O'Connor N, Wainscoat J, Pallesen G, et al. The expression of the Hodgkin's disease associated antigen Ki-1 in reactive and neoplastic lymphoid tissue: evidence that Reed-Sternberg cells and histiocytic malignancies are derived from activated lymphoid cells. Blood. 1985;66(4):848–58.
5. Kaneko Y, Frizzera G, Edamura S, Maseki N, Sakurai M, Komada Y, et al. A novel translocation, t(2;5)(p23;q35), in childhood phagocytic large T-cell lymphoma mimicking malignant histiocytosis. Blood. 1989;73(3):806–13.
6. Le Beau MM, Bitter MA, Larson RA, Doane LA, Ellis ED, Franklin WA, et al. The t(2;5)(p23;q35): a recurring chromosomal abnormality in Ki-1-positive anaplastic large cell lymphoma. Leukemia. 1989;3(12):866–70.
7. Rimokh R, Magaud JP, Berger F, Samarut J, Coiffier B, Germain D, et al. A translocation involving a specific breakpoint (q35) on

chromosome 5 is characteristic of anaplastic large cell lymphoma ('Ki-1 lymphoma'). Br J Haematol. 1989;71(1):31–6.
8. Morris SW, Kirstein MN, Valentine MB, Dittmer KG, Shapiro DN, Saltman DL, et al. Fusion of a kinase gene, ALK, to a nucleolar protein gene, NPM, in non-Hodgkin's lymphoma. Science. 1994;263(5151):1281–4.
9. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri S, Stein H, et al. WHO classification of tumours of haematopoietic and lymphoid tissues. 4th ed. Lyon: IARC; 2008.
10. Chiarle R, Voena C, Ambrogio C, Piva R, Inghirami G. The anaplastic lymphoma kinase in the pathogenesis of cancer. Nat Rev Cancer. 2008;8(1):11–23.
11. Kinney MC, Higgins RA, Medina EA. Anaplastic large cell lymphoma: twenty-five years of discovery. Arch Pathol Lab Med. 2011;135(1):19–43.
12. Alexander S, Kraveka JM, Weitzman S, Lowe E, Smith L, Lynch JC, et al. Advanced stage anaplastic large cell lymphoma in children and adolescents: results of ANHL0131, a randomized phase III trial of APO versus a modified regimen with vinblastine: a report from the children's oncology group. Pediatr Blood Cancer. 2014;61(12):2236–42.
13. Brugieres L, Deley MC, Pacquement H, Meguerian-Bedoyan Z, Terrier-Lacombe MJ, Robert A, et al. CD30(+) anaplastic large-cell lymphoma in children: analysis of 82 patients enrolled in two consecutive studies of the French Society of Pediatric Oncology. Blood. 1998;92(10):3591–8.
14. Brugieres L, Le Deley MC, Rosolen A, Williams D, Horibe K, Wrobel G, et al. Impact of the methotrexate administration dose on the need for intrathecal treatment in children and adolescents with anaplastic large-cell lymphoma: results of a randomized trial of the EICNHL Group. J Clin Oncol. 2009;27(6):897–903.
15. Han JY, Suh JK, Lee SW, Koh KN, Im HJ, Seo JJ. Clinical characteristics and treatment outcomes of children with anaplastic large cell lymphoma: a single center experience. Blood Res. 2014;49(4):246–52.
16. Laver JH, Kraveka JM, Hutchison RE, Chang M, Kepner J, Schwenn M, et al. Advanced-stage large-cell lymphoma in children and adolescents: results of a randomized trial incorporating intermediate-dose methotrexate and high-dose cytarabine in the maintenance phase of the APO regimen: a Pediatric Oncology Group phase III trial. J Clin Oncol. 2005;23(3):541–7.
17. Le Deley MC, Rosolen A, Williams DM, Horibe K, Wrobel G, Attarbaschi A, et al. Vinblastine in children and adolescents with high-risk anaplastic large-cell lymphoma: results of the randomized ALCL99-vinblastine trial. J Clin Oncol. 2010;28(25):3987–93.
18. Lowe EJ, Sposto R, Perkins SL, Gross TG, Finlay J, Zwick D, et al. Intensive chemotherapy for systemic anaplastic large cell lymphoma in children and adolescents: final results of Children's Cancer Group Study 5941. Pediatr Blood Cancer. 2009;52(3):335–9.
19. Pillon M, Gregucci F, Lombardi A, Santoro N, Piglione M, Sala A, et al. Results of AIEOP LNH-97 protocol for the treatment of anaplastic large cell lymphoma of childhood. Pediatr Blood Cancer. 2012;59(5):828–33.
20. Rosolen A, Pillon M, Garaventa A, Burnelli R, d'Amore ES, Giuliano M, et al. Anaplastic large cell lymphoma treated with a leukemia-like therapy: report of the Italian Association of Pediatric Hematology and Oncology (AIEOP) LNH-92 protocol. Cancer. 2005;104(10):2133–40.
21. Turner SD, Lamant L, Kenner L, Brugieres L. Anaplastic large cell lymphoma in paediatric and young adult patients. Br J Haematol. 2016;173(4):560–72.
22. Williams DM, Hobson R, Imeson J, Gerrard M, McCarthy K, Pinkerton CR. Anaplastic large cell lymphoma in childhood: analysis of 72 patients treated on The United Kingdom Children's Cancer Study Group chemotherapy regimens. Br J Haematol. 2002;117(4):812–20.
23. Mori T, Kiyokawa N, Shimada H, Miyauchi J, Fujimoto J. Anaplastic large cell lymphoma in Japanese children: retrospective analysis of 34 patients diagnosed at the National Research Institute for Child Health and Development. Br J Haematol. 2003;121(1):94–6.
24. Kinney MC, Collins RD, Greer JP, Whitlock JA, Sioutos N, Kadin ME. A small-cell-predominant variant of primary Ki-1 (CD30)+ T-cell lymphoma. Am J Surg Pathol. 1993;17(9):859–68.
25. Bonzheim I, Geissinger E, Roth S, Zettl A, Marx A, Rosenwald A, et al. Anaplastic large cell lymphomas lack the expression of T-cell receptor molecules or molecules of proximal T-cell receptor signaling. Blood. 2004;104(10):3358–60.
26. Benharroch D, Meguerian-Bedoyan Z, Lamant L, Amin C, Brugieres L, Terrier-Lacombe MJ, et al. ALK-positive lymphoma: a single disease with a broad spectrum of morphology. Blood. 1998;91(6):2076–84.
27. Pulford K, Lamant L, Morris SW, Butler LH, Wood KM, Stroud D, et al. Detection of anaplastic lymphoma kinase (ALK) and nucleolar protein nucleophosmin (NPM)-ALK proteins in normal and neoplastic cells with the monoclonal antibody ALK1. Blood. 1997;89(4):1394–404.
28. Perkins SL, Pickering D, Lowe EJ, Zwick D, Abromowitch M, Davenport G, et al. Childhood anaplastic large cell lymphoma has a high incidence of ALK gene rearrangement as determined by immunohistochemical staining and fluorescent in situ hybridisation: a genetic and pathological correlation. Br J Haematol. 2005;131(5):624–7.
29. Damm-Welk C, Busch K, Burkhardt B, Schieferstein J, Viehmann S, Oschlies I, et al. Prognostic significance of circulating tumor cells in bone marrow or peripheral blood as detected by qualitative and quantitative PCR in pediatric NPM-ALK-positive anaplastic large-cell lymphoma. Blood. 2007;110(2):670–7.
30. Lamant L, McCarthy K, d'Amore E, Klapper W, Nakagawa A, Fraga M, et al. Prognostic impact of morphologic and phenotypic features of childhood ALK-positive anaplastic large-cell lymphoma: results of the ALCL99 study. J Clin Oncol. 2011;29(35):4669–76.
31. Mussolin L, Damm-Welk C, Pillon M, Zimmermann M, Franceschetto G, Pulford K, et al. Use of minimal disseminated disease and immunity to NPM-ALK antigen to stratify ALK-positive ALCL patients with different prognosis. Leukemia. 2013;27(2):416–22.
32. Pileri S, Falini B, Delsol G, Stein H, Baglioni P, Poggi S, et al. Lymphohistiocytic T-cell lymphoma (anaplastic large cell lymphoma CD30+/Ki-1 + with a high content of reactive histiocytes). Histopathology. 1990;16(4):383–91.
33. Ott G, Bastian BC, Katzenberger T, Decoteau JF, Kalla J, Rosenwald A, et al. A lymphohistiocytic variant of anaplastic large cell lymphoma with demonstration of the t(2;5)(p23;q35) chromosome translocation. Br J Haematol. 1998;100(1):187–90.
34. Vassallo J, Lamant L, Brugieres L, Gaillard F, Campo E, Brousset P, et al. ALK-positive anaplastic large cell lymphoma mimicking nodular sclerosis Hodgkin's lymphoma: report of 10 cases. Am J Surg Pathol. 2006;30(2):223–9.
35. Hassler MR, Pulverer W, Lakshminarasimhan R, Redl E, Hacker J, Garland GD, et al. Insights into the pathogenesis of anaplastic large-cell lymphoma through genome-wide DNA methylation profiling. Cell Rep. 2016;17(2):596–608.
36. Marzec M, Zhang Q, Goradia A, Raghunath PN, Liu X, Paessler M, et al. Oncogenic kinase NPM/ALK induces through STAT3 expression of immunosuppressive protein CD274 (PD-L1, B7-H1). Proc Natl Acad Sci U S A. 2008;105(52):20852–7.
37. Murga-Zamalloa CA, Mendoza-Reinoso V, Sahasrabuddhe AA, Rolland D, Hwang SR, McDonnell SR, et al. NPM-ALK phos-

phorylates WASp Y102 and contributes to oncogenesis of anaplastic large cell lymphoma. Oncogene. 2017;36(15):2085–94.
38. Damm-Welk C, Klapper W, Oschlies I, Gesk S, Rottgers S, Bradtke J, et al. Distribution of NPM1-ALK and X-ALK fusion transcripts in paediatric anaplastic large cell lymphoma: a molecular-histological correlation. Br J Haematol. 2009;146(3):306–9.
39. Tsuyama N, Sakamoto K, Sakata S, Dobashi A, Takeuchi K. Anaplastic large cell lymphoma: pathology, genetics, and clinical aspects. J Clin Exp Hematop. 2017;57(3):120–42.
40. Bandini C, Pupuleku A, Spaccarotella E, Pellegrino E, Wang R, Vitale N, et al. IRF4 mediates the oncogenic effects of STAT3 in anaplastic large cell lymphomas. Cancers (Basel). 2018;10(1)
41. Turner SD, Yeung D, Hadfield K, Cook SJ, Alexander DR. The NPM-ALK tyrosine kinase mimics TCR signalling pathways, inducing NFAT and AP-1 by RAS-dependent mechanisms. Cell Signal. 2007;19(4):740–7.
42. Webb TR, Slavish J, George RE, Look AT, Xue L, Jiang Q, et al. Anaplastic lymphoma kinase: role in cancer pathogenesis and small-molecule inhibitor development for therapy. Expert Rev Anticancer Ther. 2009;9(3):331–56.
43. Wu C, Molavi O, Zhang H, Gupta N, Alshareef A, Bone KM, et al. STAT1 is phosphorylated and downregulated by the oncogenic tyrosine kinase NPM-ALK in ALK-positive anaplastic large-cell lymphoma. Blood. 2015;126(3):336–45.
44. Ait-Tahar K, Damm-Welk C, Burkhardt B, Zimmermann M, Klapper W, Reiter A, et al. Correlation of the autoantibody response to the ALK oncoantigen in pediatric anaplastic lymphoma kinase-positive anaplastic large cell lymphoma with tumor dissemination and relapse risk. Blood. 2010;115(16):3314–9.
45. Pulford K, Falini B, Banham AH, Codrington D, Roberton H, Hatton C, et al. Immune response to the ALK oncogenic tyrosine kinase in patients with anaplastic large-cell lymphoma. Blood. 2000;96(4):1605–7.
46. Stadler S, Singh VK, Knorr F, Damm-Welk C, Woessmann W. Immune response against ALK in children with ALK-positive anaplastic large cell lymphoma. Cancers (Basel). 2018;10(4)
47. Atsaves V, Tsesmetzis N, Chioureas D, Kis L, Leventaki V, Drakos E, et al. PD-L1 is commonly expressed and transcriptionally regulated by STAT3 and MYC in ALK-negative anaplastic large-cell lymphoma. Leukemia. 2017;31(7):1633–7.
48. Panjwani PK, Charu V, DeLisser M, Molina-Kirsch H, Natkunam Y, Zhao S. Programmed death-1 ligands PD-L1 and PD-L2 show distinctive and restricted patterns of expression in lymphoma subtypes. Hum Pathol. 2018;71:91–9.
49. Parrilla Castellar ER, Jaffe ES, Said JW, Swerdlow SH, Ketterling RP, Knudson RA, et al. ALK-negative anaplastic large cell lymphoma is a genetically heterogeneous disease with widely disparate clinical outcomes. Blood. 2014;124(9):1473–80.
50. King RL, Dao LN, McPhail ED, Jaffe ES, Said J, Swerdlow SH, et al. Morphologic features of ALK-negative anaplastic large cell lymphomas with DUSP22 rearrangements. Am J Surg Pathol. 2016;40(1):36–43.
51. Crescenzo R, Abate F, Lasorsa E, Tabbo F, Gaudiano M, Chiesa N, et al. Convergent mutations and kinase fusions lead to oncogenic STAT3 activation in anaplastic large cell lymphoma. Cancer Cell. 2015;27(4):516–32.
52. Le Deley MC, Reiter A, Williams D, Delsol G, Oschlies I, McCarthy K, et al. Prognostic factors in childhood anaplastic large cell lymphoma: results of a large European intergroup study. Blood. 2008;111(3):1560–6.
53. Lowe EJ, Gross TG. Anaplastic large cell lymphoma in children and adolescents. Pediatr Hematol Oncol. 2013;30(6):509–19.
54. Damm-Welk C, Schieferstein J, Schwalm S, Reiter A, Woessmann W. Flow cytometric detection of circulating tumour cells in nucleophosmin/anaplastic lymphoma kinase-positive anaplastic large cell lymphoma: comparison with quantitative polymerase chain reaction. Br J Haematol. 2007;138(4):459–66.
55. Mussolin L, Pillon M, d'Amore ES, Santoro N, Lombardi A, Fagioli F, et al. Prevalence and clinical implications of bone marrow involvement in pediatric anaplastic large cell lymphoma. Leukemia. 2005;19(9):1643–7.
56. Bayle C, Charpentier A, Duchayne E, Manel AM, Pages MP, Robert A, et al. Leukaemic presentation of small cell variant anaplastic large cell lymphoma: report of four cases. Br J Haematol. 1999;104(4):680–8.
57. Ok CY, Wang SA, Amin HM. Leukemic phase of ALK(+) anaplastic large-cell lymphoma, small-cell variant: clinicopathologic pitfalls of a rare entity. Clin Lymphoma Myeloma Leuk. 2014;14(4):e123–6.
58. Onciu M, Behm FG, Raimondi SC, Moore S, Harwood EL, Pui CH, et al. ALK-positive anaplastic large cell lymphoma with leukemic peripheral blood involvement is a clinicopathologic entity with an unfavorable prognosis. Report of three cases and review of the literature. Am J Clin Pathol. 2003;120(4):617–25.
59. Zecchini Barrese T, Sagramoso C, Bacci F, Sabattini E. Small cell variant of anaplastic large cell lymphoma with leukemic presentation: a diagnostic challenge. Rev Bras Hematol Hemoter. 2017;39(3):269–73.
60. Williams D, Mori T, Reiter A, Woessman W, Rosolen A, Wrobel G, et al. Central nervous system involvement in anaplastic large cell lymphoma in childhood: results from a multicentre European and Japanese study. Pediatr Blood Cancer. 2013;60(10):E118–21.
61. Shah M, Karnik L, Nadal-Melsio E, Reid A, Ahmad R, Bain BJ. ALK-positive anaplastic large cell lymphoma presenting with hemophagocytic lymphohistiocytosis. Am J Hematol. 2015;90(8):746.
62. Pasqualini C, Minard-Colin V, Saada V, Lamant L, Delsol G, Patte C, et al. Clinical analysis and prognostic significance of haemophagocytic lymphohistiocytosis-associated anaplastic large cell lymphoma in children. Br J Haematol. 2014;165(1):117–25.
63. Murphy SB. Classification, staging and end results of treatment of childhood non-Hodgkin's lymphomas: dissimilarities from lymphomas in adults. Semin Oncol. 1980;7(3):332–9.
64. Rosolen A, Perkins SL, Pinkerton CR, Guillerman RP, Sandlund JT, Patte C, et al. Revised international pediatric non-hodgkin lymphoma staging system. J Clin Oncol. 2015;33(18):2112–8.
65. Pillon M, Arico M, Mussolin L, Carraro E, Conter V, Sala A, et al. Long-term results of the AIEOP LNH-97 protocol for childhood lymphoblastic lymphoma. Pediatr Blood Cancer. 2015;62(8):1388–94.
66. Doghri R, HadjKacem LB, Houcine Y, Charfi L, Driss M, Kacem K, et al. Prognostic factors of ALK-negative anaplastic large-cell lymphoma: a single-institution experience. Ann Hematol. 2018;97(4):725–6.
67. Gaulard P, de Leval L. ALK-negative anaplastic large-cell lymphoma. Blood. 2016;127(2):175–7.
68. Savage KJ, Harris NL, Vose JM, Ullrich F, Jaffe ES, Connors JM, et al. ALK- anaplastic large-cell lymphoma is clinically and immunophenotypically different from both ALK+ ALCL and peripheral T-cell lymphoma, not otherwise specified: report from the international peripheral T-cell lymphoma project. Blood. 2008;111(12):5496–504.
69. Damm-Welk C, Mussolin L, Zimmermann M, Pillon M, Klapper W, Oschlies I, et al. Early assessment of minimal residual disease identifies patients at very high relapse risk in NPM-ALK-positive anaplastic large-cell lymphoma. Blood. 2014;123(3):334–7.
70. Mussolin L, Bonvini P, Ait-Tahar K, Pillon M, Tridello G, Buffardi S, et al. Kinetics of humoral response to ALK and its relationship with minimal residual disease in pediatric ALCL. Leukemia. 2009;23(2):400–2.

71. Mussolin L, Pillon M, Zimmermann M, Carraro E, Basso G, Knoerr F, et al. Course of anti-ALK antibody titres during chemotherapy in children with anaplastic large cell lymphoma. Br J Haematol. 2018;182(5):733–5.
72. Hapgood G, Savage KJ. The biology and management of systemic anaplastic large cell lymphoma. Blood. 2015;126(1):17–25.
73. Sibon D, Fournier M, Briere J, Lamant L, Haioun C, Coiffier B, et al. Long-term outcome of adults with systemic anaplastic large-cell lymphoma treated within the Groupe d'Etude des Lymphomes de l'Adulte trials. J Clin Oncol. 2012;30(32):3939–46.
74. Brugieres L, Quartier P, Le Deley MC, Pacquement H, Perel Y, Bergeron C, et al. Relapses of childhood anaplastic large-cell lymphoma: treatment results in a series of 41 children--a report from the French Society of Pediatric Oncology. Ann Oncol. 2000;11(1):53–8.
75. Fukano R, Mori T, Kobayashi R, Mitsui T, Fujita N, Iwasaki F, et al. Haematopoietic stem cell transplantation for relapsed or refractory anaplastic large cell lymphoma: a study of children and adolescents in Japan. Br J Haematol. 2015;168(4):557–63.
76. Mori T, Takimoto T, Katano N, Kikuchi A, Tabuchi K, Kobayashi R, et al. Recurrent childhood anaplastic large cell lymphoma: a retrospective analysis of registered cases in Japan. Br J Haematol. 2006;132(5):594–7.
77. Smith SM, Burns LJ, van Besien K, Lerademacher J, He W, Fenske TS, et al. Hematopoietic cell transplantation for systemic mature T-cell non-Hodgkin lymphoma. J Clin Oncol. 2013;31(25):3100–9.
78. Strullu M, Thomas C, Le Deley MC, Chevance A, Kanold J, Bertrand Y, et al. Hematopoietic stem cell transplantation in relapsed ALK+ anaplastic large cell lymphoma in children and adolescents: a study on behalf of the SFCE and SFGM-TC. Bone Marrow Transplant. 2015;50(6):795–801.
79. Woessmann W, Peters C, Lenhard M, Burkhardt B, Sykora KW, Dilloo D, et al. Allogeneic haematopoietic stem cell transplantation in relapsed or refractory anaplastic large cell lymphoma of children and adolescents--a Berlin-Frankfurt-Munster group report. Br J Haematol. 2006;133(2):176–82.
80. Woessmann W, Zimmermann M, Lenhard M, Burkhardt B, Rossig C, Kremens B, et al. Relapsed or refractory anaplastic large-cell lymphoma in children and adolescents after Berlin-Frankfurt-Muenster (BFM)-type first-line therapy: a BFM-group study. J Clin Oncol. 2011;29(22):3065–71.
81. Brugieres L, Pacquement H, Le Deley MC, Leverger G, Lutz P, Paillard C, et al. Single-drug vinblastine as salvage treatment for refractory or relapsed anaplastic large-cell lymphoma: a report from the French Society of Pediatric Oncology. J Clin Oncol. 2009;27(30):5056–61.
82. Ruf R, Brugieres L, Pillon M, Zimmermann M, Attarbaschi A, Melgrenn K, Williams D, Uyttebroeck A, Wrobel G, Reiter A, Woessman W. Risk-adapted therapy for patients with relapsed or refractory ALCL – final report of the prospective ALCL-relapse trial of the EICNHL. Br J Haematol. 2015;171
83. Chihara D, Fanale MA. Management of anaplastic large cell lymphoma. Hematol Oncol Clin North Am. 2017;31(2):209–22.
84. Morel A, Briere J, Lamant L, Loschi M, Haioun C, Delarue R, et al. Long-term outcomes of adults with first-relapsed/refractory systemic anaplastic large-cell lymphoma in the pre-brentuximab vedotin era: a LYSA/SFGM-TC study. Eur J Cancer. 2017;83:146–53.
85. Pro B, Advani R, Brice P, Bartlett NL, Rosenblatt JD, Illidge T, et al. Brentuximab vedotin (SGN-35) in patients with relapsed or refractory systemic anaplastic large-cell lymphoma: results of a phase II study. J Clin Oncol. 2012;30(18):2190–6.
86. Pro B, Advani R, Brice P, Bartlett NL, Rosenblatt JD, Illidge T, et al. Five-year results of brentuximab vedotin in patients with relapsed or refractory systemic anaplastic large cell lymphoma. Blood. 2017;130(25):2709–17.
87. Locatelli FM-KC, Neville K, Llort A, Beishuizen A, Daw S, Pillon M, Aladjidi N, Klingebiel T, Landman-Parker J, Medina-Sanson A, August K, Huebner D, Sachs J, Hoffman K, Kinley J, Song S, Song G, Zhang S, Gore L. A phase 1/2 study of brentuximab vedotin in pediatric patients with relapsed/refractory (R/R) systemic anaplastic large-cell lymphoma (sALCL) or R/R Hodgkin lymphoma (HL). Hematol Oncol. 2017;35(S2):248–9.
88. Mosse YP, Lim MS, Voss SD, Wilner K, Ruffner K, Laliberte J, et al. Safety and activity of crizotinib for paediatric patients with refractory solid tumours or anaplastic large-cell lymphoma: a Children's Oncology Group phase 1 consortium study. Lancet Oncol. 2013;14(6):472–80.
89. Mosse YP, Voss SD, Lim MS, Rolland D, Minard CG, Fox E, et al. Targeting ALK with crizotinib in pediatric anaplastic large cell lymphoma and inflammatory myofibroblastic tumor: a Children's Oncology Group Study. J Clin Oncol. 2017;35(28):3215–21.
90. Gambacorti-Passerini C, Messa C, Pogliani EM. Crizotinib in anaplastic large-cell lymphoma. N Engl J Med. 2011;364(8):775–6.
91. Costa RB, Costa RLB, Talamantes SM, Kaplan JB, Bhave MA, Rademaker A, et al. Systematic review and meta-analysis of selected toxicities of approved ALK inhibitors in metastatic non-small cell lung cancer. Oncotarget. 2018;9(31):22137–46.
92. Gambacorti-Passerini C, Mussolin L, Brugieres L. Abrupt relapse of ALK-positive lymphoma after discontinuation of crizotinib. N Engl J Med. 2016;374(1):95–6.
93. Geoerger B, Schulte J, Zwaan CM, Casanova M, Fischer M, Moreno L, Trahair T, Jimenez I, Kang HJ, Pappo AS, Schafer E, Weiss BD, Healy ME, Li K, Lin T, Boral A, Pearson AD. Phase I study of ceritinib in pediatric patients (Pts) with malignancies harboring a genetic alteration in ALK (ALK+): safety, pharmacokinetic (PK), and efficacy results. J Clin Oncol. 2015;33(Suppl; abstr 10005)
94. Richly H, Kim TM, Schuler M, Kim DW, Harrison SJ, Shaw AT, et al. Ceritinib in patients with advanced anaplastic lymphoma kinase-rearranged anaplastic large-cell lymphoma. Blood. 2015;126(10):1257–8.
95. Damm-Welk C, Siddiqi F, Fischer M, Hero B, Narayanan V, Camidge DR, et al. Anti-ALK antibodies in patients with ALK-positive malignancies not expressing NPM-ALK. J Cancer. 2016;7(11):1383–7.
96. Chan TS, Khong PL, Kwong YL. Pembrolizumab for relapsed anaplastic large cell lymphoma after allogeneic haematopoietic stem cell transplantation: efficacy and safety. Ann Hematol. 2016;95(11):1913–5.
97. Hebart H, Lang P, Woessmann W. Nivolumab for refractory anaplastic large cell lymphoma: a case report. Ann Intern Med. 2016;165(8):607–8.
98. Rigaud C, Abbou S, Minard-Colin V, Geoerger B, Scoazec JY, Vassal G, et al. Efficacy of nivolumab in a patient with systemic refractory ALK+ anaplastic large cell lymphoma. Pediatr Blood Cancer. 2018;65(4)
99. Laimer D, Dolznig H, Kollmann K, Vesely PW, Schlederer M, Merkel O, et al. PDGFR blockade is a rational and effective therapy for NPM-ALK-driven lymphomas. Nat Med. 2012;18(11):1699–704.
100. Chiarle R, Martinengo C, Mastini C, Ambrogio C, D'Escamard V, Forni G, et al. The anaplastic lymphoma kinase is an effective oncoantigen for lymphoma vaccination. Nat Med. 2008;14(6):676–80.
101. Ramos CA, Ballard B, Zhang H, Dakhova O, Gee AP, Mei Z, et al. Clinical and immunological responses after CD30-specific chimeric antigen receptor-redirected lymphocytes. J Clin Invest. 2017;127(9):3462–71.

Peripheral T-Cell Lymphoma

21

Karin Mellgren and Udo Kontny

Introduction

Non-anaplastic peripheral T-cell lymphomas (PTCLs) are rare in children. A retrospective analysis of the BFM registry on 4083 children with non-Hodgkin lymphoma (NHL) diagnosed between 1986 and 2012 identified 35 cases with non-anaplastic PTCL, comprising 0.9% of NHL in children under 18 years of age [1]. Besides this study, there has been only one other population-based report on the incidence of this rare disease in children [2]. In this report by the CCLG, comprising a lower number of children, non-anaplastic PTCL comprised 1.8% of childhood NHL. The proportion of non-anaplastic PTCL in children therefore appears to be slightly lower than in adults, in whom it constitutes about 4% of NHL in Western countries [3].

Non-anaplastic PTCL is a heterogeneous group of diseases, divided into 28 subtypes by the current WHO classification [4]. In adults, the most common subtypes are peripheral T-cell lymphoma not otherwise specified (PTCL-NOS) occurring in 25% of patients and angioimmunoblastic T-cell lymphoma (AILT) in 18.5% [5, 6]. In children, PTCL-NOS is the most common subtype, followed by NK−/T-cell lymphoma (NKTCL), hepatosplenic T-cell lymphoma (HSTCL), and subcutaneous panniculitis-like T-cell lymphoma (SPLTCL) [1, 2, 7–10]. In contrast to adults, angioimmunoblastic T-cell lymphoma only rarely occurs in children. In the largest retrospective study on non-anaplastic PTCL in children and adolescents comprising 143 patients, PTCL-NOS was diagnosed in 42%, NKTCL in 21%, and HSTCL and SPLTCL in 20% each; only 3% of cases were angioimmunoblastic lymphomas [7].

Comparing the distribution of PTCL subtypes in children does not point out major differences between geographic regions as in adults. Whereas NK-/T-cell lymphoma is more prevalent in adults in Asia than in Western countries, its incidence in children in Asia is extremely rare [11]. This, however, does not exclude that there are differences in the biology of PTCL in children among ethnicities or geographic regions which might lead to different responses to treatment regimens.

The low incidence of PTCL in children and its heterogeneity have been major challenges for making the correct diagnosis of PTCL, especially PTCL-NOS. Most retrospective analyses have encompassed patients over periods of several decades, and diagnosis has been complicated by changing classifications of PTCLs and the development of new diagnostic tools, such as anti-CD30 and ALK1 antibodies (reviewed in [1]) as well as the more knowledgeable use and interpretation of molecular methods, i.e., clonality analyses. In the BFM analysis, out of 69 cases registered as PTCL, 31 were dismissed when the cases were reviewed applying the current clinical and pathological standards of diagnosis [1, 12].

Recent reviews on PTCL in pediatric and adolescent patients show that survival rates are still poor compared to other NHL sub-entities of this age group, and that treatment results vary between PTCL subgroups [1, 2, 7–10, 13]. Histological subtypes of pediatric PTCL differ slightly from what is described in adults, and outcome varies between the subgroups with a good prognosis for patients with SPTCL, intermediate outcome for patients with PTCL-NOS, and a very poor prognosis for patients with HSTCL. In contrast to adults, pre-existing conditions are present in a large number of children with PTCL (25%) and their outcome appears to be worse than in those without pre-existing condition [7].

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As reported in the adult population [14], most pediatric patients have stage III or IV disease and high-stage patients have a worse outcome than patients with low-stage disease. This might be due to a difference in distribution of histological subtypes between low-stage patients and high-stage patients. Indeed, diseases such as SPTCL are more often found in the low-stage group and more aggressive subtypes such as HSTCL in the high-stage group. The outcome of pediatric patients after treatment with conventional chemotherapy is better than what is generally reported for adults, but the pediatric outcome varied among the different subgroups, suggesting a subtype-specific treatment approach being necessary [7].

PTCL-NOS

PTCL-NOS is the most common subtype of non-anaplastic PTCL. Most patients are diagnosed at the end of the first decade of life, with a slight predominance of males [1]. Histologically, tumors contain a mixture of cells of different sizes with atypical pleomorphic nuclei containing prominent nucleoli; the mitotic rate is usually high. T-cell antigens are variably expressed; most cases express CD4, but mature antigens such as CD5 of CD7 are frequently lost (Fig. 21.1).

Biology

PTCL-NOS is a molecularly heterogenous group [15, 16]. Using gene expression profiling (GEP), two major biologically and clinically distinctive subgroups have been defined in adults, one characterized by high expression of GATA-binding protein 3 (GATA3) and its target genes, and another marked by high expression of T-box 21 (TBX21) and eomesodermin (EOMES) and their targets [17]. With respect to the total PTCL-NOS group, the GATA3 subgroup is associated with poor overall survival [17]. Various recurrent mutations have been identified in PTCL-NOS, such as mutations in the

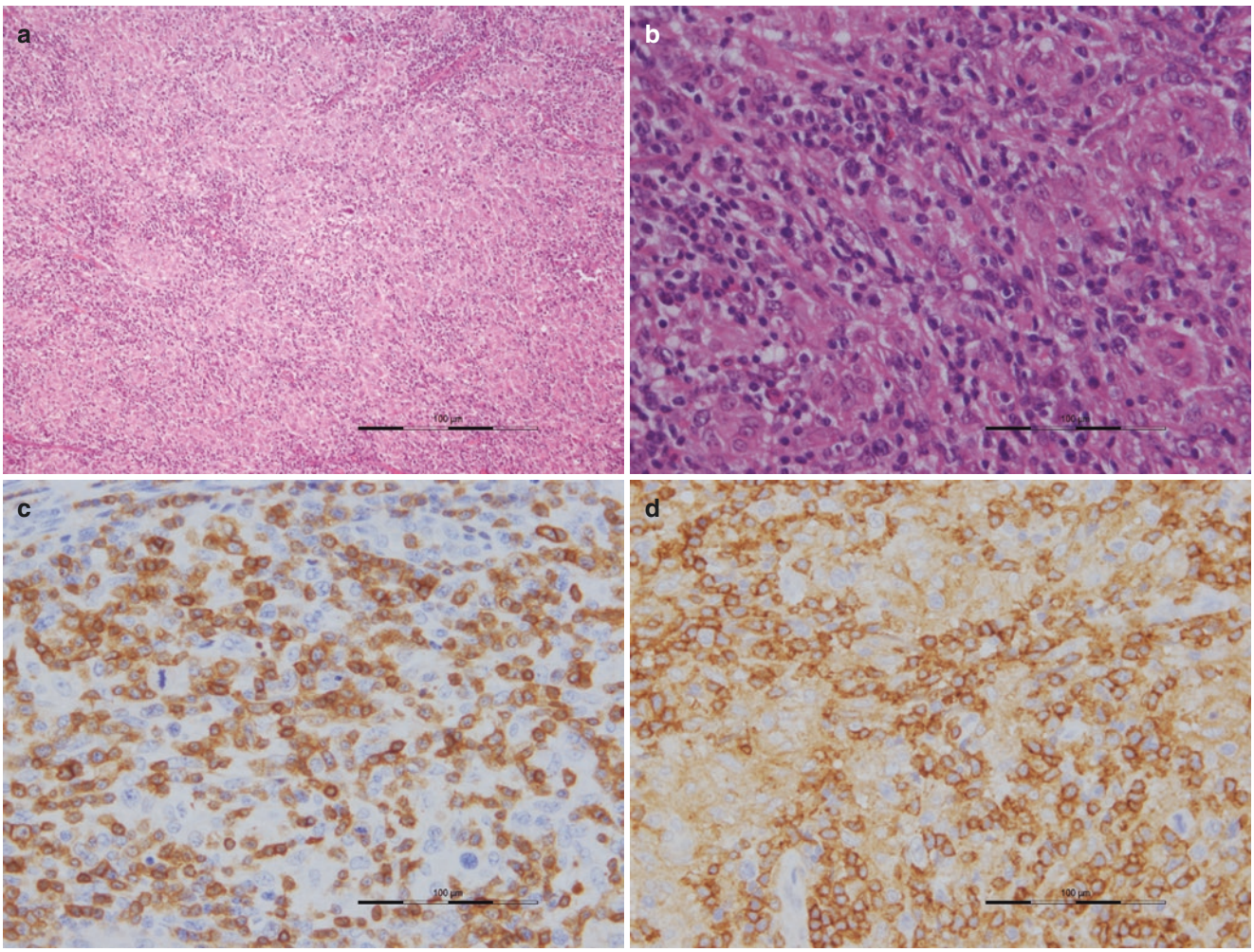

Fig. 21.1 PTCL-NOS. (**a**) Hematoxylin eosin (H&E) staining, 10 × magnification; (**b**) H&E staining, 40 × magnification; (**c**) immunohistochemistry for CD3, 40 × magnification; (**d**) immunohistochemistry for CD4, 40 × magnification

epigenetic modulator genes TET2, DNMT3A, RHOA, or the FYN gene [18–20]. The latter encodes a tyrosine kinase which plays an important role in T-cell activation. As FYN can be targeted by specific kinase inhibitor, it might be of potential therapeutic use [18]. Overexpression of spleen tyrosine kinase (SYK) has been found in a subset of PTCL-NOS characterized by a recurrent t(5;9)(q33;q22) translocation leading to ITK/SYK fusion, and making SYK a potential therapeutic target [21]. Examination of copy number variations in PTCL-NOS has revealed loss at 9p21 as a recurrent event, associated with reduced expression of CDKN2A, CDKN2B, and MTAP. Reduced levels of CDKN2A which are reported in 9% of adults with PTCL-NOS were associated with an inferior outcome [22, 23]. Other structural alterations reported in PTCL-NOS are the fusion CTLA4-CD28 and fusions of VAV1 [23]. Karyotype changes frequently seen in adult patients with PTCL-NOS were observed in 44% of children in whom cytogenetic results were available [24].

Clinical Presentation

Pediatric patients usually present with advanced disease [7]. In the BFM analysis, out of 18 patients, 10 were diagnosed with stage III and 5 with stage IV disease [1]. CNS involvement at diagnosis is rare. Interestingly, most patients with advanced diseases have an effusion, at least at one location, most commonly of pleural origin. B-symptoms occur in about half of patients at diagnosis, and LDH elevation is seen in three-fourths of patients. About half of the patients are anemic at diagnosis, and one-third has a decreased platelet count [1].

Treatment

Because of the rarity of the disease, there has been no standard of treatment for patients with PTCL-NOS. In the 60 patients with PTCL-NOS analyzed by Mellgren et al., 29 received B-cell lymphoma or anaplastic large cell (ALCL)-type therapy, and 22 had T-cell lymphoma-like therapy [7]; no significant differences in pEFS and OS between the two groups were seen. After a median follow-up of 32.5 months (range: 0–229 months), the pOS at 5 years for all patients with PTCL-NOS was 0.56 ± 0.07, and pEFS at 5 years was 0.47 ± 0.07. Relapse occurred in 33% of patients, with a median time to relapse of 8.5 months. Ten patients (17%) were reported to have progressive disease, and three died from treatment-related toxicity. Analysis of the 15 BFM cases with PTCL-NOS, which were included in the analysis by Mellgren et al., revealed a 5-y OS and EFS of 65% ± 11% and 61% ± 11%, respectively [1]. Of the 18 patients, 12 either received an ALCL- or ALCL-like regimen, 5 patients were treated on a protocol for lymphoblastic lymphoma (LBL), and 1 patient received B-NHL therapy. The BFM group has adapted a B-cell type regimen, similar to the regimen for anaplastic large cell lymphoma in 1999 [25]. This regimen consists of two alternating blocks (P1 and P2) for a total of six blocks, preceded by 2 weekly injections of vinblastine (6 mg/m^2/week). The composition of the blocks is as follows: P1: dexamethasone (10 mg/m^2/d, d1–5), vinblastine (6 mg/m^2/d, d1), etoposide (100 mg/m^2/d, d4–5), cytarabine (150 mg/m^2/d in two doses, d4–5), methotrexate (1 g/m^2/d over 24 h, d1), ifosfamide (800 mg/m^2/d, d1–5), and intrathecal triple therapy with methotrexate/cytarabine/prednisone (d2). P2: dexamethasone (10 mg/m^2/d, d1–5), vinblastine (6 mg/m^2/d, d1), doxorubicin (25 mg/m^2/d, d4–5), methotrexate (1 g/m^2/d over 24 h, d1), cyclophosphamide (200 mg/m^2/d, d1–5), and intrathecal triple therapy with methotrexate/cytarabine/prednisone (d2). After the end of sixth block, maintenance chemotherapy with vinblastine (6 mg/m^2/week) was given for a total treatment duration of 72 weeks. In the smaller retrospective report by Windsor et al., 12 pediatric PTCL-NOS patients treated with T-NHL/ALL-type therapy had a better outcome (OS 75%) compared to the 5 patients treated on a B-NHL-type regimen (OS 20%) [2]. Also, six of the eight patients with PTCL-NOS in the report by Kobayashi were treated with T-NHL/ALL-like therapy, with an overall survival of 83% [9].

As there is no clear superiority for either a T- or B-cell lymphoma-based regimen, the usage of an ALCL-like treatment regimen is currently preferred by the authors for induction, as it is tolerated well and it is of a shorter duration than the intensive chemotherapy part in current T-lymphoma/leukemia protocols.

Hematopoietic stem cell transplantation (HSCT) has been performed in 27% of patients with PTCL-NOS in the analysis by Mellgren et al. [7], 14 were allogeneic and 2 were autologous. Six patients (37.5%) were transplanted in CR1, and five of them are alive at the date of last follow-up. Nine patients were transplanted in CR2 (56%) and five are alive. One patient transplanted in partial remission died. Kobayashi reported on 26 pediatric patients with non-anaplastic PTCL, among them 16 patients with PTCL-NOS who received either an allogeneic or autologous HSCT [26]. Of the 3 patients with PTCL-NOS who received an allogeneic transplant in CR1 or PR, all stayed without evidence of disease; of the 12 PTCL-NOS patients with progressive disease or induction failure most of them received an allogeneic transplant. EFS and OS of these patients were 50%; most of the patients receiving an allotransplant were conditioned with a TBI-based regimen; interestingly, all five patients receiving a RIST regimen and all five patients with chronic GvHD survived without evidence of disease, suggesting a graft-versus-lymphoma effect in non-anaplastic PTCL.

Whereas the indication for an allogeneic HSCT in the first relapse seems to be clear, the question remains whether there is a high-risk group of patients who might benefit from an allogeneic stem cell transplant in CR1. Such a risk group could consist of patients who are not in complete remission after induction chemotherapy. MRI/CT, PET-CT as well as MRD in bone marrow or peripheral blood in case of initial involvement are used to monitor response to treatment. PET positivity after induction chemotherapy has been shown in adult patients with PTCL to have a high negative predictive value [27].

Hepatosplenic T-Cell Lymphoma

Hepatosplenic T-cell lymphoma (HSTCL) is an aggressive disease characterized by splenomegaly, often hepatomegaly without lymphadenopathy, B-symptoms, and cytopenias. A leukemic phase may develop as the disease progresses [6]. HSTCL has been shown to be associated with long-term immunosuppression and chronic antigen stimulation in 20% of the cases, in particular after solid organ recipients and patients with Crohn's disease treated with azathioprine and infliximab.

Histologically, the tumor is constituted by small to intermediate-size lymphoma cells that preferentially infiltrate the splenic red pulp cords and sinuses, hepatic sinusoids, and bone marrow sinuses. The lymphoma cells usually present with a CD3-positive, CD56-positive, CD4-negative, CD8-negative, and CD5-negative phenotype with a nonactivated cytotoxic profile (Fig. 21.2) [28]. Most cases are TCR γ/δ positive, but cases with similar clinic-pathological features and an α/β phenotype have been reported [29].

Karyotypic studies frequently show an isochromosome 7q, which may be accompanied by trisomy 8 and loss of a sex chromosome [30].

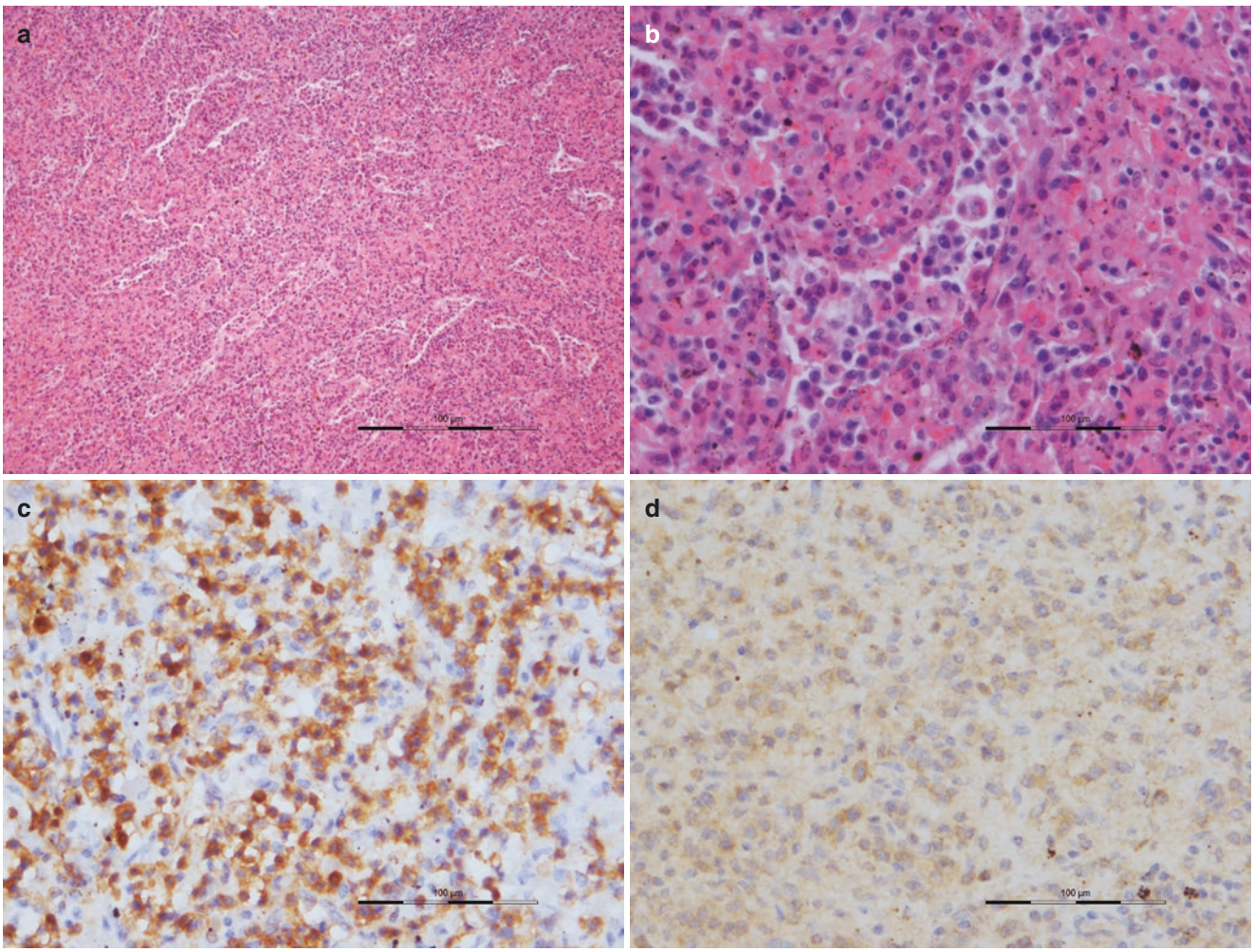

Fig. 21.2 Hepatosplenic T-cell lymphoma. (**a**) H&E staining, 10 × magnification; (**b**) H&E staining, 40 × magnification; (**c**) immunohistochemistry for CD3, 40 × magnification; (**d**) immunohistochemistry for T-cell receptor gamma chain, 40 × magnification

Clinically, patients with HSTCL typically present with B-symptoms (fever, weight loss, and night sweats), massive hepatosplenomegaly, no lymphadenopathy, moderate anemia, and marked thrombocytopenia [1, 7]. The disease is aggressive, and most patients die within 2 years of diagnosis, even if a remission is achieved initially.

The hepatosplenic T-cell lymphomas affect teenagers and young adults in most of the cases. Most of the patients are young men, and the disease is rarer in female patients. Some reports have suggested that αβ-phenotype of HSTCL is more common in female patients [29].

In the adult population, HSTCL accounts for less than 5% of PTCL but appears to be more frequent in the pediatric population. In a retrospective, international, multicenter study, HSTCL was described in 13% of the 143 pediatric cases; all of them had advanced-stage disease [7]. Patients had a dismal prognosis with pOS at 5 years of only 13%.

Subcutaneous Panniculitis-like T-Cell Lymphoma

Cytotoxic lymphomas infiltrating the subcutaneous tissue encompass two distinct entities with clinical and pathological differences, i.e., subcutaneous panniculitis-like T-cell lymphoma (SPTCL)—restricted to cases with an α/β phenotype—and primary cutaneous γδT-cell lymphoma (PCGD-TCL) [4].

Subcutaneous panniculitis-like T-cell lymphoma (SPLTCL) is defined as α/βT-cell-receptor-positive T-cell lymphoma of CD8-positive cytotoxic T cells involving exclusively the subcutaneous tissue (Fig. 21.3). SPLTCL is rare but has been shown to affect all age groups including children [7, 31–33]. The disease has been reported also in very young children. It is associated with autoimmune disease, mainly systemic lupus erythematosus in 20% of the adult cases.

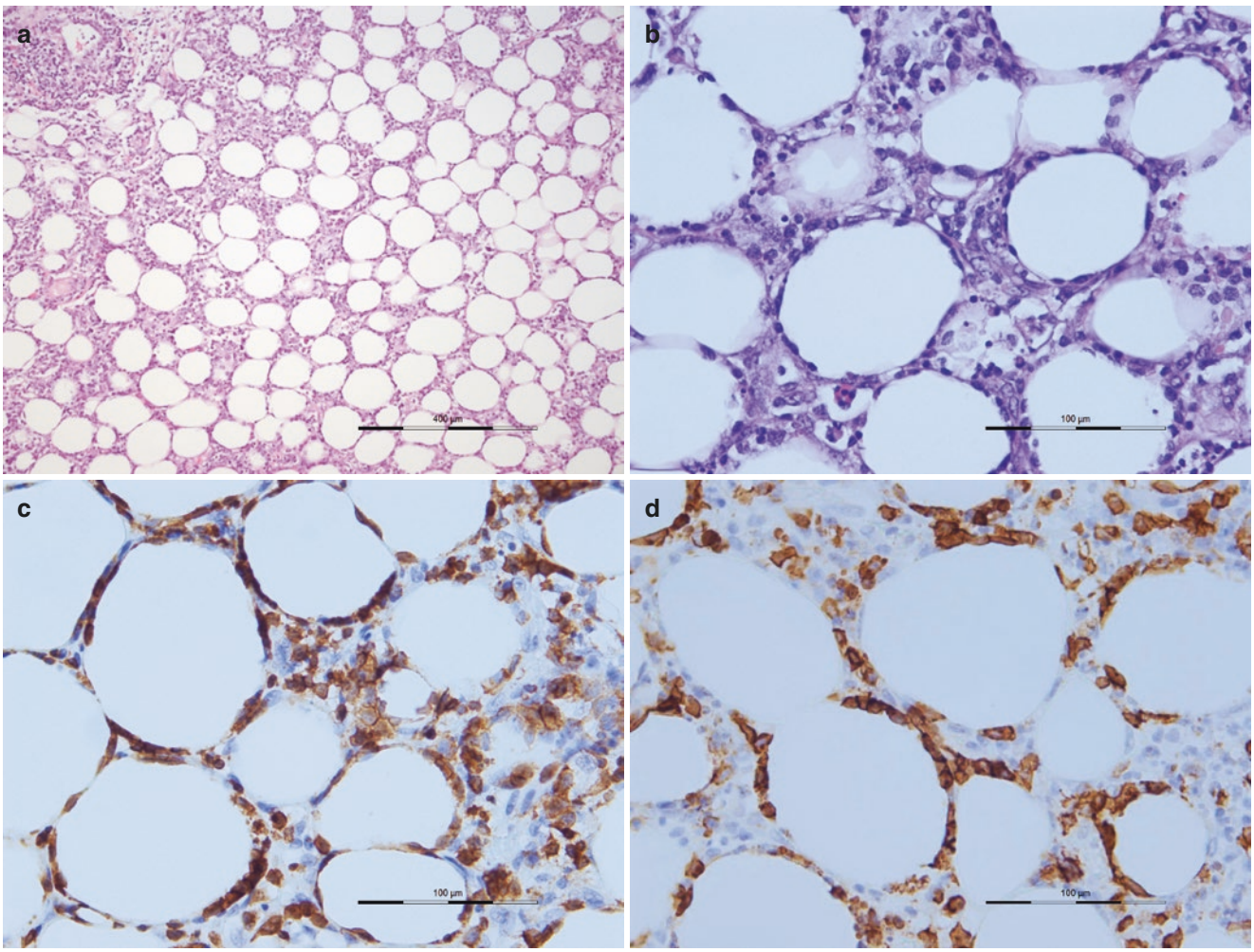

Fig. 21.3 Subcutaneous panniculitis-like T-cell lymphoma. (**a**) H&E staining, 10 × magnification; (**b**) H&E staining, 40 × magnification; (**c**) immunohistochemistry for CD3, 40 × magnification; (**d**) immunohistochemistry for CD8, 40 × magnification

The clinical course of this lymphoma depends on the presence or absence of a haemophagocytic syndrome, which has been reported to develop secondarily in approximately 20% of cases of SPLTCL, usually associated with an unfavorable outcome [32].

Typically, patients with SPTCL show selective infiltration of subcutaneous nodules located on the extremities and chest, infiltrating the subcutis but sparing the dermis. The disease has a good prognosis, in particular if it is not associated with haemophagocytic syndrome, and some series report an OS at 5 years of around 80% [7, 31, 32].

Angioimmunoblastic T-Cell Lymphoma (AITL)

AITL is a disease of the middle-aged and elderly, presenting at a median age of 60–65 years [34]. It is extremely rare in children, with less than 10 cases reported in the literature [7, 35]. The cell of origin of AITL is now thought to be the follicular helper T cell (TFH) [36, 37]. In the retrospective analysis by Mellgren et al., AITL was reported in four children; two were EBV-negative. Median follow-up of the patients with AITL was 51.5 months (range 14–86 months). Three of the patients (75%) underwent HSCT, one allogeneic and two autologous. All three patients were transplanted in CR1, and two of them survived [7]. Based on gene expression analysis of adult cases with AILT, Iqbal and colleagues constructed a molecular prognosticator that appears to be largely related to the microenvironmental signature; here, the high expression of two immunosuppressive signatures was associated with poor outcome [38].

Conclusions

The unique clinical and biological features of pediatric PTCL warrant a treatment approach that should be dependent on the PTCL subtype. Uniform diagnostic criteria for the different subtypes of PTCL in children have to be established, and based on such criteria, patients with these rare disorders will have to be entered in an international registry which could serve as a platform for future clinical trials.

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References

1. Kontny U, Oschlies I, Woessmann W, Burkhardt B, Lisfeld J, Salzburg J, Janda A, Attarbaschi A, Niggli F, Zimmermann M, Reiter A, Klapper W. Non-anaplastic peripheral T-cell lymphomas in children and adolescents – a retrospective analysis of the NHL-BFM study group. Br J Haematol. 2015;168(6):835–44.
2. Windsor R, Stiller C, Webb D. Peripheral T-cell lymphoma in childhood: population-based experience in the United Kingdom over 20 years. Pediatr Blood Cancer. 2008;50(4):784–7.
3. A clinical evaluation of the International Lymphoma Study Group classification of non-Hodgkin's lymphoma. The non-Hodgkin's lymphoma classification project. Blood. 1997;89(11):3909–18.
4. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, Advani R, Ghielmini M, Salles GA, Zelenetz AD, Jaffe ES. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127(20):2375–90.
5. William BM, Armitage JO. International analysis of the frequency and outcome of NK/T-cell lymphomas. Best Pract Res Clin Haematol. 2013;26(1):23–32.
6. Vose J, Armitage J, Weisenburger D, International T-Cell Lymphoma Project. International peripheral T-cell and natural killer/T-cell lymphoma study: pathology findings and clinical outcomes. J Clin Oncol. 2008;26(25):4124–30.
7. Mellgren K, Attarbaschi A, Abla O, Alexander S, Bomken S, Bubanska E, Chiang A, Csóka M, Fedorova A, Kabickova E, Kobayashi R, Krenova Z, Meyer-Wentrup F, Miakova N, Pillon M, Plat G, Uyttebroeck A, Williams D, Wróbel G, Kontny U, on behalf of the European Intergroup for Childhood Non-Hodgkin Lymphoma (EICNHL) and the international Berlin-Frankfurt-Münster (i-BFM) Group. Non-anaplastic peripheral T-cell lymphoma in children and adolescents – an international review of 143 cases. Ann Hematol. 2016;95(8):1295–305.
8. Hutchinson RE, Laver JH, Chang M, Muzzafar T, Desai S, Murphy S, Schwenn M, Shuster J, Link MP. Non-anaplastic peripheral T-cell lymphoma in childhood and adolescence: a Children's Oncology Group Study. Pediatr Blood Cancer. 2008;51:29–33.
9. Kobayashi R, Yamato K, Tanaka F, Takashima Y, Inada H, Kikuchi A, Kumagai MA, Sunami S, Nakagawa A, Fukano R, Fujita N, Mitsui T, Tsurusawa M, Mori T, Lymphoma Committee, Japanese Pediatric Leukemia/Lymphoma Study Group. Retrospective analysis of non-anaplastic peripheral T-cell lymphoma in pediatric patients in Japan. Pediatr Blood Cancer. 2010;54(2):212–5.
10. Al Mahmoud R, Weitzman S, Schechter T, Ngan B, Abdelhaleem M, Alexander S. Peripheral T-cell lymphomas in children and adolescents: a single-institution experience. J Pediatr Hematol Oncol. 2012;34:611–6.
11. Wang ZY, Li YX, Wang WH, Jin J, Wang H, Song YW, et al. Primary radiotherapy showed favorable outcome in treating extranodal nasal-type NK/T-cell lymphoma in children and adolescents. Blood. 2009;114(23):4771–6.
12. Langerak AW, Groenen PJ, Bruggemann M, Beldjord K, Bellan C, Bonello L, et al. EuroClonality/BIOMED-2 guidelines for interpretation and reporting of Ig/TCR clonality testing in suspected lymphoproliferations. Leukemia. 2012;26(10):2159–71.
13. Sandlund JT, Perkins SL. Uncommon non-Hodgkin lymphomas of childhood: pathological diagnosis, clinical features and treatment approaches. Br J Haematol. 2015;169(5):631–46.
14. Ellin F, Landström J, Jerkeman M, Relander T. Real-world data on prognostic factors and treatment in peripheral T-cell lymphomas: a study from the Swedish Lymphoma registry. Blood. 2014;124(10):1570–7.
15. Ballester B, Ramuz O, Gisselbrecht C, et al. Gene expression profiling identifies molecular subgroups among nodal peripheral T-cell lymphomas. Oncogene. 2006;25(10):1560–70.
16. Sakata-Yanagimoto M, Chiba S. Molecular pathogenesis of peripheral T cell lymphoma. Curr Hematol Malig Rep. 2015;10:429–37.
17. Iqbal J, Wright G, Wang C, Rosenwald A, Gascoyne RD, Weisenburger DD, Greiner TC, Smith L, Guo S, Wilcox RA, Teh BT, Lim ST, Tan SY, Rimsza LM, Jaffe ES, Campo E, Martinez A, Delabie J, Braziel RM, Cook JR, Tubbs RR, Ott G, Geissinger E, Gaulard P, Piccaluga PP, Pileri SA, Au WY, Nakamura S, Seto M, Berger F, de Leval L, Connors JM, Armitage J, Vose J, Chan WC,

Staudt LM, Lymphoma Leukemia Molecular Profiling Project and the International Peripheral T-cell Lymphoma Project. Gene expression signatures delineate biological and prognostic subgroups in peripheral T-cell lymphoma. Blood. 2014;123(19):2915–23.
18. Palomero T, Couronne L, Khiabanian H, et al. Recurrent mutations in epigenetic regulators, RHOA and FYN kinase in peripheral T cell lymphomas. Nat Genet. 2014;46(2):166–70.
19. Yoo HY, Sung MK, Lee SH, et al. A recurrent inactivating mutation in RHOA GTPase in angioimmunoblastic T cell lymphoma. Nat Genet. 2014;46(4):371–5.
20. Couronne L, Bastard C, Bernard OA. TET2 and DNMT3A mutations in human T-cell lymphoma. N Engl J Med. 2012;366(1):95–6.
21. Streubel B, Vinatzer U, Willheim M, Raderer M, Chott A. Novel t(5;9)(q33;q22) fuses ITK to SYK in unspecified peripheral T-cell lymphoma. Leukemia. 2006;20(2):313–8.
22. Fujiwara SI, Yamashita Y, Nakamura N, et al. High-resolution analysis of chromosome copy number alterations in angioimmunoblastic T-cell lymphoma and peripheral T-cell lymphoma, unspecified, with single nucleotide polymorphism-typing microarrays. Leukemia. 2008;22(10):1891–8.
23. Sandell RF, Boddicker RL, Feldman AL. Genetic landscape and classification of peripheral T cell lymphomas. Curr Oncol Rep. 2017;19:28.
24. Nelson M, Horsman DE, Weisenburger DD, Gascoyne RD, Dave BJ, Loberiza FR, Ludkovski O, Savage KJ, Armitage JO, Sanger WG. Cytogenetic abnormalities and clinical correlations in peripheral T-cell lymphoma. Br J Haematol. 2008;141(4):461–9.
25. Seidemann K, Tiemann M, Schrappe M, Yakisan E, Simonitsch I, Janka-Schaub G, et al. Short-pulse B-non-Hodgkin lymphoma-type chemotherapy is efficacious treatment for pediatric anaplastic large cell lymphoma: a report of the Berlin-Frankfurt-Munster Group Trial NHL-BFM 90. Blood. 2001;97(12):3699–706.
26. Kobayashi R, Fujita N, Mitsui T, Iwasaki F, Suzumiya J, Kuroda H, et al. Stem cell transplantation for paediatric patients with non-anaplastic peripheral T-cell lymphoma in Japan. Br J Haematol. 2012;159(1):88–93.
27. Casulo C, Schoder H, Feeney J, Lim R, Maragulia J, Zelenetz AD, et al. 18F-fluorodeoxyglucose positron emission tomography in the staging and prognosis of T cell lymphoma. Leuk Lymphoma. 2013;54(10):2163–7.
28. Attygalle AD, Cabecadas J, Gaulard P, Jaffe E, de Jong D, Hyeh Ko Y, Said J, Klapper W. Peripheral T-cell and NK-cell lymphomas and their mimics; taking a step forward – report on the lymphoma workshop of the XVIth meeting of the European Association for Haematopathology and the Society for Hematopathology. Histopathology. 2014;64:171–99.
29. Macon WR, Levy NB, Kurtin PJ, Salhany KE, Elkhalifa MY, Casey TT, Craig FE, Vnencak-Jones CL, Gulley ML, Park JP, Cousar JB. Hepatosplenic alphabeta T-cell lymphomas: a report of 14 cases and comparison with hepatosplenic gammadelta T-cell lymphomas. Am J Surg Pathol. 2001;25(3):285–96.
30. Belhadj K, Reyes F, Farcet JP, Tilly H, Bastard C, Angonin R, et al. Hepatosplenic gammadelta T-cell lymphoma is a rare clinicopathologic entity with poor outcome: report on a series of 21 patients. Blood. 2003;102(13):4261–9.
31. Willemze R. Thirty years of progress in cutaneous lymphoma research. G Ital Dermatol Venereol. 2012;147(6):515–21.
32. Oschlies I, Simonitsch-Klupp I, Maldyk J, Konovalov D, Abramov D, Myakova N, Lisfeld J, Attarbaschi A, Kontny U, Woessmann W, Klapper W. Subcutaneous panniculitis-like T-cell lymphoma in children: a detailed clinicopathological description of 11 multifocal cases with a high frequency of haemophagocytic syndrome. Br J Dermatol. 2015;172(3):793–7.
33. Sakurai E, et al. Subcutaneous Panniculitis-Like T-Cell Lymphoma (SPTCL) with Hemophagocytosis (HPS): Successful Treatment Using High-Dose Chemotherapy (BFM-NHL & ALL-90) and Autologous Peripheral Blood Stem Cell Transplantation. J Clin Exp Hematop. 2013;53(2):135–40.
34. Iannitto E, Ferreri AJ, Minardi V, Tripodo C, Kreipe HH. Angioimmunoblastic T-cell lym- phoma. Crit Rev Oncol Hematol. 2008;68:264–71.
35. Kraus T, Twist CJ, Tan BT. Angioimmunoblastic T cell lymphoma: an unusual presentation of posttransplant lymphoproliferative disorder in a pediatric patient. Acta Haematol. 2014;131:95–101.
36. de Leval L, Rickman DS, Thielen C, et al. The gene expression profile of nodal peripheral T-cell lymphoma demonstrates a molecular link between angioimmunoblastic T-cell lymphoma (AITL) and follicular helper T (TFH) cells. Blood. 2007;109(11):4952–63.
37. Piccaluga PP, Agostinelli C, Califano A, et al. Gene expression analysis of angioimmunoblastic lymphoma indicates derivation from T follicular helper cells and vascular endothelial growth factor deregulation. Cancer Res. 2007;67(22):10703–10.
38. Iqbal J, Weisenburger DD, Greiner TC, Vose JM, McKeithan T, Kucuk C, Geng H, Deffenbacher K, Smith L, Dybkaer K, Nakamura S, Seto M, Delabie J, Berger F, Loong F, Au WY, Ko YH, Sng I, Armitage JO, Chan WC, International Peripheral T-Cell Lymphoma Project. Molecular signatures to improve diagnosis in peripheral T-cell lymphoma and prognostication in angioimmunoblastic T-cell lymphoma. Blood. 2010;115(5):1026–36.

Extranodal NK-/T-Cell Lymphomas and EBV+ Lymphoproliferative Diseases of Childhood

22

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Introduction

Extranodal NK-/T-cell lymphoma, nasal type (ENKTL) is a distinct lymphoma characterized by predominant occurrence in the nasal/paranasal area, skin/soft tissue, or gastrointestinal tract [1]. Epstein-Barr virus (EBV) is one of the most important exogenous factors for lymphomagenesis of ENKTL, which became evident in the past two decades. Another important EBV-associated lymphoid neoplasm in childhood and adolescence is chronic active EBV-associated (CAEBV)-lymphoproliferative disorder (LPD), which has unique clinical characteristics and requires distinctive management [2]. Both of these diseases are rare in childhood and adolescence. There is higher prevalence in Asia and Latin America. The proportion among pediatric T-cell lymphomas (excluding anaplastic large cell lymphoma) is about 5% [3]. Therefore, the following information are mostly based on adult data.

Extranodal NK-/T-Cell Lymphomas

Pathophysiology

ENKTL is transformed from functionally mature NK-cells [1, 4]. The genetic mechanism of lymphomagenesis specific for ENKTL remains unknown. No recurrent genetic or chromosomal abnormalities were found in ENKTL, although the incidence of loss of 6q21–25 or isochromosome 6p is relatively high [4]. However, EBV is capable of transforming lymphocytes to tumor cells [5–7]. Therefore, the EBV is currently regarded as a hallmark of ENKTL, which is detected by means of in situ hybridization (ISH) or Southern blotting.

Another important issue for ENKTL is that lymphoma cells express multidrug resistance (MDR)-associated P-glycoprotein (pGP). The pGP intensely export various cytotoxic agents such as doxorubicin or vincristine [8, 9]. This property causes poor response to conventional lymphoma chemotherapy [10–14].

Incidence

ENKTL accounts for approximately 3% to 11% of all lymphomas in East Asia [15–17]. This subtype predominantly occurs in middle-aged adults (median age of 40s to 50s) with male predominance [4, 10–12, 18–22], but less frequently in children or young adults [3].

Clinical Presentations

The key manifestations of ENKTL include nasal obstruction, discharge, or bleeding. The lymphoma can extend to adjacent tissue, such as nasopharynx, paranasal tissues, orbit, oral cavity, palate, and oropharynx [1]. Some patients present with extensive disease with fever, bone marrow involvement, hemophagocytosis, and disseminated intravascular coagulation (DIC). Other most affected extranodal organs are skin and gastrointestinal tract [23, 24], followed by testes, ovary, pancreas, and adrenal glands. Lymphomatoid gastropathy and NK-cell enteropathy should be excluded before the diagnosis of ENKTL with gastrointestinal (GI) involvement [25, 26]. A half of nasal cases present with localized or locally advanced disease. On the contrary, two thirds of extranasal cases present with advanced stage and rapid progression [11, 12].

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Evaluation and Diagnosis

Histologic evaluation is essential for the diagnosis of ENKTL. Biopsy specimens show varying-size lymphoma infiltration with angiocentric or angiodestructive growth pattern [1, 4]. Necrosis in the inflammatory background is also frequent (Fig. 22.1). Repeated tissue biopsy may be needed to confirm diagnosis in some necrotic cases. Phenotypically, lymphoma cells express CD2, cytoplasmic CD3 (cyCD3), CD7, CD56, CD43, CD45RO, HLA-DR, CD25, FAS, and

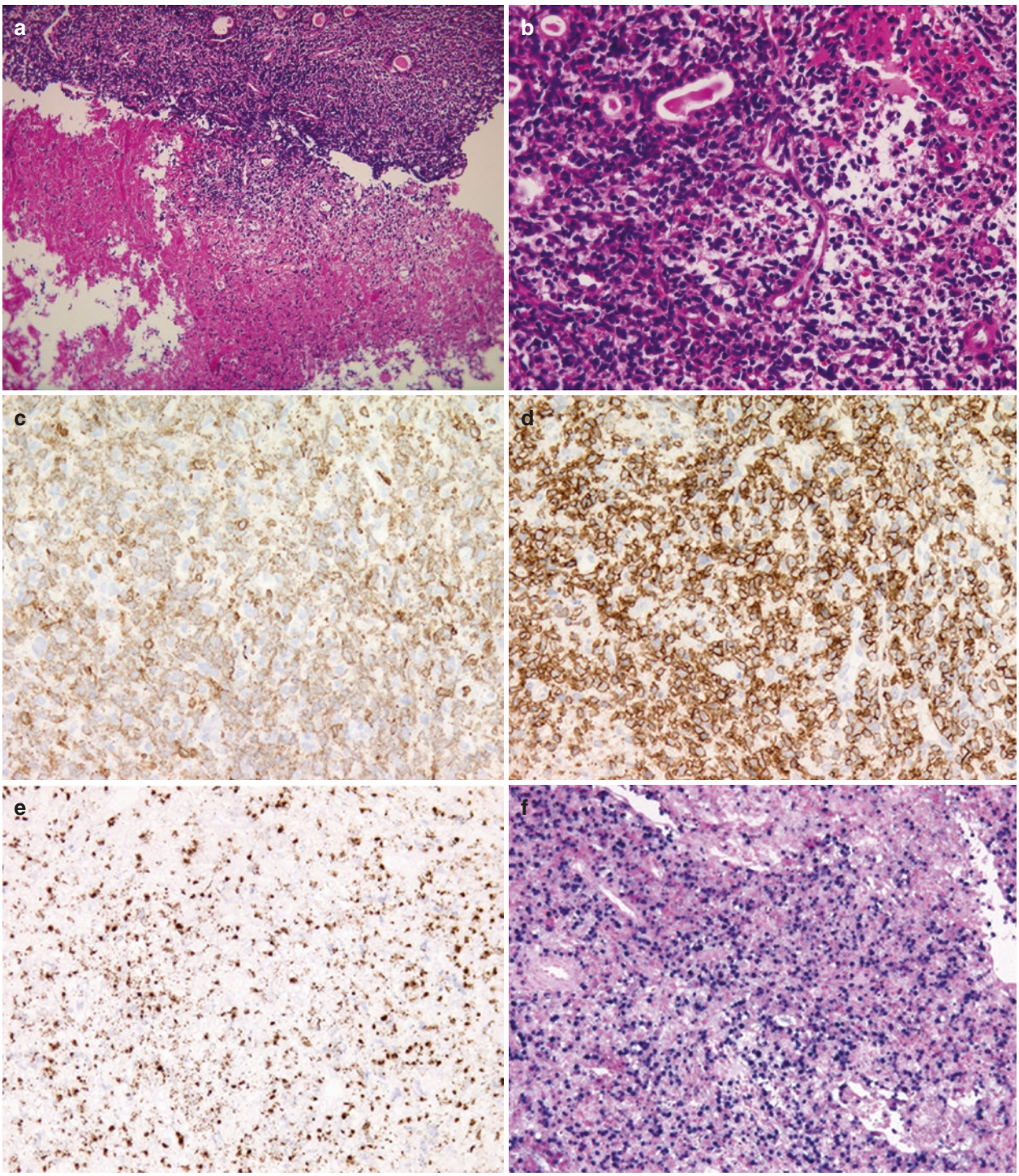

Fig. 22.1 Pathology of extranodal NK-/T-cell lymphoma, nasal type. (**a**) Biopsied specimen of ENKTL (hematoxylin-eosin staining). Diffuse proliferation of atypical lymphoid cells is seen in the upper half of the lesion, while a vast necrosis spreads in the lower half. (**b**) Angiocentric/angiodestructive growth of lymphoma cells can be seen (hematoxylin-eosin staining). (**c**–**f**) The lymphoma cells are positive for CD3 (**c**), CD56 (**d**), granzyme B (**e**), and EBV-encoded RNA (**f**)

FAS ligand. Cytotoxic molecules (TIA-1, granzyme B, and perforin) are almost always positive. Surface CD3 (sCD3), CD4, CD5, CD57, and T-cell receptor (TCR) are consistently negative. CD8 is positive in few of cases, and less frequently CD16. EBV is always positive by type II latency pattern with LMP1 and EBNA-1. In situ hybridization with EBV-encoded small RNA (EBER) is exclusively positive and is the most useful and reliable marker for EBV detection.

Staging

Staging system for ENKTL should be based on the modified Lugano classification [27]. Preferred imaging technique for the staging is whole-body positron emission tomography (PET)/CT, because ENKTL is highly PET-avid [28–30]. Few data are available for the application of the new International Pediatric NHL (IPNHL) classification [31], as well as the previous Murphy or Ann Arber classifications [32, 33].

Prognostic Score and Risk Stratification

The International Prognostic Index (IPI) is still a good practical tool for ENKTL [10–12, 34]. On the other hand, many other ENKTL-specific prognostic factors and scoring systems have been proposed: regional lymph node involvement [10], non-nasal origin [11, 12], local tumor invasiveness [35], and EBV-DNA [36, 37]. Pretreatment EBV-DNA copy number in plasma is a practical tool for predicting treatment response and overall survival (OS) [36, 37]. High expression of latent membrane protein 1 (LMP1) in lymphoma cells was reported as a favorable prognostic factor [38], while high serum-soluble interleukin-2 receptor level represents an unfavorable risk [39]. The NK-/T-cell lymphoma prognostic index (NK-PI), consisting of B symptoms, clinical stage, serum lactate dehydrogenase (LDH) level, and regional lymph node involvement, was developed as the next model for ENKTL patients receiving conventional chemotherapy such as CHOP [10]. Recently, the Prognostic Index for Natural Killer cell lymphoma (PINK) and PINK with EBV-DNA (PINK-E) have been developed from patient data who received non-anthracycline-based treatment (Table 22.1) [40]. The PINK score consists of age, stage, distant nodal involvement, and non-nasal origin. Detectable EBV-DNA at diagnosis is added in the PINK-E score.

Treatment of ENKTL

For limited-stage or localized ENKTL, radiotherapy is a core component of treatment and can be given either before [41] or concomitant with platinum-based (cisplatin or carboplatin) chemotherapy [42, 43]. The latter strategy is termed as concurrent chemoradiotherapy (CCRT). The JCOG0211DI study demonstrated that the OS and progression-free survival (PFS) of RT-2/3DeVIC was not different from RT-full dose (100%) DeVIC [38]. Both RT-2/3DeVIC (Table 22.2) and VIPD (Table 22.3) regimens are recommended for localized ENKTL (Fig. 22.2). CCRT-VIDL showed outstanding outcomes with 2-year OS of 80% and 5-year OS of 70% [3, 42, 43] (Table 22.4). RT-2/3DeVIC has benefits in terms of short treatment duration, tolerable side effects, and promising obtainable long-term information. Sequential chemoradiotherapy by using SMILE [44] or GELOX [45] is the optional first-line treatment of localized ENKTL with 5-year OS of 85% and PFS of 74%. Radiotherapy alone is reasonable for selected patients, but is complicated by distant recur-

Table 22.1 PINK and PINK-E score

Risk factors	PINK	PINK-E
Age >60 years	*	*
Stage III or IV	*	*
Distant LN involvement	*	*
Non-nasal type	*	*
EBV-DNA		*
PINK risk category	No. of above four risk factors	
Low	0	
Intermediate	1	
High	2, 3, 4	
PINK-E risk category	No. of above five risk factors	
Low	0, 1	
Intermediate	2	
High	3, 4, 5	

Abbreviations: *PINK* prognostic index for NK-cell lymphoma, *LN* lymph node, *EBV* Epstein-Barr virus, *No* number
* These factors are components of each prognostic index

Table 22.2 RT-2/3 DeVIC therapy

Agent	Dose	Administration	Days
Radiotherapy	1.8–2.0 Gy	(Total 50 Gy)	1–33 or 38 (5–6 weeks)
Carboplatin	200 mg/m^2	30 min	1, (22, 43)
Etoposide	67 mg/m^2	2 h	1–3, (22–24, 43–45)
Ifosfamide	1000 mg/m^2	3 h	1–3, (22–24, 43–45)
Dexamethasone	40 mg/body	30 min	1–3, (22–24, 43–45)

DeVIC of 2/3 dose is repeated every 3 weeks
Abbreviations: *Min* minutes, *h* hour

Table 22.3 CCRT-VIPD therapy

Agent	Dose	Administration	Days
CCRT part			
Radiotherapy	1.8–2.0 Gy	(Total 40–52.8 Gy)	1–19 to 33 (3–5 weeks)
Cisplatin	30 mg/m^2	15–30 min	1, 8, 15, (22, 29)
VIPD part (3 cycles)			
Etoposide	100 mg/m^2	90 min	1–3
Ifosfamide	1200 mg/m^2	1 h	1–3
Cisplatin	33 mg/m^2	1 h	1–3
Dexamethasone	40 mg/body	Oral or iv	1–4

VIPD is repeated every 3 weeks
Abbreviations: *Min* minutes, *h* hour

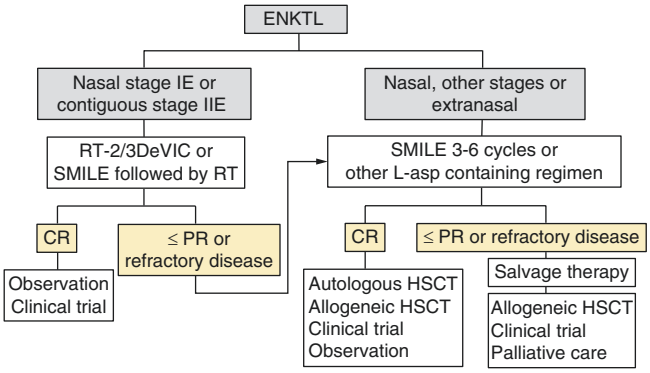

Fig. 22.2 Treatment algorithm of patients with ENKTL. The treatment strategy is divided according to the original site and clinical stage. Patients with nasal stage IE or contiguous stage IIE should be treated with concurrent chemoradiotherapy (RT) such as RT-2/3DeVIC or SMILE followed by RT. If the patients achieve a CR, they should be kept observed. For patients with other stage or extranasal origin, chemotherapy with L-asparaginase containing regimen like SMILE is recommended, as well as limited-stage patients who could not attain CR by the initial treatment. Patients should receive either autologous or allogeneic HSCT after entering CR with SMILE. Abbreviations: CR complete response, PR partial response, HSCT hematopoietic stem cell transplantation

Table 22.4 Comparison of concurrent chemoradiotherapy

	RT-2/3 DeVIC	CCRT + VIPD
Cycle interval	3 weeks	3 weeks
No. of chemotherapy	3 cycles	3 cycles
Treatment period	9 weeks	16–20 weeks
Radiation dose	50 Gy	40–50.8 Gy (median: 40 Gy)
Platinum agent	Carboplatin	Cisplatin
Patient numbers	27	30
Age > 60 years	26%	13.3%
Stage I	67%	50%
NK-PI III or IV	36%	30%
Grade 3/4 toxicities		
Anemia	15%	26.7%
Thrombocytopenia	12%	23.3%
Febrile neutropenia	18%	60%
CR rate	77%	90%
ORR	81%	100%
2y OS (95% CI)	78% (57–89%)	86% (74–99%)
Median F/U (range)	32 months (24–62)	23.7 months (17.3–37)
5y OS (95% CI)	70% (49–84%)	–
Median F/U	67 months (61–94)	–

Abbreviations: *NK-PI* NK-cell lymphoma prognostic index, *CR* complete response, *ORR* overall response rate, *y* year, *OS* overall survival, *CI* confidence interval, *F/U* follow-up

rences. Risk of central nervous system involvement is usually not so high, with less than 10% [46]. Therefore, the CNS prophylaxis for localized disease is controversial and may be considered in patients with high-risk features.

On the other hand, a completely different treatment strategy is required for advanced stage, extranasal origin, relapsed, or refractory ENKTL. From the history of past several decades, CHOP/CHOP-like regimen is insufficient in obtaining the satisfactory outcomes [11]. L-asparaginase-containing regimens produced better outcomes and became the standard of care for these patients. SMILE (steroid, methotrexate, ifosfamide, L-asparaginase, and etoposide, Table 22.5) and AspaMetDex (L-asparaginase, methotrexate, and dexamethasone, Table 22.6) regimens are commonly used. Comparisons of these regimens are summarized in Table 22.7. SMILE regimen demonstrated a 5-year sur-

Table 22.5 SMILE regimen

Agent	Dose	Administration	Days
Methotrexate	2 g/m^2	6 h	1
Ifosfamide	1500 mg/m^2	3 h	2–4
Etoposide	100 mg/m^2	2 h	2–4
Dexamethasone	40 mg/day	30 min	2–4
L-asparaginase	6000 unit/m^2	2 h	8, 10, 12, 14, 16, 18, 20
G-CSF			7-recovery

SMILE is repeated every 4 weeks
Abbreviations: *SMILE* steroid/methotrexate/ifosfamide/L-asparaginase and etoposide, *G-CSF* granulocyte colony-stimulating factor, *h* hour, *min* minutes

Table 22.6 AspaMetDex regimen

Agent	Dose	Administration	Days
Methotrexate (MTX)	3 g/m^2	Drip intravenous	1
L-asparaginase	6000 unit/m^2	Intramuscularly	2, 4, 6, 8
Dexamethasone (DEX)	40 mg/day	Oral	1–4

For patients older than 70 years, MTX is reduced to 2 g/m^2, and DEX to 20 mg/day
AspaMetDex is repeated every 3 weeks

Table 22.7 Comparison of L-asparaginase containing regimens

Regimen	SMILE	AspaMetDex
Cycle interval	4 weeks	3 weeks
Cytotoxic agents	MTX	MTX
	L-asp	L-asp
	Dexa	Dexa
	ETP	
	IFM	
Patient numbers	38	19
Age, median (range)	47 (16–67) years	60 (45–76) years
Stage IE/IIE	29%	63%
Neutropenia grade 4	92%	5.3%
Median duration	3 days	–
Anti-asparaginase antibodies	No data	55%
CR rate	45%	61%
ORR	79%	78%
2y OS	51%	41%
Median f/u	24 mo.	26 mo.
Range	(13–35 mo.)	(17–49 mo.)
5y OS	47%	–
In case of L-asp allergy	If severe enough: omit L-asp	MTX monotherapy

Abbreviations: *NKTSG* NK-cell Tumor Study Group, *MTX* methotrexate, *L-asp* L-asparaginase, *Dexa* dexamethasone, *ETP* etoposide, *IFM* ifosfamide, *CR* complete response, *ORR* overall response rate, *OS* overall survival, *f/u* follow-up

vival of 47% in Asia and 54% in Europe [3]. The grade 4 neutropenia after SMILE is common, and several severe infections or abnormal liver function tests are reported [47–49]. Modification of dose reduction or omission of several agents is needed for the elderly, unfit, or lymphopenic patients [47, 50]. Granulocyte colony-stimulating factor (G-CSF) starting from day 6 of SMILE is mandatory. In several countries, conventional *E. coli* L-asp can be replaced by single dose per cycle of PEG-asparaginase [51]. After obtaining the response, consolidative autologous or allogeneic hematopoietic stem cell transplantation (HSCT) is preferred for these patients [52–54]. Currently, no differential recommendations can be made for the type of HSCT (autologous vs. allogeneic or reduced intensity vs. myeloablative conditioning) [53, 55]. The optimal indications for HSCT in ENKTCL are yet to be well defined, but the prognosis of ENKTL has been dramatically improved by adopting the above treatment strategies in the latest decade [39, 52].

Novel treatment agents include checkpoint inhibitors, daratumumab, lenalidomide, and vorinostat. Two molecular pathways are responsible for the immune regulation, and PD-1/PD-L1 and CTLA4 are the target for antitumor effect [56]. EBV was shown to increase the expression of PD-L1 in Hodgkin and B-cell lymphoma cells [57], which is also applicable for ENKTL [58]. Pembrolizumab and nivolumab were highly effective for relapsed/refractory ENKL who failed L-asparaginase-containing salvage therapies [59–61], suggesting that checkpoint inhibitors are effective for the treatment of ENKTL. CD38 is a transmembrane glycoprotein expressed on plasma cells and NK-cells. Daratumumab is an anti-CD38 antibody, which shows antibody-dependent cell-mediated cytotoxicity [62]. The efficacy of daratumumab was also reported in a single case of relapsed/refractory ENKTL [63].

Response Assessment and Follow-Up

Since ENKTL is highly PET-avid, PET/CT scans should be performed with contrast-enhanced diagnostic CT for response assessment. The Lugano response criteria are commonly used [27]. PET/CT is particularly effective for ENKTL assessment because many patients are complicated

with nasal sinusitis with inflammatory changes on conventional CT. Reduction of EBV-DNA copy number in plasma is well correlated with disease response [34, 36, 37]. In addition to the routine physical examination of lymphoma, these assessments should also be applied for follow-up.

EBV+ LPD of Childhood

Pathophysiology and Characteristics

In healthy individuals normal infectious immunity can be achieved after the initial infection by EBV [64]. However, very few patients cannot eliminate the EBV and may develop recurrent fevers often accompanied by wasting syndrome and failure to thrive [2]. Although the exact cause remains unknown, it is speculated that genetic defects and/or immune deregulation after primary EBV infection are responsible for this condition [6, 7]. This is termed as chronic active EBV infection (CAEBV), but the essential nature is an underlying LPD by EBV-infected lymphocytes. Therefore, this is not a simple infection. EBV+ LPD generally presents with systemic symptoms, including fever and weight loss (Fig. 22.3). In the early phase of CAEBV, the EBV-infected lymphocytosis is polyclonal. However, after several years of waxing and waning course, the clonal selection of lymphocytes progresses. The proliferating EBV-infected lymphocytes become monoclonal with either T-cells or NK-cells [65–67].

Severe mosquito bite allergy (SMBA) is another aspect of CAEBV-LPD [2]. SMBA patients initially show symptoms limited to skin with erythema, bulla, ulcers, necrosis, and scarring (Fig. 22.3). These patients develop systemic symptoms by years, and later can be categorized to CAEBV-LPD. The mosquito's salivary gland secretions activate CD4+ T-cell proliferation and induce LMP1 expression, which cause NK-cell proliferation [2, 67, 68]. In SMBA patients, crusted ulcerative lesions develop after the mosquito bite. After several years, most of them develop EBV-positive lymphocytosis and fall in the category of EBV-LPD. Therefore, SMBA is not a simple allergic condition, but a manifestation of broad spectrum of EBV-LPD. Hydroa vacciniforme (HV) is a self-limited cutaneous disease that occurs in childhood, which manifests with a sun-related eruption showing edema, vesicles, and necrotic areas, as well as scars on the face and dorsa of the hands, forearms, and legs [69]. This form does not impair the general health of the patient and spontaneously remits in adolescence or young adulthood. However, approximately 10% of HV patients also develop EBV-LPD and are termed HV-like LPD [70].

Systemic EBV+ T-cell lymphoma of childhood is a de novo progressive form of LPD (Figure). The official nomenclature has been replaced from LPD to lymphoma in the revised WHO classification 2017 [3, 71]. This is a highly aggressive lymphoma of mostly T-cell, but sometimes NK-cells. Hemophagocytosis is frequent, and therefore, the alternative term of fulminant hemophagocytic lymphohistiocytosis (HLH) exists. However, the systemic EBV+ T-cell lymphoma of childhood is a distinct disease apart from conventional HLH with known genetic abnormalities. A considerable part of patients develop the lymphoma after their initial infection to EBV, but others occur in those with past EBV infection. The difference between these two EBV-related entities remains unknown.

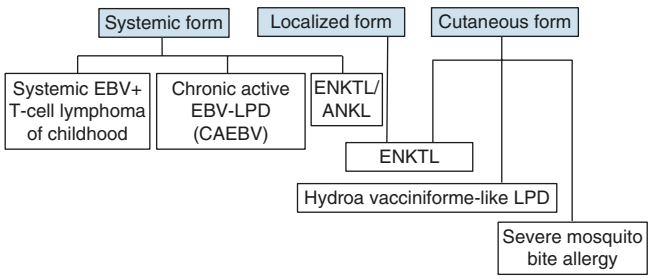

Fig. 22.3 Total scheme of EBV+ T-cell and NK-cell LPD in childhood and AYA. EBV+ T-cell and NK-cell LPD in childhood, adolescence, and young adults can be divided into three categories of systemic, localized, or cutaneous forms. The systemic form includes systemic EBV+ T-cell lymphoma of childhood, chronic active EBV-LPD, advanced stage ENKL, and aggressive NK-cell leukemia. Localized form represents limited-stage ENKL. The cutaneous form consists of cutaneous ENKTL, hydroa vacciniforme-like LPD, and severe mosquito bite allergy. Abbreviations: ENKTL extranodal NK-/T-cell lymphoma, nasal type, ANKL aggressive NK-cell leukemia, LPD lymphoproliferative disease

Incidence

EBV+ LPD of childhood is a rare condition that is most prevalent in East Asia. No sexual predilection is observed. The condition mainly develops in children, adolescence, and young adults [3, 67]. The median age at diagnosis is 14 years, ranging from 1 to 51 years [67]. Patients aged over 40 years are particularly rare. Older patients often lack the chronic active phase and directly develop lymphoma or leukemia. Those patients should be diagnosed with genuine T-cell or NK-cell lymphoma, but should not be categorized to CAEBV-LPD.

Treatment

Allogeneic HSCT is the only curative strategy, but there are several obstacles for the immediate transplant. Patients with fever or high cytokinemia are sometimes troubled by transplant-associated complications. To control this condition, a "cooling" chemotherapy regimen with prednisone, etoposide, and cyclophosphamide has been introduced and yielded good results [72]. After the cooling phase, multi-agent chemotherapy reduces the amount of EBV-infected cells. Finally,

consolidative allogeneic HSCT is recommended, which also contributes to the immunologic reconstruction of CAEBV-LPD patients [72]. Antiviral treatments are consistently ineffective. Chemotherapy and immunomodulating therapy are effective, but the effect is temporary. The conventional therapy alone cannot cure the systemic CAEBV-LPD [67]. Recently, reduced-intensity conditioning (RIC) regimens produced better outcomes than myeloablative conditioning ones [72]. Therefore, the RIC allogeneic HSCT is preferred for children and adolescence, considering the patients' growth.

Prognosis

The prognosis of EBV-LPD varies by subtypes. Systemic EBV+ T-cell lymphoma has a poor prognosis, often accompanied by a fulminant clinical course with days to weeks after diagnosis [71]. Only patients who receive allogeneic transplant can survive long-term. Regarding the systemic CAEBV-LPD, the course and prognosis is relatively indolent. T-cell type, particularly CD4+ type shows more aggressive course than NK-cell type [67]. However, the survival curve gradually declines by years in any subtypes unless the patients receive allogeneic HSCT. Both HV-like LPD and SMBA have a prolonged clinical course, and patients need to be monitored for the risk of malignant transformation. The major causes of death were multiple organ failure and hepatic failure [2]. Age of onset (≥8 years) and liver dysfunction were independent prognostic factors for mortality [67].

Future Consideration for Cell Therapy

Upcoming treatment strategies are the use of EBV-specific cytotoxic T-cells for EBV+ lymphoma. EBV-infected B-cells are controlled by EBV-specific T-cells. The imbalance between malignant EBV-infected B-cells and T-cell immunity causes EBV+ LPD or lymphoma. Transferring EBV-specific cytotoxic T-cells restores tumor destroying property of T-cell immunity. Phase II studies demonstrated its efficacy to prevent and treat EBV+ disorders especially EBV+ PTLPD, but the persistence of T-cell function and long-term outcomes should be investigated [73–78]. Novel targeted agents will be validated in the near future especially in childhood and adolescence.

References

1. Chan JKC, Quintanilla-Martinez L, Ferry JA. Extranodal NK/T-cell lymphoma, nasal type. In: Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, editors. WHO classification of tumours of haematopoietic and lymphoid tissues, Revised 4th edition. Lyon: International Agency for Research on Cancer; 2017. p. 368–71.
2. Quintanilla-Martinez L, Ko Y-H, Kimura H, Jaffe ES. EBV-positive T-cell and NK-cell lymphoproliferative diseases of childhood. In: Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, editors. WHO classification of tumours of haematopoietic and lymphoid tissues, Revised 4th edition. Lyon: International Agency for Research on Cancer; 2017. p. 355–63.
3. Pillai V, Tallarico M, Bishop MR, Lim MS. Mature T- and NK-cell non-Hodgkin lymphoma in children and young adolescents. Br J Haematol. 2016;173:573–81.
4. Suzuki R, Takeuchi K, Ohshima K, Nakamura S. Extranodal NK/T-cell lymphoma: diagnosis and treatment cues. Hematol Oncol. 2008;26:66–72.
5. Suzuki R. Pathogenesis and treatment of extranodal natural killer/T-cell lymphoma. Semin Hematol. 2014;51:42–51.
6. Kawa K. Epstein-Barr virus-associated diseases in humans. Int J Hematol. 2000;71:108–17.
7. Kawa K. Diagnosis and treatment of Epstein-Barr virus-associated natural killer cell lymphoproliferative disease. Int J Hematol. 2003;78:24–31.
8. Egashira M, Kawamata N, Sugimoto K, Kaneko T, Oshimi K. P-glycoprotein expression on normal and abnormally expanded natural killer cells and inhibition of P-glycoprotein function by cyclosporin A and its analogue, PSC833. Blood. 1999;93:599–606.
9. Yamaguchi M, Kita K, Miwa H, Nishii K, Oka K, Ohno T, Shirakawa S, Fukumoto M. Frequent expression of P-glycoprotein/MDR1 by nasal T-cell lymphoma cells. Cancer. 1995;76:2351–6.
10. Lee J, Suh C, Park YH, et al. Extranodal natural killer T-cell lymphoma, nasal-type: a prognostic model from a retrospective multicenter study. J Clin Oncol. 2006;24:612–8.
11. Suzuki R, Suzumiya J, Yamaguchi M, et al. Prognostic factors for mature natural killer (NK) cell neoplasms: aggressive NK cell leukemia and extranodal NK cell lymphoma, nasal type. Ann Oncol. 2010;21:1032–40.
12. Au W, Weisenburger DD, Intragumtornchai T, Nakamura S, Kim W-S, Sng I, Vose J, Armitage JO, Liang R, International Peripheral T-Cell Lymphoma Project. Clinical differences between nasal and extranasal natural killer/T-cell lymphoma: a study of 136 cases from the International Peripheral T-Cell Lymphoma Project. Blood. 2009;113:3931–7.
13. Oshimi K. Progress in understanding and managing natural killer-cell malignancies. Br J Haematol. 2007;139:532–44.
14. Liang R. Advances in the management and monitoring of extranodal NK/T-cell lymphoma, nasal type. Br J Haematol. 2009;147:13–21.
15. Sukpanichnant S, Sonakul D, Piankijagum A, Wanachiwanawin W, Veerakul G, Mahasandana C, Tanphaichitr VS, Suvatte V. Malignant lymphoma in Thailand: changes in the frequency of malignant lymphoma determined from a histopathologic and immunophenotypic analysis of 425 cases at Siriraj Hospital. Cancer. 1998;83:1197–204.
16. Ko YH, Kim CW, Park CS, Jang HK, Lee SS, Kim SH, Ree HJ, Lee JD, Kim SW, Huh JR. REAL classification of malignant lymphomas in the Republic of Korea: incidence of recently recognized entities and changes in clinicopathologic features. Hematolymphoreticular Study Group of the Korean Society of Pathologists. Revised European-American lymphoma. Cancer. 1998;83:806–12.
17. Sun J, Yang Q, Lu Z, et al. Distribution of lymphoid neoplasms in China: analysis of 4,638 cases according to the World Health Organization classification. Am J Clin Pathol. 2012;138:429–34.
18. Avilés A, Díaz NR, Neri N, Cleto S, Talavera A. Angiocentric nasal T/natural killer cell lymphoma: a single centre study of prognostic factors in 108 patients. Clin Lab Haematol. 2000;22:215–20.
19. Barrionuevo C, Zaharia M, Martinez MT, et al. Extranodal NK/T-cell lymphoma, nasal type: study of clinicopathologic and prognosis factors in a series of 78 cases from Peru. Appl Immunohistochem Mol Morphol. 2007;15:38–44.

20. Gualco G, Domeny-Duarte P, Chioato L, Barber G, Natkunam Y, Bacchi CE. Clinicopathologic and molecular features of 122 Brazilian cases of nodal and extranodal NK/T-cell lymphoma, nasal type, with EBV subtyping analysis. Am J Surg Pathol. 2011;35:1195–203.
21. Ng SB, Lai KW, Murugaya S, Lee KM, Loong SLE, Fook-Chong S, Tao M, Sng I. Nasal-type extranodal natural killer/T-cell lymphomas: a clinicopathologic and genotypic study of 42 cases in Singapore. Mod Pathol. 2004;17:1097–107.
22. Pongpruttipan T, Sukpanichnant S, Assanasen T, et al. Extranodal NK/T-cell lymphoma, nasal type, includes cases of natural killer cell and αβ, γδ, and αβ/γδ T-cell origin: a comprehensive clinicopathologic and phenotypic study. Am J Surg Pathol. 2012;36:481–99.
23. Ahn HK, Suh C, Chuang SS, et al. Extranodal natural killer/T cell lymphoma from skin or soft tissue: suggestion of treatment from multinational retrospective analysis. Ann Oncol. 2012;23(10):2703–7.
24. Kim SJ, Jung HA, Chuang SS, et al. Extranodal natural killer/T-cell lymphoma involving the gastrointestinal tract: analysis of clinical features and outcomes from the Asia Lymphoma Study Group. J Hematol Oncol. 2013;6:86.
25. Takeuchi K, Yokoyama M, Ishizawa S, et al. Lymphomatoid gastropathy: a distinct clinicopathologic entity of self-limited pseudomalignant NK-cell proliferation. Blood. 2010;116:5631–7.
26. Mansoor A, Pittaluga S, Beck PL, Wilson WH, Ferry JA, Jaffe ES. NK-cell enteropathy: a benign NK-cell lymphoproliferative disease mimicking intestinal lymphoma: clinicopathologic features and follow-up in a unique case series. Blood. 2011;117:1447–52.
27. Cheson BD, Fisher RI, Barrington SF, et al. Recommendations for initial evaluation, staging, and response assessment of Hodgkin and non-Hodgkin lymphoma: the Lugano classification. J Clin Oncol. 2014;32:3059–68.
28. Weiler-Sagie M, Bushelev O, Epelbaum R, et al. ^{18}F-FDG avidity in lymphoma readdressed: a study of 766 patients. J Nucl Med. 2010;51:25–30.
29. Chan WK, Au WY, Wong CY, et al. Metabolic activity measured by F-18 FDG PET in natural killer-cell lymphoma compared to aggressive B- and T-cell lymphomas. Clin Nucl Med. 2010;35(8):571–5.
30. Fujiwara H, Maeda Y, Nawa Y, et al. The utility of positron emission tomography/computed tomography in the staging of extranodal natural killer/T-cell lymphoma. Eur J Haematol. 2011;87:123–9.
31. Rosolen A, Perkins SL, Pinkerton CR, Guillerman RP, Sandlund JT, Patte C, Reiter A, Cairo MS. Revised international pediatric non-Hodgkin lymphoma staging system. J Clin Oncol. 2015;33:2112–8.
32. Murphy SB. Classification, staging and end results of treatment of childhood non-Hodgkin's lymphomas: dissimilarities from lymphomas in adults. Semin Oncol. 1980;7:332–9.
33. Lister TA, Crowther D, Sutcliffe SB, et al. Report of a committee convened to discuss the evaluation and staging of patients with Hodgkin's disease: Cotswolds meeting. J Clin Oncol. 1989;7:1630–6.
34. Chim C-S, Ma S-Y, Au W-Y, Choy C, Lie AKW, Liang R, Yau C-C, Kwong Y-L. Primary nasal natural killer cell lymphoma: long-term treatment outcome and relationship with the International Prognostic Index. Blood. 2004;103:216–21.
35. Kim TM, Park YH, Lee S-Y, et al. Local tumor invasiveness is more predictive of survival than International Prognostic Index in stage I_E/II_E extranodal NK/T-cell lymphoma, nasal type. Blood. 2005;106:3785–90.
36. Au W-Y, Pang A, Choy C, Chim C-S, Kwong Y-L. Quantification of circulating Epstein-Barr virus (EBV) DNA in the diagnosis and monitoring of natural killer cell and EBV-positive lymphomas in immunocompetent patients. Blood. 2004;104:243–9.
37. Suzuki R, Yamaguchi M, Izutsu K, et al. Prospective measurement of Epstein-Barr virus-DNA in plasma and peripheral blood mononuclear cells of extranodal NK/T-cell lymphoma, nasal type. Blood. 2011;118:6018–22.
38. Yamaguchi M, Takata K, Yoshino T, et al. Prognostic biomarkers in patients with localized natural killer/T-cell lymphoma treated with concurrent chemoradiotherapy. Cancer Sci. 2014;105:1435–41.
39. Yamaguchi M, Suzuki R, Oguchi M, et al. Treatments and outcomes of patients with extranodal natural killer/T-cell lymphoma diagnosed between 2000 and 2013: a cooperative study in Japan. J Clin Oncol. 2017;35:32–9.
40. Kim SJ, Yoon DH, Jaccard A, et al. A prognostic index for natural killer cell lymphoma after non-anthracycline-based treatment: a multicentre, retrospective analysis. Lancet Oncol. 2016;17:389–400.
41. Li Y-X, Yao B, Jin J, et al. Radiotherapy as primary treatment for stage IE and IIE nasal natural killer/T-cell lymphoma. J Clin Oncol. 2006;24:181–9.
42. Yamaguchi M, Tobinai K, Oguchi M, et al. Phase I/II study of concurrent chemoradiotherapy for localized nasal natural killer/T-cell lymphoma: Japan Clinical Oncology Group Study JCOG0211. J Clin Oncol. 2009;27:5594–600.
43. Kim SJ, Kim K, Kim BS, et al. Phase II trial of concurrent radiation and weekly cisplatin followed by VIPD chemotherapy in newly diagnosed, stage IE to IIE, nasal, extranodal NK/T-Cell Lymphoma: Consortium for Improving Survival of Lymphoma study. J Clin Oncol. 2009;27:6027–32.
44. Kwong Y-L, Kim WS, Lim ST, Kim SJ, Tang T, Tse E, Leung AYH, Chim C-S. SMILE for natural killer/T-cell lymphoma: analysis of safety and efficacy from the Asia Lymphoma Study Group. Blood. 2012;120:2973–80.
45. Wang L, Wang Z-H, Chen X-Q, Wang K-F, Huang H-Q, Xia Z-J. First-line combination of GELOX followed by radiation therapy for patients with stage IE/IIE ENKTL: an updated analysis with long-term follow-up. Oncol Lett. 2015;10:1036–40.
46. Ferreri AJM. Risk of CNS dissemination in extranodal lymphomas. Lancet Oncol. 2014;15:e159–69.
47. Yamaguchi M, Kwong Y-L, Kim WS, et al. Phase II study of SMILE chemotherapy for newly diagnosed stage IV, relapsed, or refractory extranodal natural killer (NK)/T-cell lymphoma, nasal type: the NK-Cell Tumor Study Group study. J Clin Oncol. 2011;29:4410–6.
48. Chan A, Tang T, Ng T, Shih V, Tay K, Tao M, Quek R, Lim ST. To SMILE or not: supportive care matters. J Clin Oncol. 2012;30:1015–6; author reply 1016-1017
49. Kim SJ, Park S, Kang ES, et al. Induction treatment with SMILE and consolidation with autologous stem cell transplantation for newly diagnosed stage IV extranodal natural killer/T-cell lymphoma patients. Ann Hematol. 2015;94:71–8.
50. Suzuki R. Treatment of advanced extranodal NK/T-cell lymphoma, nasal-type and aggressive NK-cell leukemia. Int J Hematol. 2010;92:697–701.
51. Qi S, Yahalom J, Hsu M, Chelius M, Lunning M, Moskowitz A, Horwitz S. Encouraging experience in the treatment of nasal type extra-nodal NK/T-cell lymphoma in a non-Asian population. Leuk Lymphoma. 2016;57:2575–83.
52. Suzuki R. NK/T cell lymphoma: updates in therapy. Curr Hematol Malig Rep. 2018;13:7–12.
53. Yamaguchi M, Suzuki R, Oguchi M. Advances in the treatment of extranodal NK/T-cell lymphoma, nasal type. Blood. 2018;131:2528.
54. Kharfan-Dabaja MA, Kumar A, Ayala E, et al. Clinical practice recommendations on indication and timing of hematopoietic cell transplantation in mature T-cell and NK/T-cell lymphomas: an international collaborative effort on behalf of the Guidelines Committee of the American Society for Blood and Marrow Transplantation. Biol Blood Marrow Transplant. 2017;23:1826–38.
55. Kwong Y-L. Hematopoietic stem cell transplantation in natural killer cell lymphoma and leukemia. Int J Hematol. 2010;92:702–7.

56. Buchbinder EI, Desai A. CTLA-4 and PD-1 pathways: similarities, differences, and implications of their inhibition. Am J Clin Oncol. 2016;39:98–106.
57. Green MR, Rodig S, Juszczynski P, et al. Constitutive AP-1 activity and EBV infection induce PD-L1 in Hodgkin lymphomas and post-transplant lymphoproliferative disorders: implications for targeted therapy. Clin Cancer Res. 2012;18:1611–8.
58. Chen BJ, Chapuy B, Ouyang J, et al. PD-L1 expression is characteristic of a subset of aggressive B-cell lymphomas and virus-associated malignancies. Clin Cancer Res. 2013;19:3462–73.
59. Kwong YL, Chan TSY, Tan D, et al. PD1 blockade with pembrolizumab is highly effective in relapsed or refractory NK/T-cell lymphoma failing L-asparaginase. Blood. 2017;129:2437–42.
60. Chan TSY, Li J, Loong F, et al. PD1 blockade with low-dose nivolumab in NK/T cell lymphoma failing L-asparaginase: efficacy and safety. Ann Hematol. 2018;97:193–6.
61. Li X, Cheng Y, Zhang M, et al. Activity of pembrolizumab in relapsed/refractory NK/T-cell lymphoma. J Hematol Oncol. 2018;11:15.
62. Lokhorst HM, Plesner T, Laubach JP, et al. Targeting CD38 with daratumumab monotherapy in multiple myeloma. N Engl J Med. 2015;373:1207–19.
63. Hari P, Raj RV, Olteanu H. Targeting CD38 in refractory extranodal natural killer cell-T-cell lymphoma. N Engl J Med. 2016;375:1501–2.
64. Gulley ML. Molecular diagnosis of Epstein-Barr virus-related diseases. J Mol Diagn. 2001;3:1–10.
65. Kimura H, Hoshino Y, Kanegane H, Tsuge I, Okamura T, Kawa K, Morishima T. Clinical and virologic characteristics of chronic active Epstein-Barr virus infection. Blood. 2001;98:280–6.
66. Okano M, Kawa K, Kimura H, et al. Proposed guidelines for diagnosing chronic active Epstein-Barr virus infection. Am J Hematol. 2005;80:64–9.
67. Kimura H, Ito Y, Kawabe S, et al. EBV-associated T/NK-cell lymphoproliferative diseases in nonimmunocompromised hosts: prospective analysis of 108 cases. Blood. 2012;119:673–86.
68. Ishihara S, Okada S, Wakiguchi H, Kurashige T, Hirai K, Kawa-Ha K. Clonal lymphoproliferation following chronic active Epstein-Barr virus infection and hypersensitivity to mosquito bites. Am J Hematol. 1997;54:276–81.
69. Ruiz-Maldonado R, Parrilla F, Orozco-Covarrubias M, et al. Edematous, scarring vasculitic panniculitis: a new multisystemic disease with malignant potential. J Am Acad Dermatol. 1995;32:37–44.
70. Iwatsuki K, Xu Z, Takata M, et al. The association of latent Epstein-Barr virus infection with hydroa vacciniforme. Br J Dermatol. 1999;140:715–21.
71. Quintanilla-Martinez L, Kumar S, Fend F, et al. Fulminant EBV(+) T-cell lymphoproliferative disorder following acute/chronic EBV infection: a distinct clinicopathologic syndrome. Blood. 2000;96:443–51.
72. Kawa K, Sawada A, Sato M, et al. Excellent outcome of allogeneic hematopoietic SCT with reduced-intensity conditioning for the treatment of chronic active EBV infection. Bone Marrow Transplant. 2011;46:77–83.
73. Papadopoulos EB, Ladanyi M, Emanuel D, Mackinnon S, Boulad F, Carabasi MH, Castro-Malaspina H, Childs BH, Gillio AP, Small TN. Infusions of donor leukocytes to treat Epstein-Barr virus-associated lymphoproliferative disorders after allogeneic bone marrow transplantation. N Engl J Med. 1994;330:1185–91.
74. Leen AM, Myers GD, Sili U, et al. Monoculture-derived T lymphocytes specific for multiple viruses expand and produce clinically relevant effects in immunocompromised individuals. Nat Med. 2006;12:1160–6.
75. Heslop HE, Slobod KS, Pule MA, et al. Long-term outcome of EBV-specific T-cell infusions to prevent or treat EBV-related lymphoproliferative disease in transplant recipients. Blood. 2010;115:925–35.
76. Barth MJ, Chu Y, Hanley PJ, Cairo MS. Immunotherapeutic approaches for the treatment of childhood, adolescent and young adult non-Hodgkin lymphoma. Br J Haematol. 2016;173:597–616.
77. Cho S-G, Kim N, Sohn H-J, et al. Long-term outcome of extranodal NK/T cell lymphoma patients treated with postremission therapy using EBV LMP1 and LMP2a-specific CTLs. Mol Ther J Am Soc Gene Ther. 2015;23:1401–9.
78. Bollard CM, Gottschalk S, Torrano V, et al. Sustained complete responses in patients with lymphoma receiving autologous cytotoxic T lymphocytes targeting Epstein-Barr virus latent membrane proteins. J Clin Oncol. 2014;32:798–808.

Cutaneous T-Cell Lymphomas in Childhood and Adolescence

23

Rein Willemze

Introduction

Malignant lymphomas can present in the skin either as primary or secondary manifestation of the disease. The term primary cutaneous lymphoma (PCL) refers to a heterogeneous group of cutaneous T-cell lymphomas (CTCLs) and cutaneous B-cell lymphomas (CBCLs) that present in the skin with no evidence of extracutaneous disease at the time of diagnosis [1]. After the gastrointestinal lymphomas, PCLs are the second most common group of extranodal non-Hodgkin lymphomas, with an estimated annual incidence of almost 10 per million persons [2]. PCLs must be distinguished from nodal or systemic malignant lymphomas involving the skin secondarily, which often have another clinical behavior, have a different prognosis, and require a different therapeutic approach. In recent lymphoma classifications, PCLs are therefore included as separate entities. The frequency and prognosis of the different types of CTCL and CBCL are presented in Table 23.1. In the Western world, CTCLs constitute ~75–80% of all PCLs and CBCLs ~20–25% [1, 3]. However, different distributions have been observed in other parts of the world. In Southeast Asian countries, CTCLs other than MF, in particular, Epstein-Barr virus (EBV)-associated natural killer (NK)/T-cell lymphomas, are much more common than in Western countries, while CBCLs are much more uncommon [4, 5].

PCLs generally affect adult and often elderly patients, while PCLs arising in childhood and adolescence are rare with an estimated annual incidence of 0.1 and 0.3 per million persons in age groups 0–9 and 10–19 years, respectively [2]. Consistently, published reports on pediatric PCLs are sparse. There are only few reviews or studies including more than 50 patients with a pediatric PCL [6–11]. Most reports concern small cohorts or case studies. The most common types of pediatric PCL with over 100 reported cases are MF and primary cutaneous CD30-positive lymphoproliferative disorders (LPDs) (Table 23.1). Other CTCLs that may present in childhood, although less commonly (50–100 reported cases), are subcutaneous panniculitis-like T-cell lymphoma (SPTCL) and, particularly in Central and South America and Asia, EBV-positive lymphoproliferative disorders of childhood, now commonly referred to as chronic active EBV infections (CEABV). However, pediatric cases of Sezary syndrome, extranodal NK/T-cell lymphoma, primary cutaneous CD8-positive aggressive epidermotropic cytotoxic T-cell lymphoma, and primary cutaneous gamma/delta T-cell lymphoma are extremely rare or have not been reported at all. Regarding CBCL, there are more than 20 published cases of primary cutaneous marginal zone lymphoma presenting in childhood, while pediatric cases of primary cutaneous follicle center lymphoma are extremely rare and pediatric cases of primary cutaneous diffuse large B-cell lymphoma, leg type have never been reported [8, 12, 13]. In addition, B-lymphoblastic lymphoma/leukemia not uncommonly presents in the skin, usually the head, and may be the first and sometimes even the only manifestation of the disease [14].

This chapter will focus on the clinicopathologic features, treatment, and prognosis of the more common types of CTCL presenting in childhood and adolescence.

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Table 23.1 WHO-EORTC classification for cutaneous lymphomas: relative frequency and survival in adults and number of reported cases in children

	Frequency in adults (%)	5-year DSS	Reported cases in children[c]
Cutaneous T-cell lymphoma			
Mycosis fungoides (MF)	44	88	++++
Variants of MF			
Folliculotropic MF	5	80	++
Pagetoid reticulosis	<1	100	+
Granulomatous slack skin	<1	100	+
Sézary syndrome	3	30	–
Primary cutaneous CD30-positive LPD			
Cutaneous anaplastic large cell lymphoma	8	95	++
Lymphomatoid papulosis	12	100	++++
Subcutaneous panniculitis-like T-cell lymphoma	1	82	+++
Primary cutaneous extranodal NK/T-cell lymphoma, nasal-type	<1	<20	–
Primary cutaneous γ/δ T-cell lymphoma	<1	<20	+
Primary cutaneous CD8+ aggressive epidermotropic cytotoxic T-cell lymphoma[a]	<1	<20	–
Primary cutaneous acral CD8+ T-cell lymphoma[b]	<1	100%	–
Primary cutaneous CD4+ small/medium T-cell LPD[a]	4	100%	+
Cutaneous B-cell lymphoma			
Primary cutaneous marginal zone B-cell lymphoma	8	99	++
Primary cutaneous follicle center lymphoma	11	95	–
Primary cutaneous diffuse large B-cell lymphoma, leg type	4	50	–

Adapted from [1]
DSS disease-specific survival, *LPD* lymphoproliferative disorder
[a]Provisional entities
[b]New provisional entity in the revised 2017 WHO classification [3]
[c]Number of reported cases: 0–1; +: 2–10; ++: 11–50; +++: 51–100; ++++: >100

Mycosis Fungoides

Mycosis fungoides (MF) is the most common type of cutaneous T-cell lymphoma (CTCL) and accounts for approximately 50% of all PCLs [1]. Patients with classical MF present with patches and plaques that are preferentially located on the buttocks and other covered sites of the trunk and limbs (sun-protected areas). Histologically, these early stages are characterized by superficial band-like or lichenoid infiltrates of small to medium-sized atypical T-cells with cerebriform and sometimes hyperchromatic nuclei, which characteristically infiltrate into the epidermis (epidermotropism). The neoplastic T-cells usually have a mature CD3+, CD4+ and, CD8− T-cell phenotype. Most patients have a protracted clinical course over years or even decades. However, a proportion of patients may develop nodules or tumors and eventually progress to extracutaneous disease [15, 16].

MF is also one of the two most common types of CTCL in children and adolescents with over 300 reported cases [10, 11, 17–22]. The male-to-female ratio in juvenile MF varies between 1.5:1 and 1:1. In most series the median age at diagnosis varies between 9 and 13 years, but infants as young as 10 months with MF have been reported [23]. Clinically, juvenile MF may present with erythematous patches and plaques typical of classic MF but more often shows an atypical clinical presentation, such as hypopigmented, hyperpigmented, or folliculotropic MF (Figs. 23.1 and 23.2). Hypopigmented MF, which is often found in dark-skinned individuals, is the most common variant in children and adolescents [10, 11, 17, 20, 24]. Patients present with asymptomatic hypopigmented patches that are mainly located on trunk and extremities. Differentiation from various benign skin diseases, including vitiligo, lichen sclerosus et atrophicus,

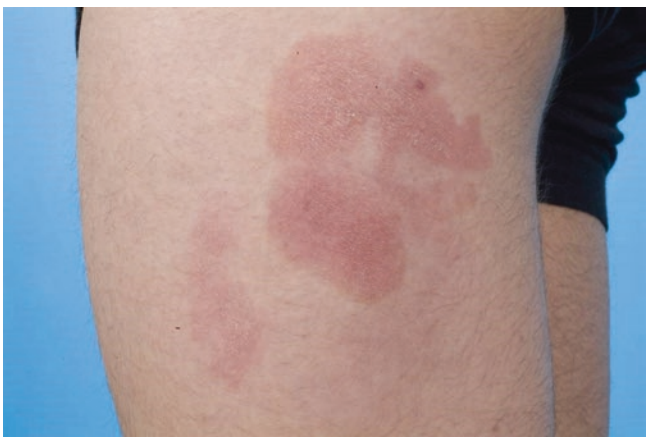

Fig. 23.1 Mycosis fungoides. A 15-year-old boy with multiple patches and slightly infiltrated plaques on the left upper leg for more than 10 years

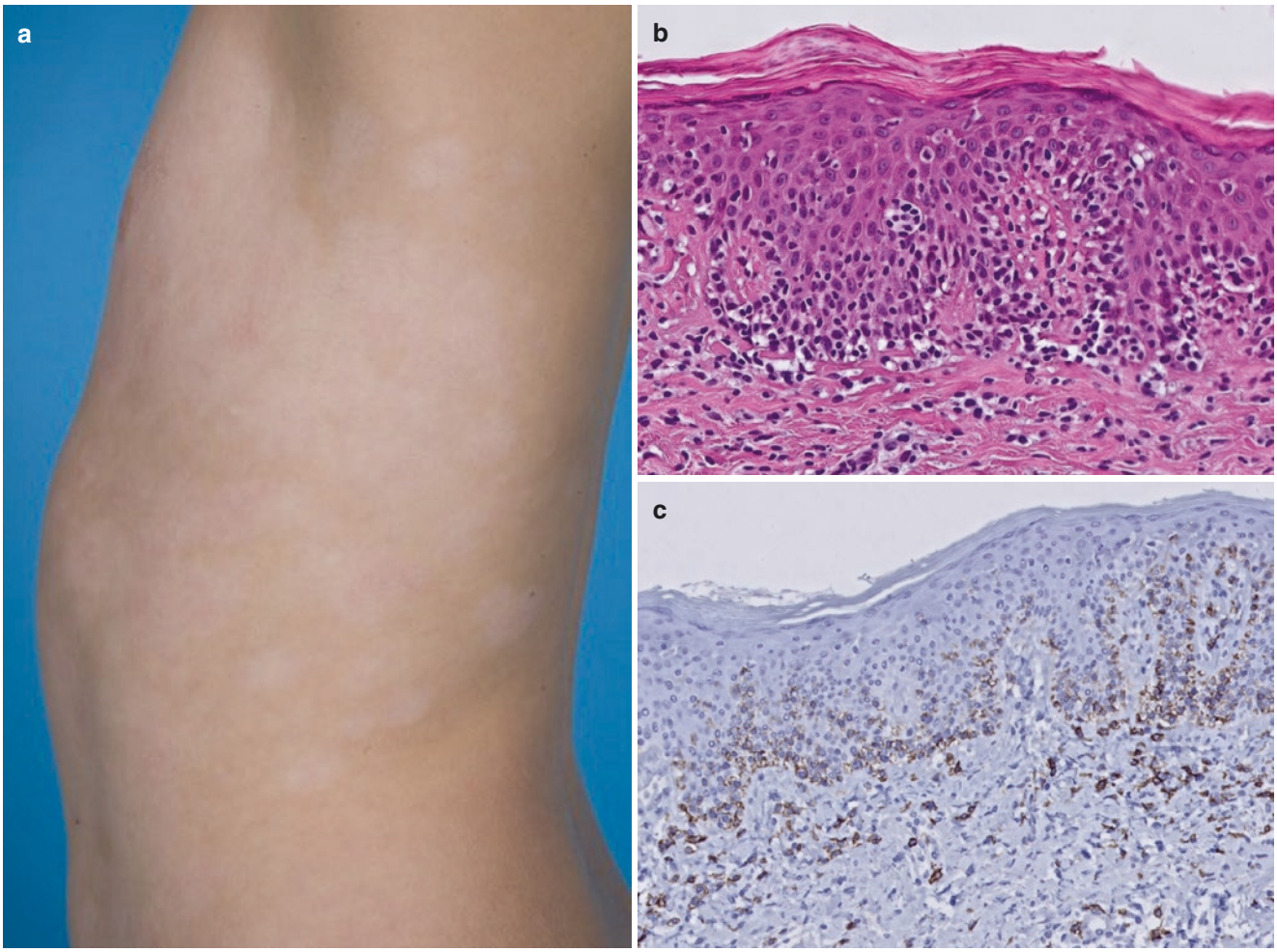

Fig. 23.2 Hypopigmented mycosis fungoides. A 9-year-old girl with generalized hypopigmented patches (**a**); Histologic examination shows infiltration of the epidermis along the basal layer (**b**); the neoplastic T-cells are CD8+ (**c**)

pityriasis alba, and post-inflammatory hypopigmentation, may be difficult. In Caucasians, hypopigmented lesions usually coexist with erythematous lesions, as observed in classic MF. Histopathology shows the typical features of early patch-stage MF. However, in contrast to classic MF, hypopigmented MF usually has a CD8+ cytotoxic T-cell phenotype (Fig. 23.2) [10, 11, 24].

The prognosis of patients with juvenile MF is usually excellent [10, 11, 20]. Most patients present with early patch/plaque stage disease (stage IA–IB) and rarely with more advanced stage MF (stage IIB–IV) [11]. In addition, progression from early-stage MF to tumor stage MF or beyond is rarely observed [25–27]. Patients respond very well to skin-directed therapies, such as topical steroids, narrowband UVB (NB-UVB), or (bath) PUVA, but recurrences after treatment are common. In patients with hypopigmented MF, NB-UVB is the preferred mode of treatment, since its efficacy for patch-stage disease has been well-established and because it has fewer side effects and less carcinogenic, when compared to PUVA therapy [11, 20, 28].

Variants of MF

Apart from classical MF, many clinical and/or histopathologic variants of MF mimicking a wide variety of inflammatory skin diseases have been described [29–31]. Most variants, including hypopigmented MF described above, have a clinical behavior similar to that of classic MF and have therefore not been classified separately. In recent classifications, only folliculotropic MF (FMF), pagetoid reticulosis, and granulomatous slack skin are recognized as distinct variants of MF, because of their distinctive clinicopathologic features, clinical behavior, and/or prognosis [1, 3].

Folliculotropic MF

Folliculotropic MF (FMF) is characterized by the presence of folliculotropic infiltrates, often with sparing of the interfollicular epidermis and preferential involvement of the head and neck area [1]. Patients may present with (grouped) follicular

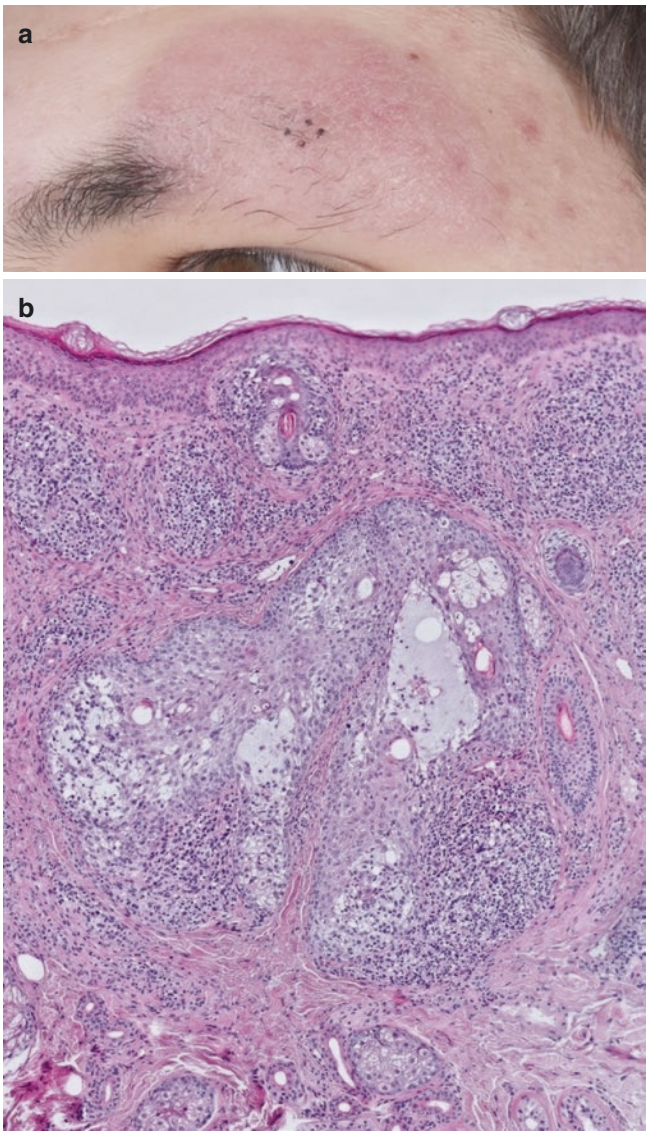

Fig. 23.3 Folliculotropic mycosis fungoides. A 18-year-old man with a slightly infiltrated plaque with associated alopecia in the left eyebrow (**a**); histology shows perifollicular infiltrates with infiltration of the follicular epithelium and extensive follicular mucinosis (**b**)

papules, acneiform lesions, indurated plaques, or tumors [32–36]. Infiltrated plaques or tumors in the eyebrow region with concurrent hair loss are a highly characteristic feature (Fig. 23.3). Some patients may show keratosis pilaris-like lesions that are mainly localized on trunk and extremities (Fig. 23.4) [37]. The skin lesions are often associated with alopecia. Histologically, FMF is characterized by the presence of perifollicular to diffuse infiltrates with variable infiltration of the follicular epithelium by small- and medium-sized or sometimes large T-cells with cerebriform and hyperchromatic nuclei [32, 38]. Many cases show mucinous degeneration of the follicular epithelium (follicular mucinosis), but cases without follicular mucinosis have been

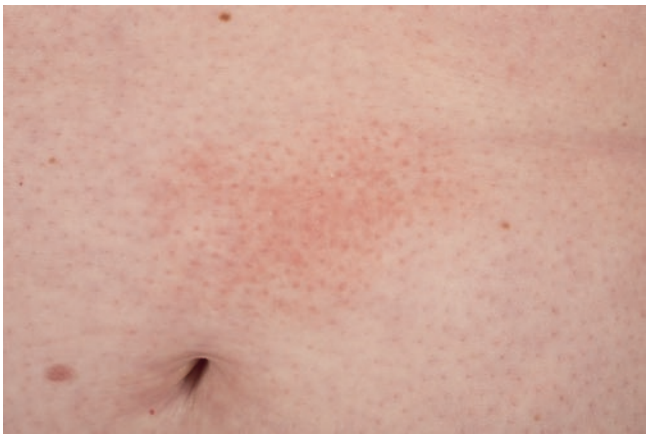

Fig. 23.4 Folliculotropic mycosis fungoides. A 17-year-old female with keratosis pilaris-like lesions on the abdomen

described as well [39]. In virtually all cases, the neoplastic cells in FMF have a CD3+, CD4+, and CD8− T-cell phenotype as in classic MF [38].

FMF mostly presents in adults, but has also been reported in children and adolescents [11, 36]. In these young patients, FMF usually presents with grouped follicular papules or follicle-based patches with associated alopecia on the arms, the legs, or the trunk (Fig. 23.4). Because of the deep localization of the perifollicular infiltrates, bath or systemic PUVA is the preferred type of treatment in these patients with early-stage FMF [11]. Presentation with or progression to more advanced disease with infiltrated plaques or tumors is very uncommon [11, 25, 40]. The relationship between FMF and the so-called idiopathic form of follicular mucinosis (alopecia mucinosa), which may show a similar clinical presentation, is a matter of debate. In adults, this idiopathic form of follicular mucinosis often presents with widespread and persistent lesions and is generally considered as a variant of MF [41, 42]. In children and adolescents, idiopathic follicular mucinosis usually presents with one or few localized patches with follicular accentuation, which can be treated effectively with topical steroids or phototherapy and has an excellent prognosis. These cases should not be equated with MF [43, 44].

Pagetoid Reticulosis (Woringer-Kolopp Disease)

Pagetoid reticulosis is a rare unilesional variant of MF, clinically characterized by the presence of a solitary, slowly progressive, psoriasiform or hyperkeratotic patch or plaque, which is usually localized on an extremity, particularly hands or feet (Fig. 23.5a) [1, 3]. Histologically, these lesions show a hyperplastic epidermis with marked infiltration by small- to medium-sized atypical pagetoid cells, arranged singly or

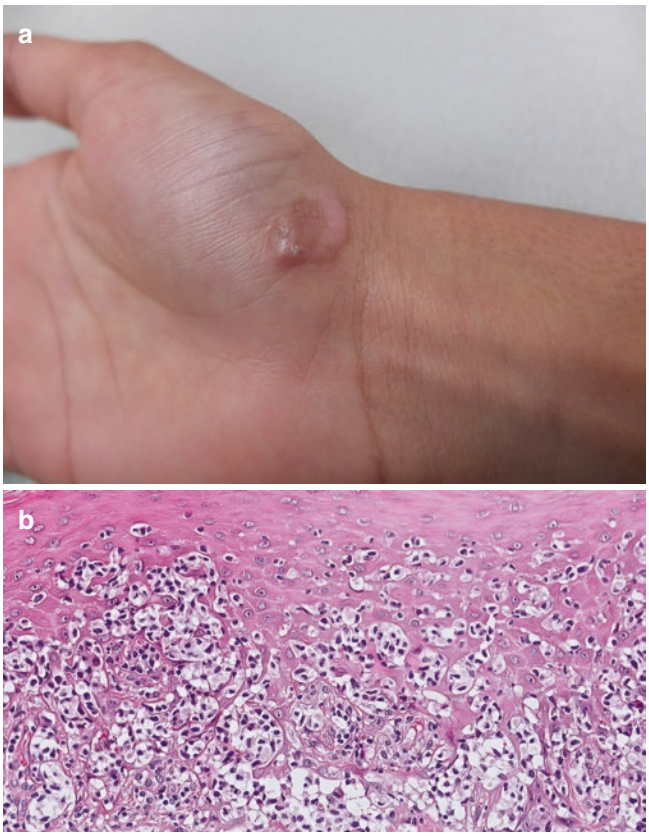

Fig. 23.5 Pagetoid reticulosis. A 14-year-old boy with a slowly progressive plaque on the right wrist (**a**); histologic examination shows epidermal hyperplasia and extensive epidermotropism of small- to medium-sized atypical lymphocytes in a pagetoid pattern (**b**)

in nests or clusters (Fig. 23.5b). The superficial dermis may have an infiltrate of mostly small lymphocytes but rarely contains neoplastic T-cells. The neoplastic T-cells may show either a CD3+, CD4−, CD8+, or less commonly a CD3+, CD4+, CD8−, or CD3+, CD4−, and CD8− phenotype. Cases with a CD8+ or CD4−, CD8− phenotype express cytotoxic proteins. CD30 is often expressed [45].

Publications on pediatric pagetoid reticulosis are extremely rare, but they appear to have the same clinicopathologic features as adult cases. The original report by Woringer and Kolopp concerned a 13-year-old with an erythematous scaly well-demarcated patch on the left forearm that had been present for 6 years and was completely excised. Other reported cases concern a 5-year-old boy with an erythematous scaly patch on the left buttock and a 6-year-old boy with an erythematous plaque on his left thigh, which had slowly progressed since few months after birth [46, 47]. In the series published by Fink-Puches et al., three of 24 MF cases had pagetoid reticulosis, but further details are not provided [6]. The preferred mode of treatment in these patients is radiotherapy or surgical excision. The prognosis of pagetoid reticulosis is excellent; extracutaneous dissemination or disease-related deaths have never been reported [48].

Granulomatous Slack Skin

Granulomatous slack skin (GSS) is a very rare variant of MF, clinically characterized by the slow development of pendulous folds of lax skin in the major skin folds (axilla and groins) and histologically by the presence of dense infiltrates of small clonal CD4-positive T-cells admixed with numerous macrophages and many scattered multinucleated giant cells [1, 49]. Loss of elastic tissue, elastophagocytosis, and emperipolesis (engulfment of lymphocytes) by multinucleated cells are commonly observed. Extracutaneous dissemination is rare, but in approximately one-third of patients an association with other malignant lymphomas, particularly MF and Hodgkin lymphoma, has been reported [50]. Treatment of GSS is unsatisfactory. Patients have been treated with PUVA, radiotherapy, surgical excision, interferon-α, and other systemic therapies, but complete responses have never been reported. There are only few reports of GSS in children or adolescents and, in two of four reported patients, an association with another type of lymphoma was reported [51–55]. Tronnier et al. described a 13-year-old patient with GSS that was preceded by a diagnosis of folliculotropic MF for 2 years [55]. Long-term follow-up in an 11-year-old boy who presented with widespread GSS lesions showed progression to a systemic CD30-positive peripheral T-cell lymphoma, which had a fatal outcome despite chemotherapy [51, 52]. Because of the increased risk of a second malignant lymphoma, long-term follow-up is mandatory in patients with GSS [56].

Primary Cutaneous CD30-Positive T-Cell Lymphoproliferative Disorders

Primary cutaneous CD30-positive lymphoproliferative disorders (LPDs) are the second most common group of the cutaneous T-cell lymphomas, accounting for approximately 25% of all CTCLs (Table 23.1) [1]. This group includes primary cutaneous anaplastic large lymphoma (C-ALCL) and lymphomatoid papulosis (LyP), which show overlapping clinical, histologic, and phenotypic features and form a spectrum of disease [1]. The clinical appearance and course are used as decisive criteria for the definite diagnosis and choice of treatment. Both C-ALCL and LyP have an excellent prognosis, with a 10-year survival of 90% and almost 100%, respectively [57]. Also in children, primary cutaneous CD30-positive LPD represents one of the two most common subgroups of CTCL. In the Dutch registry, primary cutaneous CD30-positive LPDs account for 64 of 91 (70%) CTCL patients younger than 20 years of age. In this age group, LyP is much more common than C-ALCL. From the 503 patients with LyP in our database, 53 cases (10.5%) were younger than 20 years of age, compared to 11 of 283 patients (3.9%)

with a diagnosis of C-ALCL. In a study of the Mayo Clinic on 123 patients with LyP, a similar proportion (14 patients; 11%) were in the pediatric age group [58].

Lymphomatoid Papulosis (LyP)

Lymphomatoid papulosis (LyP) is a chronic recurrent condition clinically characterized by the presence of self-healing papular, papulonecrotic, and/or nodular skin lesions with histologic features of a (CD30-positive) CTCL [1, 3]. Although there has been continued discussion whether LyP is a malignant, a premalignant, or a benign condition, LyP is currently regarded as a low-grade malignant CTCL. The histologic picture of LyP is extremely variable, and in recent classifications six histologic subtypes are recognized: five histologic subtypes resembling different types of CTCL, including C-ALCL (types A and C), plaque stage MF (type B), aggressive CD8+ CTCL (type D) and angio-centric lymphomas (type E), and a new subtype characterized by the presence of chromosomal rearrangements involving the *DUSP-IRF4* locus on 6p25.3 [59, 60]. The same rearrangement is found in approximately 25% of C-ALCL [61]. Recognition of these different types of LyP is important to avoid misdiagnosis of other often more aggressive types of CTCL, but has no therapeutic or prognostic implications. LyP should not only be differentiated from other types of CTCL, but also from a wide variety of infectious and inflammatory dermatoses that can contain substantial numbers of CD30-positive cells [60].

LyP most often occurs in adults (median age, 45 years), but children may also be affected. In different cohort studies, the male-to-female ratio varied between 1.2 and 1.5, and the median age at diagnosis was between 7.5 and 12 years of age [22, 58, 62, 63]. The youngest patient published to date is an 8-month-old child. An association with atopy has been reported in 30–60% of children with LyP [62, 63].

In most children LyP has the same clinicopathologic features as in adults. Characteristically, skin lesions in different stages of evolution coexist (Fig. 23.6) [57, 64]. The number of lesions may vary from a few to more than a hundred. Individual skin lesions disappear within 3–12 weeks and may leave behind superficial scars. In some children, LyP may start with large, rapidly growing ulcerating lesions in addition to papular lesions. Spontaneous resolution of these large lesions may take months rather than weeks. During follow-up, these large lesions stop to develop and LyP continues with small papular or papulonecrotic lesions or disappears completely. The duration of the disease may vary from several months to decades. Studies of LyP in adults indicate that it may be associated with another type of malignant lymphoma, generally MF, C-ALCL, or Hodgkin lymphoma in up to 20% [57, 60, 65]. There are also few reports of children with LyP, who developed a second malignant lymphoma during follow-up, most commonly a C-ALCL [57, 62, 63].

Treatment of LyP is generally unsatisfactory. Since a curative therapy is not available and none of the available treatment modalities affects the natural course of the disease, the short-term benefits of active treatment should be balanced carefully against potential side effects [57, 66]. In patients with relatively few non-scarring lesions, an expectant policy can be followed. In the case of cosmetically disturbing lesions (e.g., scarring or many papulonodules), low-dose oral methotrexate (MTX; 5–20 mg/week) is the

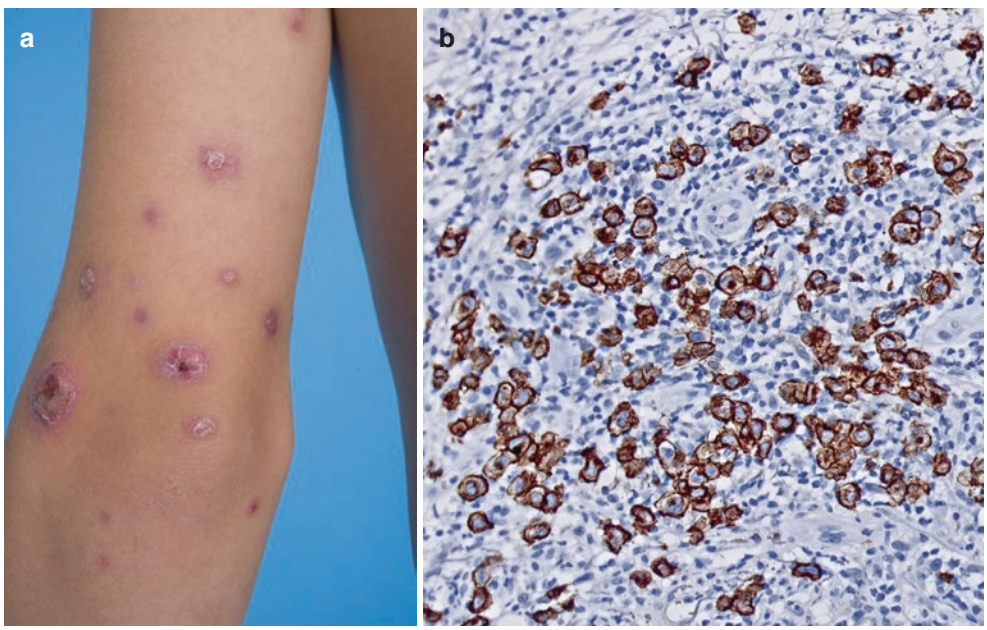

Fig. 23.6 Lymphomatoid papulosis. A 9-year-old boy with papules and nodules on arms and legs in various stages of evolution (**a**); histology shows a dense inflammatory infiltrate with many scattered large atypical CD30+ cells (**b**)

most effective therapy for reducing the number of skin lesions and can also be used safely in children [67]. PUVA therapy is also effective, but is less attractive in case maintenance treatment is required. LyP has an excellent prognosis in the vast majority of patients. However, because of the risk of late secondary lymphomas, long-term follow-up is advised.

Primary Cutaneous Anaplastic Large Cell Lymphoma (C-ALCL)

Most C-ALCL patients present with solitary or localized often ulcerating nodules or tumors, histologically showing dense dermal infiltrates of large CD30-positive tumor cells with an anaplastic or pleomorphic cytomorphology [1, 3]. In most cases the neoplastic cells have an activated CD4+ T-cell phenotype. Approximately 10% of patients present with multifocal lesions. Similarly to LyP, the skin lesions may show partial or complete spontaneous regression [57]. C-ALCL presenting with solitary or localized skin lesions are treated with radiotherapy or surgical excision, while patients presenting with multifocal skin lesions can best be treated with low-dose MTX, as in LyP, or with low-dose radiotherapy in the case of only a few lesions [57, 66, 68]. C-ALCL frequently relapses in the skin, but extracutaneous dissemination is uncommon, and their prognosis is usually excellent with a 10-year disease-related survival of approximately 90% [57, 69].

C-ALCL may also occur in childhood, but it is much less common than LyP [57]. Reports on pediatric C-ALCL are few, but clearly show that children have the same clinicopathologic features and the same favorable prognosis as adults (Fig. 23.7) [57, 70–72]. None of these patients developed extracutaneous disease or had a fatal outcome. Most patients had been treated with aggressive combination chemotherapy, but cutaneous relapses were observed in almost all of them. Current evidence indicates that systemic chemotherapy is no longer warranted in such patients. As in adult patients, local radiotherapy and surgical excision are the first choice of treatment and, in case of spontaneous resolution, an expectant policy is even justified [57, 66]. Systemic chemotherapy is only recommended in exceptional cases developing extracutaneous disease.

It is important to differentiate pediatric C-ALCL from systemic ALCL, which is relatively common in this age group and frequently shows secondary cutaneous involvement [72]. Unlike this systemic ALCL, C-ALCL usually does not carry the t(2;5) chromosomal translocation and does not express ALK (anaplastic lymphoma kinase) [73].

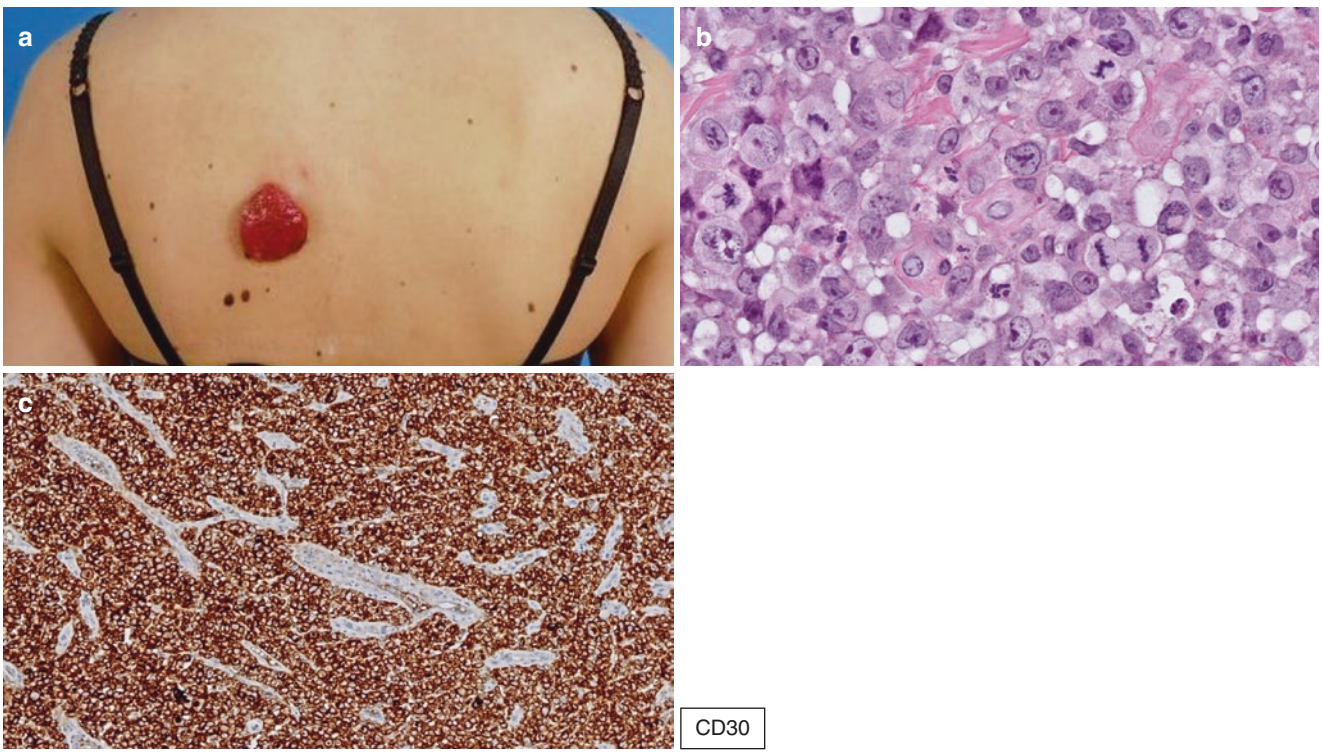

Fig. 23.7 Primary cutaneous anaplastic large cell lymphoma. A 17-year-old female presenting with a large ulcerating tumor on the back (**a**); histologic examination shows a monotonous infiltrate of large cells with anaplastic morphology (**b**); the tumor cells show a diffuse staining for CD30 (**c**)

Expression of ALK protein therefore strongly suggests secondary cutaneous involvement of a systemic ALK-positive ALCL. However, unusual cases of ALK+ C-ALCL have been reported both in children and adults, and many of these cases had an excellent prognosis [74–77].In one study, five of six pediatric cases of ALK-positive C-ALCL had presented with a solitary lesion. Surgical excision, followed by local radiotherapy in two of them, resulted in sustained complete remission in all patients [75].

Subcutaneous Panniculitis-Like T-Cell Lymphoma (SPTCL)

SPTCL was initially defined as a cytotoxic T-cell lymphoma with either an α/β or a γ/δ T-cell phenotype, which preferentially infiltrates the subcutaneous tissue, and often complicated by a hemophagocytic syndrome (HPS) with an aggressive clinical course, and that should therefore be treated with aggressive multi-agent chemotherapy [78]. However, more recent studies showed clinical, histological, and immunophenotypical differences between SPTCL with an α/β T-cell phenotype and SPTCL with a γ/δ T-cell phenotype, suggesting that these may represent different entities [79–81]. In recent classifications, the term SPTCL is therefore only used for cases with an α/β T-cell phenotype, while cases expressing the γ/δ T-cell receptor are reclassified as primary cutaneous gamma/delta T-cell lymphoma (PCGD-TCL) [1, 3]. Differentiation between SPTCL and PCGD-TCL is important, since both conditions have a different prognosis and require a different therapeutic approach.

SPTCL is a rare type of lymphoma accounting for <1% of all CTCL. It is slightly more common in females than in males and may affect both children and adults [1, 82]. Clinically, patients present with solitary, but more commonly multiple nodules or deeply seated plaques resembling panniculitis, which mainly involve the legs and the arms trunk and less commonly the face. Systemic symptoms such as fever, fatigue, and weight loss, and laboratory abnormalities, including cytopenias and elevated liver function tests, are common, but a frank hemophagocytic syndrome (HPS) is observed in only 15–20% of patients [81]. Up to 20% of patients may have an associated autoimmune disease, most commonly systemic lupus erythematosus [81]. The differential diagnosis with lupus panniculitis may sometimes be challenging [83]. Histologically, SPTCL shows strictly subcutaneous infiltrates resembling a lobular panniculitis with rimming of individual fat cells by small- to medium-sized neoplastic T-cells, which usually have a mature CD3+, CD4−, CD8+, and CD56− T-cell phenotype and typically express βF1, but not TCR γ/TCRδ facilitating differentiation from PCGD-TCL (Fig. 23.8) [81].

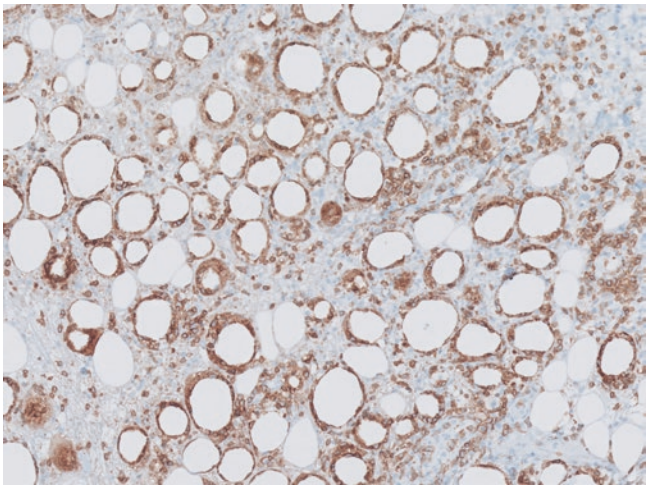

Fig. 23.8 Subcutaneous panniculitis-like T-cell lymphoma. Histologic examination shows rimming of adipocytes by CD8-positive neoplastic T-cells

Reports on pediatric cases of SPTCL are rare. In a series of 63 SPTCL patients, 12 of 63 patients (19%) were 20 years or younger [81]. The youngest reported patients were 4 and 5 months old, respectively [82, 84]. The clinicopathologic features of pediatric SPTCL are similar to those described in adult patients. Most cases of SPTCL have a favorable prognosis, particularly if not associated with a HPS. One study reported 5-year overall survival (OS) rates of 91% and 46% in SPTCL patients without and with an HPS, respectively [81]. Most patients with pediatric SPTCL have been treated with combination chemotherapy, in some of them followed by an allogeneic stem cell transplant [84]. More recent studies indicate that in SPTCL without associated HPS, systemic steroids or other immunosuppressive agents (cyclosporine, MTX) should be considered first, while combination chemotherapy should be reserved for cases with progressive disease not responding to immunosuppressive therapy and cases with associated HPS [81, 85]. In cases presenting with a solitary skin lesion, radiotherapy can be used. Bexarotene may also be effective in SPTCL [86]. In some cases of pediatric SPTCL, the subcutaneous lesions disappeared spontaneously without active treatment [87–89].

EBV-Positive Lymphoproliferative Disorders of Childhood

EBV-positive lymphoproliferative disorders of childhood include hydroa vacciniforme-like lymphoproliferative disorders (HV-like LPD) and hypersensitivity reactions to mosquito bites (HMB). Both are cutaneous manifestations of chronic active EBV (CAEBV) infection with a risk for progression to systemic EBV-positive T-cell or NK-cell lymphoma [3]. HV-like LPD is used as an encompassing term

for cases previously referred to as HV and HV-like lymphoma. These disorders are seen mainly in children and adolescents from Asia, or in indigenous populations from Central and South America and Mexico [90, 91]. Both conditions are rare in adults.

Clinically, classic HV presents with a papulovesicular eruption on sun-exposed skin areas, in particular, the face, the earlobes, and the back of the hands, often with seasonal activity, but without systemic symptoms [92]. In more severe cases (HV-like lymphoma), skin lesions are localized in sun-exposed and nonexposed skin areas, facial swelling and extensive ulceration are common, and systemic symptoms, such as fever, wasting, lymphadenopathy, and hepatosplenomegaly, may be present [93, 94]. Patients with mosquito bite allergy typically show bullous lesions that become necrotic at the site of the mosquito bite and may demonstrate similar systemic symptoms as seen in patients with HV-like lymphoma [90, 95].

Histologically, skin lesions show variable degrees of epidermal spongiosis, necrosis, and ulceration, and a variably dense dermal infiltrate mainly consisting of small- to medium-sized lymphocytes [93]. Angio-centricity and angio-destruction are frequently found. The number of EBER-positive cells is variable. Most cases of HV-like LPD have a CD8+ T-cell phenotype, while hypersensitivity reactions to mosquito bites more often have a NK-cell phenotype [96].

Most reported cases run an aggressive clinical course and have a poor prognosis, in particular patients presenting with systemic manifestations [93]. However, patients may have recurrent skin lesions for many years before progression to systemic lymphoma. There is no standard treatment for these conditions. Most reported patients have been treated with multi-agent chemotherapy, but sustained complete remissions are rarely achieved [96]. In patients with only skin lesions, a conservative approach should be considered.

Conclusions

Cutaneous T-cell lymphomas (CTCLs) usually affect adult and elderly patients and are rare in childhood and adolescence. CTCLs most commonly seen in children are mycosis fungoides and lymphomatoid papulosis. Pediatric CTCLs generally have the same clinicopathologic features and clinical behavior as their adult counterparts. Most patients with juvenile mycosis fungoides present with early patch/plaque stage disease, and progression to advanced stage disease is rarely observed. Diagnosis and treatment of these rare conditions can be challenging. Clinicopathologic correlation and a multidisciplinary approach with close collaboration between pediatric oncologists, dermatologists, and pathologists are the best guarantee for correct diagnosis and adequate treatment.

References

1. Willemze R, Jaffe ES, Burg G, et al. WHO-EORTC classification for cutaneous lymphomas. Blood. 2005;105:3768–85.
2. Criscione VD, Weinstock MA. Incidence of cutaneous T-cell lymphoma in the United States, 1973-2002. Arch Dermatol. 2007;143:854–9.
3. Swerdlow SH, Campo E, Harris NL, et al. World Health Organization classification of tumours of hematopoietic and lymphoid tissue (Revised 4th edition). Lyon: IARC Press; 2017.
4. Tan SH, Sim CS, Ong BH. Cutaneous lymphomas other than mycosis fungoides in Singapore: a clinicopathological analysis using recent classification systems. Br J Dermatol. 2003;149:542–53.
5. Park JH, Shin HT, Lee DY, et al. World Health Organization-European Organization for Research and Treatment of Cancer classification of cutaneous lymphoma in Korea: a retrospective study at a single tertiary institution. J Am Acad Dermatol. 2012;67:1200–9.
6. Fink-Puches R, Chott A, Ardigo M, et al. The spectrum of cutaneous lymphomas in patients less than 20 years of age. Pediatr Dermatol. 2004;21:525–33.
7. Boccara O, Blanche S, de PY, Brousse N, Bodemer C, Fraitag S. Cutaneous hematologic disorders in children. Pediatr Blood Cancer. 2012;58:226–32.
8. Kempf W, Kazakov DV, Belousova IE, Mitteldorf C, Kerl K. Paediatric cutaneous lymphomas: a review and comparison with adult counterparts. J Eur Acad Dermatol Venereol. 2015;29:1696–709.
9. Ceppi F, Pope E, Ngan B, Abla O. Primary cutaneous lymphomas in children and adolescents. Pediatr Blood Cancer. 2016;63:1886–94.
10. Castano E, Glick S, Wolgast L, et al. Hypopigmented mycosis fungoides in childhood and adolescence: a long-term retrospective study. J Cutan Pathol. 2013;40:924–34.
11. Hodak E, Amitay-Laish I, Feinmesser M, et al. Juvenile mycosis fungoides: cutaneous T-cell lymphoma with frequent follicular involvement. J Am Acad Dermatol. 2014;70:993–1001.
12. Amitay-Laish I, Feinmesser M, Ben-Amitai D, et al. Juvenile onset of primary low-grade cutaneous B-cell lymphoma. Br J Dermatol. 2009;161:140–7.
13. Amitay-Laish I, Tavallaee M, Kim J, et al. Paediatric primary cutaneous marginal zone B-cell lymphoma: does it differ from its adult counterpart? Br J Dermatol. 2017;176:1010–20.
14. Muljono A, Graf NS, Arbuckle S. Primary cutaneous lymphoblastic lymphoma in children: series of eight cases with review of the literature. Pathology (Phila). 2009;41:223–8.
15. van Doorn R, van Haselen CW, van Voorst Vader PC, et al. Mycosis fungoides: disease evolution and prognosis of 309 Dutch patients. Arch Dermatol. 2000;136:504–10.
16. Agar NS, Wedgeworth E, Crichton S, et al. Survival outcomes and prognostic factors in mycosis fungoides/Sezary syndrome: validation of the revised International Society for Cutaneous Lymphomas/European Organisation for Research and Treatment of Cancer staging proposal. J Clin Oncol. 2010;28:4730–9.
17. Nanda A, AlSaleh QA, Al-Ajmi H, et al. Mycosis fungoides in Arab children and adolescents: a report of 36 patients from Kuwait. Pediatr Dermatol. 2010;27:607–13.
18. Yazganoglu KD, Topkarci Z, Buyukbabani N, Baykal C. Childhood mycosis fungoides: a report of 20 cases from Turkey. J Eur Acad Dermatol Venereol. 2013;27:295–300.
19. Laws PM, Shear NH, Pope E. Childhood mycosis fungoides: experience of 28 patients and response to phototherapy. Pediatr Dermatol. 2014;31:459–64.
20. Boulos S, Vaid R, Aladily TN, Ivan DS, Talpur R, Duvic M. Clinical presentation, immunopathology, and treatment of juvenile-onset mycosis fungoides: a case series of 34 patients. J Am Acad Dermatol. 2014;71:1117–26.

21. Heng YK, Koh MJ, Giam YC, Tang MB, Chong WS, Tan SH. Pediatric mycosis fungoides in Singapore: a series of 46 children. Pediatr Dermatol. 2014;31:477–82.
22. Moon HR, Lee WJ, Won CH, et al. Paediatric cutaneous lymphoma in Korea: a retrospective study at a single institution. J Eur Acad Dermatol Venereol. 2014;28:1798–804.
23. Tsianakas A, Kienast AK, Hoeger PH. Infantile-onset cutaneous T-cell lymphoma. Br J Dermatol. 2008;159:1338–41.
24. El-Shabrawi-Caelen L, Cerroni L, Medeiros LJ, McCalmont TH. Hypopigmented mycosis fungoides: frequent expression of a CD8+ T-cell phenotype. Am J Surg Pathol. 2002;26:450–7.
25. Hickham PR, McBurney EI, Fitzgerald RL. CTCL in patients under 20 years of age: a series of five cases. Pediatr Dermatol. 1997;14:93–7.
26. Crowley JJ, Nikko A, Varghese A, Hoppe RT, Kim YH. Mycosis fungoides in young patients: clinical characteristics and outcome. J Am Acad Dermatol. 1998;38:696–701.
27. Quaglino P, Zaccagna A, Verrone A, Dardano F, Bernengo MG. Mycosis fungoides in patients under 20 years of age: report of 7 cases, review of the literature and study of the clinical course. Dermatology. 1999;199:8–14.
28. Trautinger F, Eder J, Assaf C, et al. European Organisation for Research and Treatment of Cancer consensus recommendations for the treatment of mycosis fungoides/Sezary syndrome - Update 2017. Eur J Cancer. 2017;77:57–74.
29. Nashan D, Faulhaber D, Stander S, Luger TA, Stadler R. Mycosis fungoides: a dermatological masquerader. Br J Dermatol. 2007;156:1–10.
30. Kazakov DV, Burg G, Kempf W. Clinicopathological spectrum of mycosis fungoides. J Eur Acad Dermatol Venereol. 2004;18:397–415.
31. Martinez-Escala ME, Gonzalez BR, Guitart J. Mycosis Fungoides variants. Surg Pathol Clin. 2014;7:169–89.
32. van Doorn R, Scheffer E, Willemze R. Follicular mycosis fungoides, a distinct disease entity with or without associated follicular mucinosis: a clinicopathologic and follow-up study of 51 patients. Arch Dermatol. 2002;138:191–8.
33. Gerami P, Rosen S, Kuzel T, Boone SL, Guitart J. Folliculotropic mycosis fungoides: an aggressive variant of cutaneous T-cell lymphoma. Arch Dermatol. 2008;144:738–46.
34. Lehman JS, Cook-Norris RH, Weed BR, et al. Folliculotropic mycosis fungoides: single-center study and systematic review. Arch Dermatol. 2010;146:607–13.
35. Wieser I, Wang C, Alberti-Violetti S, et al. Clinical characteristics, risk factors and long-term outcome of 114 patients with folliculotropic mycosis fungoides. Arch Dermatol Res. 2017;309:453–9.
36. van Santen S, Roach RE, van Doorn R, et al. Clinical staging and prognostic factors in Folliculotropic Mycosis Fungoides. JAMA Dermatol. 2016;152:992–1000.
37. Hodak E, Amitay-Laish I, Atzmony L, et al. New insights into folliculotropic mycosis fungoides (FMF): a single-center experience. J Am Acad Dermatol. 2016;75:347–55.
38. Gerami P, Guitart J. The spectrum of histopathologic and immunohistochemical findings in folliculotropic mycosis fungoides. Am J Surg Pathol. 2007;31:1430–8.
39. Vergier B, Beylot-Barry M, Beylot C, et al. Pilotropic cutaneous T-cell lymphoma without mucinosis. A variant of mycosis fungoides? French Study Group of Cutaneous Lymphomas. Arch Dermatol. 1996;132:683–7.
40. LeBoit PE, Abel EA, Cleary ML, et al. Clonal rearrangement of the T cell receptor beta gene in the circulating lymphocytes of erythrodermic follicular mucinosis. Blood. 1988;71:1329–33.
41. Boer A, Guo Y, Ackerman AB. Alopecia mucinosa is mycosis fungoides. Am J Dermatopathol. 2004;26:33–52.
42. Cerroni L, Fink-Puches R, Back B, Kerl H. Follicular mucinosis: a critical reappraisal of clinicopathologic features and association with mycosis fungoides and Sezary syndrome. Arch Dermatol. 2002;138:182–9.
43. Alikhan A, Griffin J, Nguyen N, Davis DM, Gibson LE. Pediatric follicular mucinosis: presentation, histopathology, molecular genetics, treatment, and outcomes over an 11-year period at the Mayo Clinic. Pediatr Dermatol. 2013;30:192–8.
44. Zvulunov A, Shkalim V, Ben-Amitai D, Feinmesser M. Clinical and histopathologic spectrum of alopecia mucinosa/follicular mucinosis and its natural history in children. J Am Acad Dermatol. 2012;67:1174–81.
45. Haghighi B, Smoller BR, LeBoit PE, Warnke RA, Sander CA, Kohler S. Pagetoid reticulosis (Woringer-Kolopp disease): an immunophenotypic, molecular, and clinicopathologic study. Mod Pathol. 2000;13:502–10.
46. Miedler JD, Kristjansson AK, Gould J, Tamburro J, Gilliam AC. Pagetoid reticulosis in a 5-year-old boy. J Am Acad Dermatol. 2008;58:679–81.
47. Matsuzaki Y, Kimura K, Nakano H, Hanada K, Sawamura D. Localized pagetoid reticulosis (Woringer-Kolopp disease) in early childhood. J Am Acad Dermatol. 2009;61:120–3.
48. Steffen C. Ketron-Goodman disease, Woringer-Kolopp disease, and pagetoid reticulosis. Am J Dermatopathol. 2005;27:68–85.
49. LeBoit PE. Granulomatous slack skin. Dermatol Clin. 1994;12:375–89.
50. van Haselen CW, Toonstra J, van der Putte SJ, van Dongen JJ, van Hees CL, van Vloten WA. Granulomatous slack skin. Report of three patients with an updated review of the literature. Dermatology. 1998;196:382–91.
51. Camacho FM, Burg G, Moreno JC, Campora RG, Villar JL. Granulomatous slack skin in childhood. Pediatr Dermatol. 1997;14:204–8.
52. Moreno-Gimenez JC, Jimenez-Puya R, Galan-Gutierrez M, Perez-Seoane C, Camacho FM. Granulomatous slack skin disease in a child: the outcome. Pediatr Dermatol. 2007;24:640–5.
53. Gokdemir G, Argon A, Sakiz D, Argon D, Koslu A. Granulomatous slack skin: report of a case with response to electron beam therapy. Med Oncol. 2008;25:178–81.
54. Sheng Y, Chen L, Huang Z, et al. Granulomatous slack skin: assessment of disease progression and treatment response using positron emission tomography/computed tomography. Clin Lymphoma Myeloma. 2009;9:455–8.
55. Tronnier M. Foreign-body-associated granulomatous slack skin in folliculotropic mycosis fungoides of childhood. J Cutan Pathol. 2009;36:578–81.
56. Kempf W, Ostheeren-Michaelis S, Paulli M et al. Granulomatous mycosis fungoides and granulomatous slack skin: a multicenter study of thr Cutaneous Lymphoma Histopathology Task Force Group of the European Organization for Research and Treatment of Cancer (EORTC). Arch Dermatol. 2008;144:1609–17.
57. Bekkenk MW, Geelen FA, van Voorst Vader PC, et al. Primary and secondary cutaneous CD30(+) lymphoproliferative disorders: a report from the Dutch Cutaneous Lymphoma Group on the long-term follow-up data of 219 patients and guidelines for diagnosis and treatment. Blood. 2000;95:3653–61.
58. de Souza A, Camilleri MJ, Wada DA, Appert DL, Gibson LE, el-Azhary RA. Clinical, histopathologic, and immunophenotypic features of lymphomatoid papulosis with CD8 predominance in 14 pediatric patients. J Am Acad Dermatol. 2009;61:993–1000.
59. Karai LJ, Kadin ME, Hsi ED, et al. Chromosomal rearrangements of 6p25.3 define a new subtype of lymphomatoid papulosis. Am J Surg Pathol. 2013;37:1173–81.
60. Kempf W. A new era for cutaneous CD30-positive T-cell lymphoproliferative disorders. Semin Diagn Pathol. 2017;34:22–35.
61. Wada DA, Law ME, Hsi ED, et al. Specificity of IRF4 translocations for primary cutaneous anaplastic large cell lymphoma: a multicenter study of 204 skin biopsies. Mod Pathol. 2011;24:596–605.
62. Nijsten T, Curiel-Lewandrowski C, Kadin ME. Lymphomatoid papulosis in children: a retrospective cohort study of 35 cases. Arch Dermatol. 2004;140:306–12.

63. Miquel J, Fraitag S, Hamel-Teillac D, et al. Lymphomatoid papulosis in children: a series of 25 cases. Br J Dermatol. 2014;171:1138–46.
64. de Souza A, el-Azhary RA, Camilleri MJ, Wada DA, Appert DL, Gibson LE. In search of prognostic indicators for lymphomatoid papulosis: a retrospective study of 123 patients. J Am Acad Dermatol. 2012;66:928–37.
65. Cordel N, Tressieres B, D'Incan M, et al. Frequency and risk factors for associated lymphomas in patients with lymphomatoid papulosis. Oncologist. 2016;21:76–83.
66. Kempf W, Pfaltz K, Vermeer MH, et al. EORTC, ISCL, and USCLC consensus recommendations for the treatment of primary cutaneous CD30-positive lymphoproliferative disorders: lymphomatoid papulosis and primary cutaneous anaplastic large-cell lymphoma. Blood. 2011;118:4024–35.
67. Bruijn MS, Horvath B, van Voorst Vader PC, Willemze R, Vermeer MH. Recommendations for treatment of lymphomatoid papulosis with methotrexate: a report from the Dutch Cutaneous Lymphoma Group. Br J Dermatol. 2015;173:1319–22.
68. Melchers RC, Willemze R, Bekkenk MW et al. Evaluation of treatment results in multifocal primary cutaneous anaplastic large cell lymphoma: report of the Dutch Cutaneous Lymphoma Group. Br J Dermatol. 2018;179:724–31.
69. Liu HL, Hoppe RT, Kohler S, Harvell JD, Reddy S, Kim YH. CD30+ cutaneous lymphoproliferative disorders: the Stanford experience in lymphomatoid papulosis and primary cutaneous anaplastic large cell lymphoma. J Am Acad Dermatol. 2003;49:1049–58.
70. Tomaszewski MM, Moad JC, Lupton GP. Primary cutaneous Ki-1(CD30) positive anaplastic large cell lymphoma in childhood. J Am Acad Dermatol. 1999;40:857–61.
71. Hazneci E, Aydin NE, Dogan G, Turhan IO. Primary cutaneous anaplastic large cell lymphoma in a young girl. J Eur Acad Dermatol Venereol. 2001;15:366–7.
72. Kumar S, Pittaluga S, Raffeld M, Guerrera M, Seibel NL, Jaffe ES. Primary cutaneous CD30-positive anaplastic large cell lymphoma in childhood: report of 4 cases and review of the literature. Pediatr Dev Pathol. 2005;8:52–60.
73. DeCoteau JF, Butmarc JR, Kinney MC, Kadin ME. The t(2;5) chromosomal translocation is not a common feature of primary cutaneous CD30+ lymphoproliferative disorders: comparison with anaplastic large-cell lymphoma of nodal origin. Blood. 1996;87:3437–41.
74. Kadin ME, Pinkus JL, Pinkus GS, et al. Primary cutaneous ALCL with phosphorylated/activated cytoplasmic ALK and novel phenotype: EMA/MUC1+, cutaneous lymphocyte antigen negative. Am J Surg Pathol. 2008;32:1421–6.
75. Oschlies I, Lisfeld J, Lamant L, et al. ALK-positive anaplastic large cell lymphoma limited to the skin: clinical, histopathological and molecular analysis of 6 pediatric cases. A report from the ALCL99 study. Haematologica. 2013;98:50–6.
76. Quintanilla-Martinez L, Jansen PM, Kinney MC, Swerdlow SH, Willemze R. Non-mycosis fungoides cutaneous T-cell lymphomas: report of the 2011 Society for Hematopathology/European Association for Haematopathology workshop. Am J Clin Pathol. 2013;139:491–514.
77. Attygalle AD, Cabecadas J, Gaulard P, et al. Peripheral T-cell and NK-cell lymphomas and their mimics; taking a step forward - report on the lymphoma workshop of the XVIth meeting of the European Association for Haematopathology and the Society for Hematopathology. Histopathology. 2014;64:171–99.
78. Jaffe ES, Harris NL, Stein H, Vardiman JW. World Health Organization classification of tumours: pathology and genetics of tumours of hematopoietic and lymphoid tissues. Lyon: IARC Press; 2001.
79. Salhany KE, Macon WR, Choi JK, et al. Subcutaneous panniculitis-like T-cell lymphoma: clinicopathologic, immunophenotypic, and genotypic analysis of alpha/beta and gamma/delta subtypes. Am J Surg Pathol. 1998;22:881–93.
80. Massone C, Chott A, Metze D, et al. Subcutaneous, blastic natural killer (NK), NK/T-cell, and other cytotoxic lymphomas of the skin: a morphologic, immunophenotypic, and molecular study of 50 patients. Am J Surg Pathol. 2004;28:719–35.
81. Willemze R, Jansen PM, Cerroni L, et al. Subcutaneous panniculitis-like T-cell lymphoma: definition, classification, and prognostic factors: an EORTC Cutaneous Lymphoma Group Study of 83 cases. Blood. 2008;111:838–45.
82. Huppmann AR, Xi L, Raffeld M, Pittaluga S, Jaffe ES. Subcutaneous panniculitis-like T-cell lymphoma in the pediatric age group: a lymphoma of low malignant potential. Pediatr Blood Cancer. 2013;60:1165–70.
83. Pincus LB, LeBoit PE, McCalmont TH, et al. Subcutaneous panniculitis-like T-cell lymphoma with overlapping clinicopathologic features of lupus erythematosus: coexistence of 2 entities? Am J Dermatopathol. 2009;31:520–6.
84. Oschlies I, Simonitsch-Klupp I, Maldyk J, et al. Subcutaneous panniculitis-like T-cell lymphoma in children: a detailed clinicopathological description of 11 multifocal cases with a high frequency of haemophagocytic syndrome. Br J Dermatol. 2015;172:793–7.
85. Mizutani S, Kuroda J, Shimura Y, et al. Cyclosporine A for chemotherapy-resistant subcutaneous panniculitis-like T cell lymphoma with hemophagocytic syndrome. Acta Haematol. 2011;126:8–12.
86. Mehta N, Wayne AS, Kim YH, et al. Bexarotene is active against subcutaneous panniculitis-like T-cell lymphoma in adult and pediatric populations. Clin Lymphoma Myeloma Leuk. 2012;12:20–5.
87. Kawachi Y, Furuta J, Fujisawa Y, Nakamura Y, Ishii Y, Otsuka F. Indolent subcutaneous panniculitis-like T cell lymphoma in a 1-year-old child. Pediatr Dermatol. 2012;29:374–7.
88. Bittencourt AL, Vieira M, Carvalho EG, Cunha C, Araujo I. Subcutaneous Panniculitis-Like T-Cell Lymphoma (SPTL) in a Child with Spontaneous Resolution. Case Rep Oncol Med. 2011;2011:639240.
89. Johnston EE, LeBlanc RE, Kim J, et al. Subcutaneous panniculitis-like T-cell lymphoma: Pediatric case series demonstrating heterogeneous presentation and option for watchful waiting. Pediatr Blood Cancer. 2015;62:2025–8.
90. Tokura Y, Ishihara S, Tagawa S, Seo N, Ohshima K, Takigawa M. Hypersensitivity to mosquito bites as the primary clinical manifestation of a juvenile type of Epstein-Barr virus-associated natural killer cell leukemia/lymphoma. J Am Acad Dermatol. 2001;45:569–78.
91. Rodriguez-Pinilla SM, Barrionuevo C, Garcia J, et al. EBV-associated cutaneous NK/T-cell lymphoma: review of a series of 14 cases from peru in children and young adults. Am J Surg Pathol. 2010;34:1773–82.
92. Iwatsuki K, Xu Z, Takata M, et al. The association of latent Epstein-Barr virus infection with hydroa vacciniforme. Br J Dermatol. 1999;140:715–21.
93. Sangueza M, Plaza JA. Hydroa vacciniforme-like cutaneous T-cell lymphoma: clinicopathologic and immunohistochemical study of 12 cases. J Am Acad Dermatol. 2013;69:112–9.
94. Barrionuevo C, Anderson VM, Zevallos-Giampietri E, et al. Hydroa-like cutaneous T-cell lymphoma: a clinicopathologic and molecular genetic study of 16 pediatric cases from Peru. Appl Immunohistochem Mol Morphol. 2002;10:7–14.
95. Kawa K, Okamura T, Yagi K, Takeuchi M, Nakayama M, Inoue M. Mosquito allergy and Epstein-Barr virus-associated T/natural killer-cell lymphoproliferative disease. Blood. 2001;98:3173–4.
96. Kimura H, Ito Y, Kawabe S, et al. EBV-associated T/NK-cell lymphoproliferative diseases in nonimmunocompromised hosts: prospective analysis of 108 cases. Blood. 2012;119:673–86.

Part VII

Treatment

Principles of Immunotherapy

24

Stanton Goldman and Jessica Hochberg

Introduction

Aggressive cytotoxic therapy cures most children and adolescents with NHL of their primary disease with expected EFS of ~75%, 85%, and 90% in anaplastic large cell lymphoma (ALCL), lymphoblastic lymphoma, and mature B-NHL. With such impressive primary cure rates, it becomes a tricky proposition to incorporate noncytotoxic therapies in these diseases. Nevertheless, recent advances are emerging in the use of immunotherapy specifically in pediatric NHL in both the minority of children who have refractory/recurrent disease and recently incorporation in high-risk populations in conjunction with cytotoxic therapies. The goal of immunotherapy may be to reduce the long-term burden of exposures to cytotoxic therapy in the pediatric and adolescent patient with NHL.

Immunotherapy as it applies to therapy for cancer is a rather broad term with the basic theme of relying on either delivering greater specificity to a cytotoxic agent (conjugated antibodies) and/or enhancing the patient's own (presumably deficient) immune response to malignancy. The advances in manufacturing highly specific monoclonal antibodies have allowed for targeting of tumor antigens with directing the immune system toward antibody-dependent cellular cytotoxicity. A more recent advance in adult malignancies has been the realization that many tumors and/or tumor microenvironments actively repress T-cells through the PD-1/PD-L1 inhibitory axis. Remarkable responses have been seen with antibodies directed at inhibiting PD-1 or PD-L1 repression allowing for reactivation of a brisk T-cell response. While this has been primarily seen in adult solid tumors, recent work and approval in refractory Hodgkin lymphoma, including children, indicate that these immune manipulating antibodies may play a role in NHL in the future. In a similar manner, bispecific antibodies "BitE" that bring T-cells and malignant cells into proximity through dual antigen binding can overcome T-cell anergy. Finally, the most exciting advance in immunotherapy in the past several years has been the pioneering and now FDA approval of manufacturing patient's own T-cells, ex vivo, with introduction of chimeric T-cell antigen receptors that specifically engage the manipulated T-cells to target specific malignant antigens. Chimeric antigen receptor T-cells are currently FDA approved in pediatric (AYA) precursor B-ALL and more recently in adult DLBCL. Table 24.1 attempts to categorize the ever-increasing expanse of immunotherapy into broad categories with "first in class" examples of the categories. Rather than overwhelm the reader with broad lists of targets/agents, we would like to specifically emphasize those immunotherapies that we feel have later-stage clinical data in pediatric NHL and may be FDA approved in pediatric NHL in the next 5–10 years.

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Table 24.1 Immunotherapies with Definite/Potential impact in Pediatric NHL

Category	First in class example	Mechanism of action	Approval in adult NHL	Approval in pediatric NHL	Comments
Naked antibodies	Rituximab	Anti-CD20; induces ADCC	Yes; follicular and DLBCL	No	Published pediatric data in recurrent and primary therapy of Burkitt, DLBCL and PTLD *(see text)*
Conjugated antibodies	Brentuximab Vedotin	Anti-CD30 conjugated with MMAE	Yes: Recurrent ALCL	No (over 12 years in ALCL refractory disease trial)	Ongoing study in upfront pediatric ALCL *(see text)*
Antibodies PD-1/PD-L1 axis	Pembrolizumab	PD-1 inhibition	No	No	Ongoing study in pediatric Hodgkin
Bispecific antibody	Blinatumomab	Bispecific monoclonal antibody CD3 and CD19	No (but approved in refractory and MRD + B- ALL)	No (but approved in refractory and MRD + B- ALL)	Potential for B-lymphoblastic lymphoma and possibly mature B-NHL
CAR T	Tisagenlecleucel	Chimeric antigen receptor T-cell against CD19	Yes; refractory CD19 + DLBCL	No (but primary approval in pediatric CD19 precursor B-ALL)	Potential for B-lymphoblastic lymphoma and possibly mature B-NHL *(see text)*

Antibody Therapies

Antibodies targeted to tumor cell surface antigen kill tumor cells through complement- and antibody-dependent cytotoxicity and induction of apoptosis. Monoclonal antibodies can sensitize cells to induction of apoptosis, thus accounting for their success in combination with cytotoxic therapy. Antibodies can also be utilized as a mechanism for delivery of toxins directly to the malignant cell. Ideal targets for monoclonal antibodies are those present on malignant cells only in order to minimize unwanted side effects.

Monoclonal Antibody Targeting CD20

Rituximab

Treatment of B-cell non-Hodgkin lymphoma (B-NHL) has been significantly enhanced by the addition of monoclonal antibody therapy in recent decades. Rituximab, a chimeric monoclonal antibody targeting the B-cell-associated antigen CD20, has exhibited significant activity in adults with B-NHL. As a single agent, rituximab led to responses in nearly half of adults with follicular lymphoma (FL) and when added to the chemotherapy regimen incorporating cyclophosphamide, doxorubicin, vincristine, and prednisone (CHOP) led to an improvement in response rates and survival compared to CHOP alone in adults with both indolent and aggressive forms of B-NHL [1–4]. While data is more limited and results mixed in the setting of adult Burkitt lymphoma (BL), recent data suggests that there is in fact a survival benefit to adding rituximab to chemotherapy for BL [5, 6]. With these findings, rituximab is currently standard of care for treatment of B-NHL in adults.

While all evidence points to a survival advantage with the addition of rituximab to chemotherapy in adult B-NHL, the question is still unsettled in pediatric B-NHL, though the data on rituximab use in pediatric B-NHL continues to grow. Rituximab was first investigated in pediatric B-NHL in the setting of relapsed/refractory disease in combination with ifosfamide, carboplatin, and etoposide (R-ICE). In the Children's Oncology Group (COG) study, R-ICE led to an overall response rate (ORR) of 60% in 20 patients with relapsed/refractory diffuse large B-cell lymphoma (DLBCL), BL, or mature B-cell acute lymphocytic leukemia (B-ALL) [7]. Of note, 8 of the 12 responders were survivors 13–30 months from study entry with no survivors in the non-responders and a median survival of only 2.5 months, highlighting the significant chemotherapy-resistant state of relapsed tumors in childhood B-NHL and the need to develop novel approaches to treating relapsed/refractory disease.

In the upfront setting, rituximab has been formally studied in several trials. The first study of rituximab in de novo pediatric B-NHL was undertaken by the German/Austrian/Swiss Berlin-Frankfurt-Munster (BFM) group. In a window study of a single dose of rituximab with only a 5-day response assessment period, 45% of children with B-NHL responded to single-agent rituximab [8]. While the study did not meet the preset target for response rate, the goal was aggressive considering the short assessment period, and overall the study demonstrated activity of rituximab in the setting of de novo pediatric B-NHL. The COG undertook a pilot study to investigate the addition of rituximab to FAB/LMB 96 backbone chemotherapy in FAB Group B and C patients. Based on evidence suggesting improved responses in adult patients that achieved higher-peak plasma rituximab levels, the study utilized a dose-dense approach with the addition of two

rituximab doses prior to each of two induction cycles and one rituximab dose prior to each of two consolidation cycles with Group C patients and then receiving four additional maintenance cycles [9, 10]. There was no significant difference in toxicity noted with rituximab administration in combination with chemotherapy suggesting rituximab can be safely given to children receiving intensive B-NHL chemotherapy [11, 12]. Additionally, survival outcomes compared favorably to historical values with 45 Group B and 40 Group C patients exhibiting a 3-year event-free survival (EFS) of 95% and 90%, respectively [11, 12]. A pharmacokinetic analysis demonstrated that rituximab exhibited similar pharmacokinetics to those reported in adults, though with a trend toward higher peak levels and a higher rate of clearance in younger children [13].

While these three studies support the safety and potential efficacy of rituximab in treating pediatric B-NHL, there had still been no currently published definitive evidence of the superiority of rituximab-containing regimens for B-NHL. Thus, a large international cooperative group Phase III study was initiated to investigate the addition of rituximab to FAB/LMB-96 backbone chemotherapy in a randomized fashion with planned accrual of 600 patients with higher risk disease (high lactate dehydrogenase [LDH]) Group B and Group C patients (NCT01595048). Accrual was halted after the first interim analysis after only 310 patients were enrolled. The preliminary analysis demonstrated a 1-year EFS of 94% in the rituximab arm vs. 81.5% in the control arm, one-sided p-value of 0.006 [14]. Though follow-up is ongoing, this finding suggests a likely benefit of rituximab at least in the high-risk setting.

Another area of investigation includes the possibility of therapeutic de-intensification with the addition of rituximab. Multiagent chemotherapy regimens utilized for pediatric B-NHL are associated with a high rate of acute toxicity, in particular mucositis and infectious complications/febrile neutropenia, and include anthracyclines, though generally at fairly low cumulative dose levels [15, 16]. While de-escalation of chemotherapy in a very curable disease remains a controversial issue, one potential application of rituximab would be to decrease cytotoxic chemotherapy associated with acute and/or late toxicities by the incorporation of rituximab into chemotherapy regimens for lower risk patients. The ongoing Reduced Burden of Oncologic Therapy (REBOOT) trial sponsored by the Childhood, Adolescent and Young Adult NHL Translational Research and Treatment (CAN TREAT) consortium is investigating a 60% reduction in the doxorubicin dose in Group B childhood, adolescent, and young adult patients with the addition of rituximab to FAB/LMB-96-based chemotherapy while also investigating the addition of rituximab to standard Group C therapy with both groups also receiving less total intrathecal chemotherapy doses with the addition of intrathecal liposomal cytarabine (NCT01859819). While accrual is ongoing, in an initial report on 24 patients accrued (18 Group B and 6 Group C), the failure-free survival and overall survival (OS) are 100% with a median time from study entry of 52 weeks (6–152 weeks) [17]. Table 24.2 summarizes the published trials of rituximab in de novo pediatric mature B-NHL.

PTLD most often arises in pediatric patients with solid organ transplant in the setting of immunosuppressive therapy. Majority of PTLD is either polyclonal or monoclonal EBV positive and CD20 positive. While the reduction in immunosuppressive agent can lead to resolution of disease, this is balanced with risk of organ rejection. The COG incorporated rituximab (days 1, 8, and 15) in the first two cycles of a low-dose chemotherapy regimen consisting of cyclophosphamide and prednisone. Fifty-five patients were enrolled including 4 with fulminant presentations. The 2-year EFS (including allograft preservation) of 71% (95% CI: 57–82%) was considered reasonably safe and effective [18]. The current ongoing COG study attempts to eliminate cytotoxic therapy all together with initial treatment with rituximab alone followed by infusion of third party-latent membrane protein-specific cytotoxic T-cells in patients with less than CR to rituximab (NCT02900976).

Obinutuzumab

Obinutuzumab is a humanized type II anti-CD20 monoclonal antibody with a glycoengineered Fc portion enhancing its affinity for Fc receptors including in the setting of Fc receptor polymorphisms known to inhibit rituximab binding [19]. In preclinical studies, obinutuzumab shows enhanced antibody-dependent cellular cytotoxicity compared to rituximab. Clinically, obinutuzumab has exhibited promising activity in both indolent and aggressive adult B-NHLs with significantly higher overall response rates compared to rituximab in relapsed/refractory indolent B-NHL patients [20], mantle cell lymphoma, or DLBCL [21]. This included responses even in patients deemed rituximab refractory. This is also seen in preclinical studies where obinutuzumab had higher activity than rituximab in rituximab-sensitive and rituximab-resistant Burkitt lymphoma cell line [22]. Based on these preclinical results, obinutuzumab use in relapsed/refractory childhood B-NHL is being investigated in a trial sponsored by the CAN TREAT consortium combining obinutuzumab with ICE chemotherapy (NCT02393157).

Ibritumomab Tiuxetan

Ibritumomab tiuxetan (Zevalin) is a monoclonal antibody directed toward CD20 attached to a radioactive molecule, Yttrium-90 (^{90}Y). This allows the antibody to deliver radiation directly to the lymphoma cells, limiting radiation toxicities to surrounding tissues. The use of radiolabeled antibodies for treatment of lymphomas is supported by the radiosensitivity of lymphomas making them ideal targets. In addition,

Table 24.2 Rituximab in pediatric mature B-NHL

Study design	Disease state and number patients	Chemotherapy backbone	Rituximab doses	Outcomes	Reference(s)
Phase II	Recurrent disease/ $n = 20$	ICE	Day 1,3 each cycle	CR/PR 60%	Griffin et al. [7]
Phase II window	Newly diagnosed/ $n = 87$ evaluable	BFM 2004 (after window)	One dose prior to chemo	ORR 41.4% to window therapy	Meinhardt et al. [8]
Pilot	Newly diagnosed/ $n = 85$	FAB 96	Day 1,3 each induction and day 1 each consolidation	EFS 92% at 3 years/safe feasible to combine with multiagent chemotherapy	Goldman et al. [11, 12]
Phase III randomized	Newly diagnosed/ $n = 310$ (155/ arm)	FAB 96	Day 1,3 each induction and day 1 each consolidation (randomized)	1-year EFS 94.2 (chemo + rituximab) vs. 81.5% (chemo alone); trial halted at first interim analysis for apparent benefit rituximab	Minard-Colin et al. [14] (abstract)

ICE ifosfamide, carboplatin, etoposide, *BFM 2004* Berlin-Frankfurt-Munster chemotherapy, *Modified FAB 96* superior arms of French–American–British (FAB) 96 with a reduced infusion time of Adriamycin, *CR* complete response, *PR* partial response, *EFS* event-free survival

the direct killing of tumor cells does not rely on the hosts' immune system for efficacy. Furthermore, the radioactive particles emitted are cytotoxic across many cell diameters leading to enhanced killing even of antigen-negative tumor cells and the ability to overcome tumor penetration barriers [23]. While the use of anti-CD20 radioimmunoconjugates has been approved for use in adults with follicular lymphoma, limited data exists on their use in children. In children, ^{90}Y-IT was studied in a Phase I COG trial in relapsed/refractory B-NHL. While no dose-limiting toxicity or excessive radiation exposure was identified, there were also no responses noted in the five heavily pretreated study patients [24]. Currently, 90Y-IT is indicated for consolidation in the front-line and may be effective as a part of myeloablative transplant regimens for aggressive B-NHLs [25].

Monoclonal Antibody Targeting CD30

Brentuximab Vedotin (Bv)

Most ALCLs have been shown to be of the T-cell phenotype and are associated with a characteristic genetic alteration involving the ALK locus on chromosome 2 and expression of CD30. Accumulating evidence indicates that the immune system plays a major role in both the pathogenesis and final control of anaplastic lymphoma kinase (ALK)-positive ALCL [26–28]. In recent trials with very diverse first-line chemotherapy regimens in terms of the duration of treatment as well as the number and cumulative doses of drugs, there are reported similar EFS rates of about 65–75% in children, adolescents, and young adults [29–32]. No intervention has been able to improve on the approximate failure rate of 25–30% that exists regardless of treatment strategy. The role of the immune system in the control of ALCL makes monoclonal antibody therapy particularly attractive.

Brentuximab vedotin combines the antitubulin cytotoxic agent monomethyl auristatin E (MMAE) attached by an enzymatically cleavable linker to the CD30-specific monoclonal antibody. After binding to CD30 antigen on target tumor cell, the antibody–drug conjugate is rapidly internalized and undergoes intralysosomal cleavage allowing the MMAE component to bind tubulin and lead to cell cycle arrest and eventual apoptosis (Fig. 24.1). Consistent with other tubulin inhibitors (like vincristine), a frequent toxicity appears to be peripheral neuropathy.

There have been two pivotal Phase I studies of Bv involving patients with relapsed ALCL. Younes published the first Phase I study using Bv given every 3 weeks to patients with refractory or relapsed CD30-positive lymphomas, including ALCL (NCT00430846) [33]. Of the 45 patients enrolled, 33 (73%) had previously received an SCT. Despite this high amount of pretreatment, toxicity was tolerable and the vast majority of toxicities were Grade 1 or 2. In the second Phase I dose escalation study, Fanale et al. gave Bv on days 1, 8, and 15, of each 28-day cycle at doses ranging from 0.4 to 1.4 mg/kg [34]. Forty-four patients were enrolled including five with systemic ALCL and one with peripheral T-cell lymphoma not otherwise specified. The maximum tolerated dose (MTD) was found to be 1.2 mg/kg. Tumor regression occurred in 85% of patients, and the overall objective response rate was 59% ($n = 24$), with 34% ($n = 14$) CRs. The median duration of response was not reached at a median follow-up of 45 weeks on study [34]. In these early Phase I trials, six of seven patients with ALCL had a CR and one had stable disease [33, 34].

A Phase II multicenter trial using Bv 1.8 mg/kg every 3 weeks for patients over 12 years of age with relapsed or refractory ALCL has recently been completed (NCT00866047). Bv was administered over 30 minutes as an outpatient every 3 weeks. A total of 58 patients were enrolled with an ORR of 86% (53% CR, 33% partial response [PR]), and 97% of patients demonstrated tumor reduction. Adverse events were manageable with a toxicity profile like the Phase I studies [35]. This has led to the accelerated FDA approval

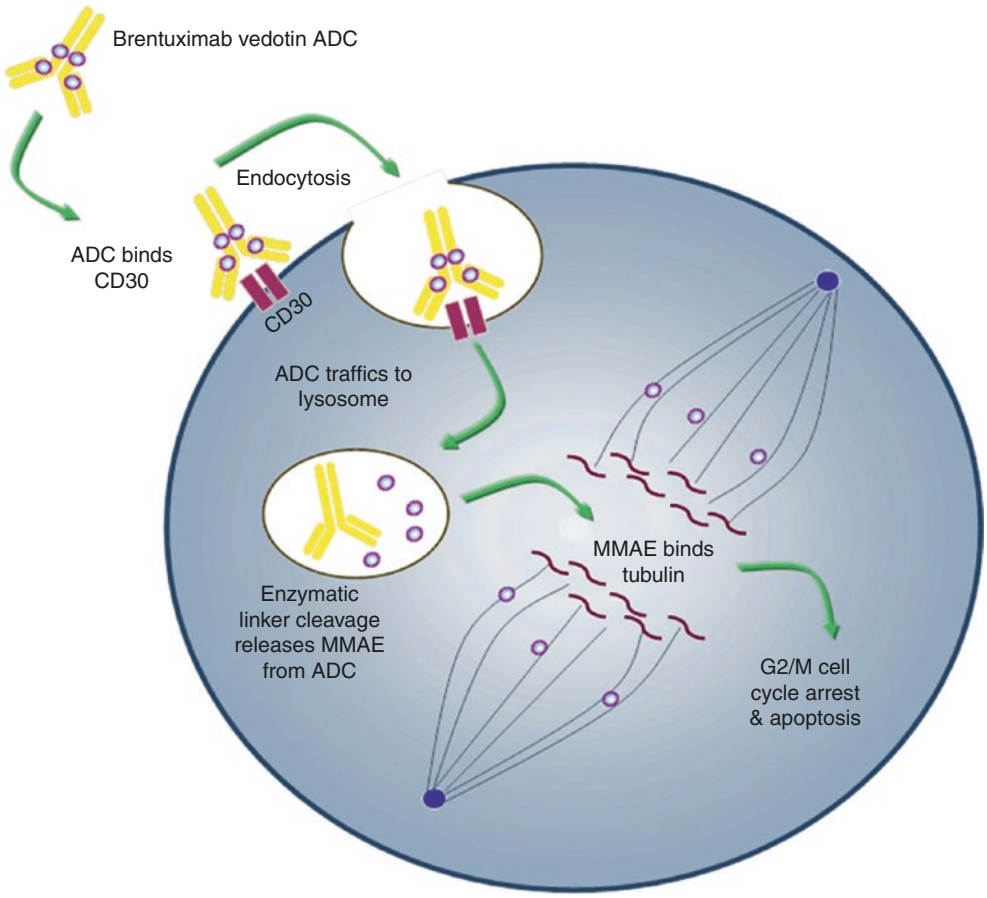

Fig. 24.1 Brentuximab vedotin mechanism of action. Brentuximab vedotin (Bv) is an anti-CD30 antibody–drug conjugate, paired with the microtubule stabilizer monomethyl auristatin E (MMAE)

of Bv in systemic ALCL after failure of multiagent chemotherapy. A recent update of the Phase II study at a median observation period of 6 years observed no progressions noted after 40 months. About half of patients who achieved CR to brentuximab proceeded to autologous stem cell transplant. Reassuringly, peripheral neuropathy resolved or improved in most patients. The long-term data provide evidence that single-agent brentuximab may be a potentially curative treatment option [36].

In adult CD30-positive NHL including ALCL, combination chemotherapy with Bv and standard-dose CHOP chemotherapy or cyclophosphamide, doxorubicin and prednisone without vincristine (CHP) has been trialed (NCT01309789). Patients received sequential treatment with Bv 1.8 mg/kg (two cycles) followed by CHOP (six cycles) or Bv 1.8 mg/kg plus CHP (BV + CHP) for six cycles. Responders then received single-agent Bv for 8 to 10 additional cycles (total of 16 cycles). The MTD of Bv in combination with CHP chemotherapy was 1.8 mg/kg administered every 3 weeks. All treated patients (100%) achieved an objective response, with 23 (88%) of 26 evaluable patients achieving a CR [37]. Based on these promising results, a randomized trial of Bv with CHP chemotherapy compared with CHOP chemotherapy in the first-line management of patients with CD30-positive T-cell NHL is currently in progress (NCT01777152). Although not yet approved in upfront therapy for ALCL, Bv has recently shown superior EFS and gained FDA approval for stage III/IV classical adult Hodgkin lymphoma when combined with AVD chemotherapy for 4–6 cycles.

The role of Bv added to front-line treatment of pediatric/adolescent disseminated ALCL is also currently being evaluated in a prospective trial by the COG. The COG regimen is piloting a more aggressive backbone based on the international European therapy (ALCL99) which includes high-dose methotrexate and more aggressive alkylators plus etoposide exposure than standard adult CHOP regimen (NCT01979536). Although not applicable to ALCL chemotherapy backbones, it should be noted that there is an absolute contraindication to combine Bv with bleomycin due to excessive pulmonary toxicity.

Monoclonal Antibody Targeting CD19

CD19 is expressed on nearly all B-cell malignancies, making it an attractive target for therapy. Coltuximab ravtansine (SAR3419) is a humanized anti-CD19 antibody conjugated to maytansinoid, a potent inhibitor of tubulin polymerization [38]. In a Phase I study in 44 adults with relapsed/

refractory B-NHL, SAR3419 led to an ORR of 30% [39]. Subsequent findings in Phase II testing in adults with relapsed/refractory DLBCL showed an ORR of 44% in 41 patients with acceptable toxicity. Response rates were significantly higher in patients with relapsed vs. primary refractory disease (58% vs. 12%) in this high-risk population [40, 41]. Denintuzumab mafodotin (SGN-CD19A) has also been tested in relapsed/refractory B-NHL. SGN-CD19A is conjugated with the microtubule stabilizing agent monomethyl auristatin F (MMAF). In two ongoing Phase I trials in relapsed/refractory B-NHL, 33% of 12 adult patients with BL/leukemia or B-lymphoblastic lymphoma (B-LBL) (NCT01786096) and 62 adult patients with indolent lymphomas (NCT01786135) responded [42, 43]. However, no responses have been observed in a small trial of pediatric patients with BL/leukemia reported to date [44].

Monoclonal Antibody Targeting CD22

CD22 is another B-cell associated antigen frequently expressed on B-NHL cells. Epratuzumab appears to function differently from rituximab and may be synergistic based on preclinical testing [45]. Single-agent treatment with unconjugated epratuzumab in Phase I/II trials led to ORRs of 43% in 14 follicular lymphoma patients, though only 15% in 33 DLBCL patients [46, 47]. However, when combined with rituximab or R-CHOP, ORRs improved to 88% and 96%, respectively [48, 49]. A conjugated form with ^{90}Y showed a promising ORR of 62% in 64 adults with relapsed/refractory B-NHL [50]. Though epratuzumab has been studied by the COG in relapsed/refractory childhood B-ALL, no data exists on its use in childhood B-NHL.

Pinatuzumab vedotin is an anti-CD22 antibody conjugated with the microtubule stabilizer monomethyl auristatin E (MMAE). Initial Phase I trials showed promising activity in 41% of DLBCL patients and 50% of indolent B-NHL patients [51]. The Phase II ROMULUS study is investigating pinatuzumab vedotin in combination with rituximab in a randomized fashion with rituximab combined with another MMAE conjugated ADC targeting CD79B, polatuzumab vedotin (see below), in relapsed/refractory B-NHL. The pinatuzumab vedotin–rituximab combination led to an ORR of 57% in DLBCL and 62% in follicular lymphoma [52]. However, toxicities include diarrhea, neutropenia, and peripheral neuropathy with a large number of patients discontinuing treatment due to peripheral neuropathy.

Another conjugated anti-CD22 antibody inotuzumab ozogamicin has also exhibited promise in B-ALL; however, Phase III trials in B-NHL were halted prematurely due to either poor enrollment (NCT00562965) or lack of efficacy (NCT01232556) [53, 54].

Monoclonal Antibody Targeting CD79B

The B-cell receptor is a heterodimer of CD79A and CD79B in combination with surface immunoglobulin. Polatuzumab vedotin is an anti-CD79B antibody conjugated to MMAE. Following an initial Phase I dose-finding study in B-NHL and CLL, a Phase II expansion cohort continued to enroll B-NHL patients for treatment with polatuzumab vedotin as a single agent or in combination with rituximab. In all patients treated with single agent at the recommended Phase II dose, responses were observed in 56% of DLBCL and 47% of follicular lymphoma patients [55]. When combined with rituximab, 78% of patients responded [52]. This represents a promising adjunct to CD20 antibody therapy and we may see incorporation into pediatric trials pending these early adult results.

Monoclonal Antibody Toxicities

Infusion reactions occur commonly with monoclonal antibody administration. Symptoms can include fever, bronchospasm, hypoxemia, and rigors. This is most often seen during the first infusion and is managed easily with supportive care. Patients are typically able to tolerate subsequent infusions with appropriate premedication and/or slower rates. Rituximab-related B-cell depletion occurs in most patients, lasting 6–12 months, although typically without significant associated infectious risk. However, there are reports of hepatitis B reactivation with anti-CD20 therapies, necessitating screening for HBV.

Bispecific T-cell Engager (BiTE)

A novel application of monoclonal antibody therapy is the use of multivalent antibodies targeting multiple cell surface proteins. A bispecific T-cell engaging (BiTE) antibody is a single polypeptide with two specific antigen-binding sites, one which engages a specific B-cell marker and another targeting a co-stimulatory molecule on T-cells. This allows for recruitment of T-cells specifically to malignant B-cells leading to T-cell activation and apoptosis [56]. By directly engaging T-cells, we may enhance the efficacy of antibody therapy. Blinatumomab targets both CD19 and CD3 inducing activation of CD3+ cytotoxic T-cells in the presence of CD19+ B-cells. Blinatumomab has been extensively investigated in B-ALL. In adult B-NHL, a Phase I trial investigated blinatumomab in 38 relapsed/refractory B-NHL patients leading to an ORR of 29% [57]. In Phase II testing in aggressive B-NHL, an ORR of 43% was observed in 21 relapsed/refractory adult DLBCL patients [58]. Common toxicities include neurotoxicity and cytokine release syndrome, which can be managed with supportive care and the use of the anti-IL6 monoclonal antibody tocilizumab [59, 60].

Checkpoint Inhibitors

One mechanism of tumor survival and proliferation is through the avoidance of host immune surveillance. We can enhance immune surveillance utilizing antibodies targeting the checkpoint inhibitors cytotoxic T-lymphocyte-associated antigen-4 (CTLA-4) and programmed cell death protein 1 (PD-1) and the PD-1 ligands PD-L1/PD-L2 [61]. The PD-1 ligand, PD-L1, is normally expressed on antigen-presenting cells, activated T-cells, and other immune cells, while PD-L2 expression is typically found on macrophages, dendritic cells, and B-cells. Immediately after activation of the T-cell receptor, engagement of PD-1 with PD-L1/L2 inhibits PI3K activity, blocking further T-cell activation and decreasing cytokine production. This mechanism normally serves to regulate the immune response but is exploited by many cancers [62]. PD-L1 and/or PD-L2 has been shown to be expressed in a subset of non-Hodgkin lymphomas as well as in the tumor microenvironment, making this pathway a promising target [63]. Several checkpoint inhibitor antibodies have recently gained approval for treatment of malignancies and continue to be investigated in relapsed/refractory B-NHL. Ipilimumab, an anti-CTLA-4 antibody, demonstrated tumor regression in relapsed/refractory follicular lymphoma and DLBCL and has been safely administered in combination with rituximab in early Phase I results [64, 65]. Pidilizumab, targeting PD-1, has demonstrated safety and activity in relapsed/refractory DLBCL, with responses noted in half of the 35 patients with measurable disease post-SCT, and in combination with rituximab in relapsed/refractory follicular lymphoma where the combination led to an ORR of 66% in 29 patients and a CR rate of 52% [66, 67]. Nivolumab and pembrolizumab, both anti-PD-1 antibodies, exhibited an ORR of 30–40% in heavily pretreated B-NHL and PMBL patients [68]. In children, the COG is investigating nivolumab as a single agent and in combination with ipilimumab in children with relapsed/refractory solid tumors including lymphoma (NCT02304458), and there are ongoing pembrolizumab trials being investigated in children with PD-L1-positive tumors including lymphomas (NCT02332668). The side effect profile of checkpoint inhibitors is low with the most common reactions being inflammatory or autoimmune including hepatitis, pneumonitis, colitis, thyroiditis, and hypophysitis. It is recommended for patients receiving checkpoint inhibitors to have regular monitoring of thyroid and other endocrine functioning with supportive care as needed for low-grade toxicities.

Adoptive Cellular Therapy

Chimeric Antigen Receptor T-cells

Chimeric antigen receptor (CAR) T-cells in the treatment of pediatric refractory precursor B-ALL have been an example of breakthrough paradigm changer in the order of BCR-ABL inhibition with imatinib in CML. CAR T-cells are T lymphocytes (typically autologous) genetically engineered to bind to specific antigens expressed on malignant cells. They are composed of an extracellular binding domain, hinge region, transmembrane domain, and one or more intracellular signaling domains. The process of production involves multiple steps beginning with peripheral blood cell collection from the patient followed by T-cell isolation and activation and T-cell modification (CAR introduction) followed by T-cell expansion and product formulation. The product is then reinfused into the patient after a lymphodepleting chemotherapy (often with fludarabine) [69]. Once bound to the malignant cell, the signaling domains stimulate T-cell proliferation, cytolysis, and cytokine secretion to eliminate the tumor cell. There have been multiple generations of CAR T-cells which incorporate additional more effective co-stimulatory signals. As with antibody therapy, ideally the antigen targeted by CAR T-cells is present predominantly on malignant cells.

CAR T-Cells Targeting CD19

Tisagenlecleucel (Kymriah), an anti-CD19 CAR T-cell therapy originally developed at the University of Pennsylvania and currently manufactured by Novartis, was tested in children and young adults (up to 25 years) with CD19 + relapsed or refractory B precursor ALL. The remission rate after a single infusion at 3 months was remarkably high at 80% with 90% overall survival at 6 months and 76% survival at 12 months [70].

CD19 is expressed at an early stage in B-cell maturation and is normally present just up to plasma cell differentiation. However, malignant mature B-NHL has more variable expression, especially Burkitt lymphoma. Nevertheless, another anti-CD19 CAR T-cell therapy, axicabtagene ciloleucel (Yescarta), gained FDA approval for diffuse large B-cell lymphoma, primary mediastinal large B-cell lymphoma, high-grade B-cell lymphoma, and diffuse large B-cell lymphoma arising from follicular lymphoma after at least two other kinds of treatment have been tried. Among 111 adult patients, the CR rate was 54% and the overall survival at 18 months in this highly pretreated adult cohort was 52% [71]. Subsequently, tisagenlecleucel also gained FDA approval in refractory/relapsed adult DLBCL and other high-grade B-cell lymphomas.

CAR T-Cells Targeting Other Antigens

Although the currently approved products all have CD19 specificity, there are numerous other tumor antigens in development with high relevance to pediatric NHL including CD20, CD22 (Burkitt/DLBCL), and CD30 (ALCL).

CAR T-Cell Therapy Toxicities

CAR T-cell products share a significant and unique toxicity profile including cytokine release syndrome and neurological toxicities. Other severe side effects include infection, low blood cell counts, and a weakened immune system. Cytokine release syndrome (CRS) is a potentially life-threatening toxicity of CAR T-cell infusion. CRS is associated with high levels of several cytokines, including interleukin-6 (IL-6) and interferon γ. The clinical syndrome of CRS includes fever, hypotension, and hypoxia [72]. Symptom onset is typically within days to weeks, correlating with peak in vivo T-cell expansion. IL6 inhibition with tocilizumab has been proven highly effective in the management of severe CRS associated with CAR T-cell therapies. A response is typically seen within hours of administration.

Neurologic toxicity can also occur, including delirium and encephalopathy [73]. Most of the neurologic symptoms are reversible with use of dexamethasone. Most significantly, severe B-cell aplasia has been seen in 100% of patients due to the depletion of non-malignant CD19 B lymphocytes. Due to the risk for opportunistic infections due to hypogammaglobulinemia, these patients require lifelong IVIG replacement [70].

Conclusion

Children and adolescents with cancer are usually otherwise healthy enough to tolerate the rigors of aggressive multiagent chemotherapy. In addition, advances in supportive care, including anti-infective agents, rasburicase for tumor lysis, and blood product support, have helped us deliver chemotherapy safely. Nevertheless, in many pediatric malignancies, including NHL, we seem to have hit the limit of cytotoxic chemotherapy escalation/manipulation in improving cure rates. In addition, we are too often burdening our survivors with lifelong late effects of chemotherapy. One envisions a future where immunotherapy helps to improve survival in the highest risk patients and/or reduce exposure to cytotoxic therapy for all.

References

1. Maloney DG, Grillo-Lopez AJ, White CA, et al. IDEC-C2B8 (Rituximab) anti-CD20 monoclonal antibody therapy in patients with relapsed low-grade non-Hodgkin's lymphoma. Blood. 1997;90:2188–95.
2. Czuczman MS, Grillo-Lopez AJ, White CA, et al. Treatment of patients with low-grade B-cell lymphoma with the combination of chimeric anti-CD20 monoclonal antibody and CHOP chemotherapy. J Clin Oncol. 1999;17:268–76.
3. Coiffier B, Lepage E, Briere J, et al. CHOP chemotherapy plus rituximab compared with CHOP alone in elderly patients with diffuse large-B-cell lymphoma. N Engl J Med. 2002;346:235–42.
4. Pfreundschuh M, Trumper L, Osterborg A, et al. CHOP-like chemotherapy plus rituximab versus CHOP-like chemotherapy alone in young patients with good-prognosis diffuse large-B-cell lymphoma: a randomised controlled trial by the MabThera International Trial (MInT) Group. Lancet Oncol. 2006;7:379–91.
5. Rizzieri DA, Johnson JL, Byrd JC, et al. Improved efficacy using rituximab and brief duration, high intensity chemotherapy with filgrastim support for Burkitt or aggressive lymphomas: cancer and Leukemia Group B study 10 002. Br J Haematol. 2014;165:102–11.
6. Hoelzer D, Walewski J, Döhner H, et al. Improved outcome of adult Burkitt lymphoma/leukemia with rituximab and chemotherapy: report of a large prospective multicenter trial. Blood. 2014;124:3870–9.
7. Griffin TC, Weitzman S, Weinstein H, et al. A study of rituximab and ifosfamide, carboplatin, and etoposide chemotherapy in children with recurrent/refractory B-cell (CD20+) non-Hodgkin lymphoma and mature B-cell acute lymphoblastic leukemia: a report from the Children's Oncology Group. Pediatr Blood Cancer. 2009;52:177–81.
8. Meinhardt A, Burkhardt B, Zimmermann M, et al. Phase II window study on rituximab in newly diagnosed pediatric mature B-cell non-Hodgkin's lymphoma and Burkitt leukemia. J Clin Oncol. 2010;28:3115–21.
9. Jäger U, Fridrik M, Zeitlinger M, et al. Rituximab serum concentrations during immuno-chemotherapy of follicular lymphoma correlate with patient gender, bone marrow infiltration and clinical response. Haematologica. 2012;97:1431–8.
10. Berinstein NL, Grillo-Lopez AJ, White CA, et al. Association of serum Rituximab (IDEC-C2B8) concentration and anti-tumor response in the treatment of recurrent low-grade or follicular non-Hodgkin's lymphoma. Ann Oncol. 1998;9:995–1001.
11. Goldman S, Smith L, Anderson JR, et al. Rituximab and FAB/LMB 96 chemotherapy in children with Stage III/IV B-cell non-Hodgkin lymphoma: a Children's Oncology Group report. Leukemia. 2013;27:1174–7.
12. Goldman S, Smith L, Galardy P, et al. Rituximab with chemotherapy in children and adolescents with central nervous system and/or bone marrow-positive Burkitt lymphoma/leukaemia: a Children's Oncology Group report. Br J Haematol. 2014;167:394.
13. Barth MJ, Goldman S, Smith L, et al. Rituximab pharmacokinetics in children and adolescents with de novo intermediate and advanced mature B-cell lymphoma/leukaemia: a Children's Oncology Group report. Br J Haematol. 2013;162:678–83.
14. Minard-Colin V, Auperin A, Pillon M, et al. Results of the randomized intergroup trial inter-B-NHL Ritux 2010 for children and adolescents with high risk B-cell non Hodgkin's lymphoma and mature acute leukemia: evaluation of efficacy in addition to standard LMB chemotherapy regimen. J Clin Oncol. 2016;34:abstr 10507.
15. Patte C, Auperin A, Gerrard M, et al. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. Blood. 2007;109:2773–80.
16. Cairo MS, Gerrard M, Sposto R, et al. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. Blood. 2007;109:2736–43.
17. Goldman S, Barth MJ, Oesterheld JE, et al. Preliminary results of a reduced burden of therapy trial by incorporation of rituximab and intrathecal liposomal cytarabine in children, adolescents and young adults with intermediate (FAB Group B) and high risk (FAB Group C) mature B-cell lymphoma/leukemia. Chicago: ASCO Annual Meeting; 2016 (abstract).

18. Gross TG, Orjuela MA, Perkins SL, et al. Low-dose chemotherapy and rituximab for posttransplant lymphoproliferative disease (PTLD): a Children's Oncology Group Report. Am J Transplant. 2012;12:3069–75.
19. Mossner E, Brunker P, Moser S, et al. Increasing the efficacy of CD20 antibody therapy through the engineering of a new type II anti-CD20 antibody with enhanced direct and immune effector cell-mediated B-cell cytotoxicity. Blood. 2010;115:4393–402.
20. Sehn LH, Goy A, Offner FC, et al. Randomized phase II trial comparing obinutuzumab (GA101) with rituximab in patients with relapsed CD20+ indolent B-cell non-Hodgkin lymphoma: final analysis of the GAUSS study. J Clin Oncol. 2015;33:3467–74.
21. Morschhauser FA, Cartron G, Thieblemont C, et al. Obinutuzumab (GA101) monotherapy in relapsed/refractory diffuse large b-cell lymphoma or mantle-cell lymphoma: results from the phase II GAUGUIN study. J Clin Oncol. 2013;31:2912–9.
22. Awasthi A, Ayello J, Van de Ven C, et al. Obinutuzumab (GA101) compared to rituximab significantly enhances cell death and antibody-dependent cytotoxicity and improves overall survival against CD20(+) rituximab-sensitive/-resistant Burkitt lymphoma (BL) and precursor B-acute lymphoblastic leukaemia (pre-B-ALL): potential targeted therapy in patients with poor risk CD20(+) BL and pre-B-ALL. Br J Haematol. 2015;171:763–75.
23. Press OW, Leonard JP, Coiffier B, et al. Immunotherapy of non-Hodgkin's lymphomas. Hematology Am Soc Hematol Educ Program. 2001;2001:221–40.
24. Cooney-Qualter E, Krailo M, Angiolillo A, et al. A phase I study of 90yttrium-ibritumomab-tiuxetan in children and adolescents with relapsed/refractory CD20-positive non-Hodgkin's lymphoma: a Children's Oncology Group study. Clin Cancer Res. 2007;13:5652s–60s.
25. Witzig TE, Gordon LI, Cabanillas F, et al. Randomized controlled trial of yttrium-90-labeled ibritumomab tiuxetan radioimmunotherapy versus rituximab immunotherapy for patients with relapsed or refractory low-grade, follicular, or transformed B-cell non-Hodgkin's lymphoma. J Clin Oncol. 2002;20:2453–63.
26. Stein H, Foss HD, Durkop H, et al. CD30(+) anaplastic large cell lymphoma: a review of its histopathologic, genetic, and clinical features. Blood. 2000;96:3681–95.
27. Chiarle R, Voena C, Ambrogio C, et al. The anaplastic lymphoma kinase in the pathogenesis of cancer. Nat Rev Cancer. 2008;8:11–23.
28. Lamant L, McCarthy K, d'Amore E, et al. Prognostic impact of morphologic and phenotypic features of childhood ALK-positive anaplastic large-cell lymphoma: results of the ALCL99 study. J Clin Oncol. 2011;29:4669–76.
29. Alexander S, Kraveka JM, Weitzman S, et al. Advanced stage anaplastic large cell lymphoma in children and adolescents: results of ANHL0131, a randomized phase III trial of APO versus a modified regimen with vinblastine: a report from the children's oncology group. Pediatr Blood Cancer. 2014;61:2236–42.
30. Brugieres L, Quartier P, Le Deley MC, et al. Relapses of childhood anaplastic large-cell lymphoma: treatment results in a series of 41 children--a report from the French Society of Pediatric Oncology. Ann Oncol. 2000;11:53–8.
31. Le Deley MC, Rosolen A, Williams DM, et al. Vinblastine in children and adolescents with high-risk anaplastic large-cell lymphoma: results of the randomized ALCL99-vinblastine trial. J Clin Oncol. 2010;28:3987–93.
32. Lowe EJ, Sposto R, Perkins SL, et al. Intensive chemotherapy for systemic anaplastic large cell lymphoma in children and adolescents: final results of Children's Cancer Group Study 5941. Pediatr Blood Cancer. 2009;52:335–9.
33. Younes A, Bartlett NL, Leonard JP, et al. Brentuximab vedotin (SGN-35) for relapsed CD30-positive lymphomas. N Engl J Med. 2010;363:1812–21.
34. Fanale MA, Forero-Torres A, Rosenblatt JD, et al. A phase I weekly dosing study of brentuximab vedotin in patients with relapsed/refractory CD30-positive hematologic malignancies. Clin Cancer Res. 2012;18:248–55.
35. Pro B, Advani R, Brice P, et al. Brentuximab vedotin (SGN-35) in patients with relapsed or refractory systemic anaplastic large-cell lymphoma: results of a phase II study. J Clin Oncol. 2012;30:2190–6.
36. Pro B, Advani R, Brice P, et al. Five-year results of brentuximab vedotin in patients with relapsed or refractory systemic anaplastic large cell lymphoma. Blood. 2017;130:2709–17.
37. Fanale MA, Horwitz SM, Forero-Torres A, et al. Brentuximab vedotin in the front-line treatment of patients with CD30+ peripheral T-cell lymphomas: results of a phase I study. J Clin Oncol. 2014;32:3137–43.
38. Blanc V, Bousseau A, Caron A, et al. SAR3419: an anti-CD19-Maytansinoid Immunoconjugate for the treatment of B-cell malignancies. Clin Cancer Res. 2011;17:6448–58.
39. Ribrag V, Dupuis J, Tilly H, et al. A dose-escalation study of SAR3419, an anti-CD19 antibody maytansinoid conjugate, administered by intravenous infusion once weekly in patients with relapsed/refractory B-cell non-Hodgkin lymphoma. Clin Cancer Res. 2014;20:213–20.
40. Trneny M, Verhoef G, Dyer MJ, et al. Starlyte phase II study of coltuximab ravtansine (CoR, SAR3419) single agent: clinical activity and safety in patients (pts) with relapsed/refractory (R/R) diffuse large B-cell lymphoma (DLBCL; NCT01472887). J Clin Oncol2014 ASCO Annual Meeting Abstracts. 2014;32:8506.
41. Thieblemont C, de Guibert S, Dupuis J, et al. Phase II study of anti-CD19 antibody drug conjugate (SAR3419) in combination with rituximab: clinical activity and safety in patients with relapsed/refractory diffuse large B-cell lymphoma (NCT01470456). Blood. 2013;122:4395.
42. Moskowitz CH, Fanale MA, Shah BD, et al. A phase 1 study of denintuzumab mafodotin (SGN-CD19A) in relapsed/refractory B-lineage non-Hodgkin lymphoma. Blood. 2015;126:182.
43. Fathi AT, Borate U, DeAngelo DJ, et al. A phase 1 study of denintuzumab mafodotin (SGN-CD19A) in adults with relapsed or refractory B-lineage acute leukemia (B-ALL) and highly aggressive lymphoma. Blood. 2015;126:1328.
44. Fathi AT, Chen R, Trippett TM, et al. Interim analysis of a phase 1 study of the antibody-drug conjugate SGN-CD19A in relapsed or refractory B-lineage acute leukemia and highly aggressive lymphoma. Blood. 2014;124:963.
45. Coleman M, Goldenberg DM, Siegel AB, et al. Epratuzumab: targeting B-cell malignancies through CD22. Clin Cancer Res. 2003;9:3991S–4S.
46. Leonard JP, Coleman M, Ketas JC, et al. Phase I/II trial of epratuzumab (humanized anti-CD22 antibody) in indolent non-Hodgkin's lymphoma. J Clin Oncol. 2003;21:3051–9.
47. Leonard JP, Coleman M, Ketas JC, et al. Epratuzumab, a humanized anti-CD22 antibody, in aggressive non-Hodgkin's lymphoma: phase I/II clinical trial results. Clin Cancer Res. 2004;10:5327–34.
48. Micallef IN, Maurer MJ, Wiseman GA, et al. Epratuzumab with rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone chemotherapy in patients with previously untreated diffuse large B-cell lymphoma. Blood. 2011;118:4053–61.
49. Grant BW, Jung SH, Johnson JL, et al. A phase 2 trial of extended induction epratuzumab and rituximab for previously untreated follicular lymphoma: CALGB 50701. Cancer. 2013;119:3797–804.
50. Morschhauser F, Kraeber-Bodere F, Wegener WA, et al. High rates of durable responses with anti-CD22 fractionated radioimmunotherapy: results of a multicenter, phase I/II study in non-Hodgkin's lymphoma. J Clin Oncol. 2010;28:3709–16.
51. Chen AI, Lebovic D, Brunvand MW, et al. Final results of a phase I study of the anti-CD22 antibody-drug conjugate (ADC)

DCDT2980S with or without rituximab (RTX) in patients (pts) with relapsed or refractory (R/R) B-cell non-Hodgkin's lymphoma (NHL). Blood. 2013;122:4399.
52. Morschhauser F, Flinn I, Advani RH, et al. Updated results of a phase II randomized study (ROMULUS) of polatuzumab vedotin or pinatuzumab vedotin plus rituximab in patients with relapsed/refractory non-Hodgkin lymphoma. Blood. 2014;124:4457.
53. Rytting M, Triche L, Thomas D, et al. Initial experience with CMC-544 (inotuzumab ozogamicin) in pediatric patients with relapsed B-cell acute lymphoblastic leukemia. Pediatr Blood Cancer. 2014;61:369–72.
54. Wagner-Johnston ND, Goy A, Rodriguez MA, et al. A phase 2 study of inotuzumab ozogamicin and rituximab, followed by autologous stem cell transplant in patients with relapsed/refractory diffuse large B-cell lymphoma. Leuk Lymphoma. 2015;56(10):2863–9.
55. Palanca-Wessels MC, Czuczman M, Salles G, et al. Safety and activity of the anti-CD79B antibody-drug conjugate polatuzumab vedotin in relapsed or refractory B-cell non-Hodgkin lymphoma and chronic lymphocytic leukaemia: a phase 1 study. Lancet Oncol. 2015;16:704–15.
56. Haas C, Krinner E, Brischwein K, et al. Mode of cytotoxic action of T cell-engaging BiTE antibody MT110. Immunobiology. 2009;214:441–53.
57. Bargou R, Leo E, Zugmaier G, et al. Tumor regression in cancer patients by very low doses of a T cell-engaging antibody. Science. 2008;321:974–7.
58. Viardot A, Goebeler M-E, Hess G, et al. Phase 2 study of bispecific T-cell engager (BiTE®) antibody blinatumomab in relapsed/refractory diffuse large B cell lymphoma. Blood. 2016;127:1410.
59. Teachey DT, Rheingold SR, Maude SL, et al. Cytokine release syndrome after blinatumomab treatment related to abnormal macrophage activation and ameliorated with cytokine-directed therapy. Blood. 2013;121:5154–7.
60. Oak E, Bartlett NL. Blinatumomab for the treatment of B-cell lymphoma. Expert Opin Investig Drugs. 2015;24:715–24.
61. Eyre TA, Collins GP. Immune checkpoint inhibition in lymphoid disease. Br J Haematol. 2015;170:291–304.
62. Parry RV, Chemnitz JM, Frauwirth KA, et al. CTLA-4 and PD-1 receptors inhibit T-cell activation by distinct mechanisms. Mol Cell Biol. 2005;25:9543–53.
63. Andorsky DJ, Yamada RE, Said J, et al. Programmed death ligand 1 is expressed by non-Hodgkin lymphomas and inhibits the activity of tumor-associated T cells. Clin Cancer Res. 2011;17:4232–44.
64. Ansell SM, Hurvitz SA, Koenig PA, et al. Phase I study of ipilimumab, an anti-CTLA-4 monoclonal antibody, in patients with relapsed and refractory B-cell non-Hodgkin lymphoma. Clin Cancer Res. 2009;15:6446–53.
65. Galligan BM, Tsao-Wei D, Groshen S, et al. Efficacy and safety of combined rituximab and ipilimumab to treat patients with relapsed/refractory CD20+ B-cell lymphoma. Blood. 2015;126:3977.
66. Westin JR, Chu F, Zhang M, et al. Safety and activity of PD1 blockade by pidilizumab in combination with rituximab in patients with relapsed follicular lymphoma: a single group, open-label, phase 2 trial. Lancet Oncol. 2014;15:69–77.
67. Armand P, Nagler A, Weller EA, et al. Disabling immune tolerance by programmed death-1 blockade with pidilizumab after autologous hematopoietic stem-cell transplantation for diffuse large B-cell lymphoma: results of an international phase II trial. J Clin Oncol. 2013;31:4199–206.
68. Lesokhin AM, Ansell SM, Armand P, et al. Preliminary results of a phase I study of nivolumab (BMS-936558) in patients with relapsed or refractory lymphoid malignancies. Blood. 2014;124:291.
69. Kershaw MH, Teng MW, Smyth MJ, et al. Supernatural T cells: genetic modification of T cells for cancer therapy. Nat Rev Immunol. 2005;5:928–40.
70. Maude SL, Laetsch TW, Buechner J, et al. Tisagenlecleucel in children and young adults with B-cell lymphoblastic leukemia. N Engl J Med. 2018;378:439–48.
71. Neelapu SS, Locke FL, Bartlett NL, et al. Axicabtagene ciloleucel CAR T-cell therapy in refractory large B-cell lymphoma. N Engl J Med. 2017;377:2531–44.
72. Lee DW, Gardner R, Porter DL, et al. Current concepts in the diagnosis and management of cytokine release syndrome. Blood. 2014;124:188–95.
73. Schuster SJ, Svoboda J, Nasta S, et al. Phase IIa trial of chimeric antigen receptor modified T cells directed against CD19 (CTL019) in patients with relapsed or refractory CD19+ lymphomas. J Clin Oncol. 2015;33:abstr 8516.

Hematopoietic Stem Cell Transplantation

25

Tetsuya Mori and Tomoo Osumi

Introduction

Hematopoietic stem cell transplantation (HSCT) is an established treatment approach for many malignant and nonmalignant diseases that affect the hematopoietic and immune system. Advances in HSCT procedures include understanding of the critical role of histocompatibility in allogeneic HSCT, development of methods to accurately type donors and recipients, increasing numbers of donors in large registries of unrelated donors and cord blood (CB) units, identification of additional sources of hematopoietic stem cells other than bone marrow (BM) such as peripheral blood (PB) and umbilical CB, improvement in graft-versus-host disease (GVHD) prophylaxis and treatment, and improvement in supportive care during the posttransplant periods. These advances have contributed to making HSCT a more common and successful treatment option. Generally, indications for HSCT should be considered for patients in whom HSCT is likely to benefit their survival compared with other therapeutic methods. In the treatment for malignant disease, standard treatment options and outcomes for each disease have been changing over time. Careful comparisons among the various treatment approaches including HSCT should be performed to consider the indications for HSCT.

Non-Hodgkin's lymphoma (NHL) accounts for approximately 7% of childhood malignant diseases. Compared with NHL in adults, the distribution of the histologic subtypes of childhood NHL shows a preponderance of aggressive variants. Childhood NHL is classified into four major histologic subtypes, namely, Burkitt lymphoma (BL), diffuse large B-cell lymphoma (DLBCL), lymphoblastic lymphoma (LBL), and anaplastic large cell lymphoma (ALCL). To date, the long-term event-free survival (EFS) of children with newly diagnosed NHL has reached 70–90% depending upon histologic subtype [1]. These results have been achieved with the use of multi-agent chemotherapy without HSCT. Therefore, HSCT will be generally reserved for use in children with NHL only after treatment failure, such as relapse, progression, or induction failure. However, the prognosis for children with relapsed or refractory (R/R) NHL is very poor with some exceptions; the optimal treatment for these children has not been established. In fact, HSCT may provide a curative treatment option for some of these patients. It is not easy to clearly assert effectiveness of HSCT because of the small number of such patients and little consistency in their therapeutic approach.

In this chapter, the reported experiences with administering HSCT in childhood NHL are provided.

HSCT for B-NHL in Children

Major histological subtypes of childhood aggressive mature B-cell NHL (B-NHL) are BL and DLBCL. BL accounts for 30–45% of childhood NHL. The BL cells show a mature B-cell phenotype and are negative for the enzyme terminal deoxynucleotidyl transferase (TdT) and positive for surface immunoglobulin with either kappa or lambda light chains. Usually, BL cases have a chromosomal translocation involving the *c-myc* gene. Clinically, jaw involvement is common in endemic BL, and abdominal involvement is common in sporadic BL. Bone marrow involvement is observed in approximately 20% of cases of sporadic BL. Other sites of involvement include testes, bone, skin, and central nervous system (CNS) [1]. DLBCL accounts for 10–20% of childhood NHL. DLBCL in children differs biologically from DLBCL in older adults.

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The majority of pediatric DLBCL cases have a germinal center B-cell phenotype. Specific characteristic cytogenetic abnormalities associated with DLBCL have not been reported. Clinical presentations of pediatric DLBCL may be similar to that of BL, although more often it is localized, and less often it involves the BM or CNS. Unlike in adult patients, standard treatment for BL is the same as treatment for DLBCL in children [1]. In the current standard protocols, patients are stratified into several risk groups according to stage, tumor resection status, pretreatment serum LDH value, and presence of CNS/BM disease. For patients with lower-risk B-NHL, more than 95% EFS can be achieved using two to four cycles of multi-agent chemotherapy [1–6]. For patients with higher-risk B-NHL, four to eight cycles of more intensive multi-agent conventional chemotherapy result in EFS of 80% to 90% [1–4, 6, 7]. A recent international randomized trial showed the benefit of adding rituximab to standard chemotherapy for advanced B-NHL [8]. HSCT for primary pediatric B-NHL is generally not recommended because of the preferred initial treatment with excellent outcome as described above.

The prognosis of relapsed/refractory (R/R) B-NHL in children has been reported to be poor [9–15]. However, for R/R B-NHL in children, the role of HSCT has been unclear. There has been no evidence provided by prospective clinical trials for HSCT in pediatric R/R B-NHL (Table 25.1).

Table 25.1 Outcomes of HSCT for refractory or relapsed B-NHL in major reported case series

Author	No. of patients	Status at HSCT	Number of Auto-/Allo-HSCT	Survival
Ladenstein et al. [16]	89	CR = 33, PR = 27, NR = 29	89/0	HSCT in CR = 54.2%, in PR = 37.5%, NR = 0%
Attarbaschi et al. [11]	4	CR = 3, PD = 1	3/1	HSCT in CR = 33%, in NR/PD = 0%
Fujita et al. [12]	13	CR, PR = 5, non-CR, PD = 8	3/10	HSCT in CR/PR = 80%, in PD = 0%
Anoop et al. [13]	16	NA	12/4	56%, all survivors received HSCT in CR
Jourdain et al. [14]	41	CR = 32, CRu = 2, non-CR = 7	33/8	HSCT in CR/CRu = 54.3%, in NR/PD = 0%
Osumi et al. [15]	20	CR, PR = 14, non-CR, PD = 6	6/14	HSCT in CR/PR = 50%, in NR/PD = 0%

Abbreviations: *HSCT* hematopoietic stem cell transplantation, *auto-* autologous, *allo-* allogeneic, *B-NHL* mature B-cell lymphoma/leukemia, *CR* complete remission, *PR* partial remission, *NR* no response, *PD* progressive disease, *CRu* unconfirmed complete remission, *NA* not available

A UK group conducted a 10-year retrospective multicenter study of R/R B-NHL [13]. Nine of 33 (27.3%) patients survived, and all of them underwent transplantation within complete remission (CR) (eight of them underwent auto-HSCT with BEAM [carmustine, etoposide, cytarabine, and melphalan] conditioning and one had allo-HSCT). All children who did not receive transplants died. They concluded that auto-HSCT with BEAM conditioning in CR offers the best chance of cure.

A recent retrospective analysis from the French clinical trial registries reported that the 5-year overall survival (OS) rate was 29.9% for 67 cases of pediatric R/R B-NHL [14]. Among them, 41 patients who were not considered as having progressive disease after salvage treatment received high-dose chemotherapy with HSCT and 18 patients (46.3%) survived. The response status at the time of HSCT was not significantly associated with survival, although the survival rate of the patients who received transplants in definitive or unconfirmed CR was significantly better than that of the other patients (54.3% vs 0%).

A Japanese group has reported two informative nationwide retrospective analyses [12, 15]. In the first study, 33 R/R B-NHL cases from late 1990 to early 2000 were analyzed and the 4-year OS rate was 21%. Four of five patients who received HSCT in PR or CR were alive, whereas none of the eight patients who received HSCT in PD survived. The second study showed the outcome of the 33 R/R B-NHL patients from the prospective study; the 5-year OS rate statistically increased to 48.5% compared with the previous study due to improvement in the CR rate by introduction of rituximab combination chemotherapy. Among the 20 patients who achieved CR or PR by rituximab combination chemotherapy, 14 patients received HSCT and seven were alive in CR while five of six patients survived without HSCT. Also in this study, no patient survived who received HSCT in PD. Those studies suggested that HSCT for R/R patients with good chemotherapy response was expected to have some effectiveness, although it is hard to rescue the patients with resistant disease by HSCT.

There is insufficient information about which donor type is appropriate in HSCT treatment for pediatric B-NHL. Ladenstein et al. reported the analysis of 89 patients with R/R BL receiving autologous HSCT from the European Lymphoma BMT registry [16]. Among them, the response of 23 patients with primary refractory disease or chemotherapy-resistant disease was dismal (5-EFS rates 0%), as all died within 1 year after autologous HSCT. On the other hand, the 5-year EFS of 38 patients with chemotherapy-sensitive disease was 48.7%, which suggested that the effectiveness of autologous HSCT for B-NHL refractory to modern intensive chemotherapy was limited.

Gross et al. reported a retrospective analysis that examined the role of HSCT for pediatric patients with R/R NHL

registered in the Center for International Blood and Marrow Transplant Research (CIBMTR) [17]. Their data suggested that survival or relapse rates were similar after allogeneic and autologous HSCT for BL and DLBCL. Also, in the French and Japanese studies mentioned, the authors concluded that the source of stem cells did not affect the outcome [12, 14, 15]. Further analysis will be needed to conclude which type of stem cell source is suitable for HSCT for R/R B-NHL.

HSCT for LBL in Children

LBL accounts for 20% to 30% of childhood NHL. In the World Health Organization (WHO) classification of lymphoid neoplasms, LBL is classified as the same disease as acute lymphoblastic leukemia (ALL) [18], even though there are controversies. The LBL cells are usually positive for TdT. Approximately 75% of cases show a T-cell immunophenotype, and the remainder show a precursor B-cell phenotype. The majority of children with T-cell LBL have enlarged cervical and mediastinal lymphadenopathy. Subdiaphragmatic disease (such as hepatosplenomegaly) and kidney involvement are often observed. In contrast, children with precursor B-cell LBL tend to have limited disease confined to the peripheral lymph nodes, skin, and bone [1]. For children with untreated LBL, the use of treatment regimens for ALL results in EFS of 80% to 90% [19–24]. Advanced stage, the presence of minimal disseminated disease (MDD) [25, 26], response to therapy as evaluated by resolution of tumor [19] or minimal residual disease (MRD) [27], and some biological factors such as loss of heterozygosity at chromosome 6q [28, 29] and *NOTCH1* mutations [28] have been reported as prognostic factors. Consequently, HSCT for LBL in children and adolescents was not recommended as first-line treatment.

The prognosis of R/R LBL in children and adolescents was also poor, and their reported survival rates were around 10–40% [11, 30–33]. Most of the surviving patients received HSCT, while there had been no prospective trial to directly validate the effectiveness of HSCT for R/R LBL in children and adolescents (Table 25.2).

The Berlin–Frankfurt–Münster (BFM) group reported the retrospective analysis of 34 R/R LBL patients from their registration of 324 LBL patients [31]. The median survival time and the 5-year OS rate were 5.1 months and 14%, respectively. All survivors received allo-HSCT, and all other patients treated with chemotherapy or auto-HSCT died. They assumed that allo-HSCT could be beneficial for patients who could achieve a second remission.

A Japanese group reported 48 R/R LBL patients from a nationwide survey from 260 patients registered in a prospective trial [32]. Their 3-year OS was 36% with a median observation period of 27.5 months. Three patients out of 33 were alive under receiving chemotherapy alone, and 14 of 33 patients were alive after HSCT. In terms of stem cell sources, three of 14 were alive after auto-HSCT and 12 of 26 were alive after allo-HSCT.

Table 25.2 Outcomes of allogeneic HSCT for refractory or relapsed LBL in major reported case series

Author	No. of patients	Status at HSCT	Survival
Burkhardt et al. [31]	12	CR = 6, PR = 4, non-CR = 1, ND = 1	HSCT in CR = 33%, in PR = 50%
Mitsui et al. [32]	19	CR = 14, PR = 6, PD = 6	HSCT in CR = 50%, in PR = 66.6%, in PD = 16.7%
Michaux et al. [33]	8	CR = 4, non-CR/PD = 4	HSCT in CR = 50%, in non-CR/PD = 0%

Abbreviations: *HSCT* hematopoietic stem cell transplantation *LBL* lymphoblastic lymphoma, *CR* complete remission, *PR* partial remission, *ND* no data, *PD* progressive disease

The European Organization for Research and Treatment of Cancer (EORTC) reported the treatment, and outcome of 23 R/R LBL patients from 197 patients registered in their clinical trials [33]. Among them, 10 patients received HSCT (allo-HSCT for eight patients, auto-HSCT for two patients) and only two patients who underwent allo-HSCT in CR survived out of all of them. They suggested that allo-HSCT after a second CR is the only potentially curative therapeutic option for R/R LBL in children.

The CIBMTR study demonstrated the superiority of allo-HSCT to auto-HSCT [17]. They analyzed 53 cases of R/R LBL receiving HSCT, and the 5-year probabilities of relapse/progression of auto-HSCT and allo-HSCT were 86% and 23%, respectively, far lower after allo-HSCT compared to auto-HSCT. Similarly, their 5-year EFS rates were 4% and 40%, respectively. In conclusion, allo-HSCT could be the only promising option for R/R LBL patients when they achieve CR.

HSCT for ALCL in Children

ALCL accounts for 10–15% of childhood NHL. In the 2016 revision of the WHO classification of lymphoid neoplasms, ALCL is classified into a group of mature T-cell and NK-cell neoplasms, and is subdivided into "ALCL, ALK+" and "ALCL, ALK–". All ALCL cases are CD30-positive, and more than 90% of childhood ALCL cases have a chromosomal rearrangement involving the *anaplastic lymphoma kinase* (*ALK*) gene. Clinically, childhood ALCL cases are characterized by involvement of lymph nodes and a variety of extranodal sites, particularly skin and bone, and systemic symptoms such as B-symptoms [1]. Commonly used chemo-

therapy regimens for children with ALCL result in disease-free survival (DFS) of 60–80%; however, current evidence does not suggest superiority of one treatment regimen over another for the standard treatment options [34–45]. Even though there are controversies, advanced disease involving the mediastinum, viscera, skin, or bone marrow [39, 46], the presence of a small-cell or lymphohistiocytic component [47], the presence of MDD detected by RT-PCR [48, 49], an immune response against the ALK protein (i.e., anti-ALK antibody titer) [49, 50], and response to therapy evaluated by MRD [51] have been reported as prognostic factors. Because outcomes of untreated children with ALCL by current chemotherapy regimens are basically favorable, treatment options using HSCT are generally reserved for first relapse, progression, or induction failure.

The relapse rates of childhood ALCL are 25–35% with current first-line strategies. Although available data on children with relapsed ALCL are limited to retrospective analyses, children with relapsed ALCL have a DFS of approximately 40–60% [11, 52, 53]. The standard approach for them has not been established. Salvage chemotherapy, followed by auto-HSCT or allo-HSCT, has been employed in this setting. Recent and important reported experiences with HSCTs for R/R childhood ALCL include the following major results, which are summarized in Table 25.3 (auto-HSCT) and Table 25.4 (allo-HSCT).

The European Group for Blood and Marrow Transplantation (EBMT) reported on the results of auto-HSCT for 64 adult and pediatric patients with ALCL. In this series, 14 of 15 patients who received auto-HSCT in the first CR maintained the remission over time. On the other hand, none of the patients who had refractory disease achieved CR. OS and progression-free survival (PFS) at 10 years of the whole population were 70.3% and 47.0%, respectively. The OS of 18 patients aged less than 20 years was significantly better than that of the adult population, but there was no difference in PFS [54]. From this study, auto-HSCT appears to be effective for young patients with relapsed ALCL if CR can be achieved before initiating conditioning therapy.

The BFM group reported the largest series of 74 children with relapsed ALCL. The recommended salvage strategy was re-induction chemotherapy followed by auto-HSCT. A 5-year OS of all patients after the first relapse was 57%. Eighteen of 19 patients treated without HSCT experienced relapse again during or after salvage chemotherapy and died. Among them, 10 patients with the intention to perform HSCT experienced progression again before HSCT and died of the disease. According to the recommendation, 39 patients received auto-HSCT. Among them, 21 patients stayed in

Table 25.3 Outcomes of autologous HSCT for refractory or relapsed ALCL in major reported case series

Study group	EBMT [54]	BFM [53]	SFCE [56]	Japan [59]	CIBMTR [17]
Study periods	1983–1996	1990–2003	1975–1997	1990–2010	1990–2005
Number of patients	64	39	14	23	24
Status at HSCT: CR/non-CR	30(15 in CR1)/34	NA	14/0	16/7	NA
5-year EFS	56.4%	59%	45%	38%	35%
Progression or relapse	41%	41%	40%	49%	48%
TRD	2%	5%	7%	12%	14%

Abbreviations: *HSCT* hematopoietic stem cell transplantation, *ALCL* anaplastic large cell lymphoma, *CR* complete remission, *EFS* event-free survival, *TRD* treatment-related death, *NA* not available

Table 25.4 Outcomes of allogeneic HSCT for refractory or relapsed ALCL in major reported case series

Study group	BFM [55]	SFCE [57]	Japan [59]	CIBMTR [17]
Study periods	1991–2003	1993–2011	1990–2010	1990–2005
Number of patients	20	34	24	12
Status at HSCT: CR/non-CR	12/6 NA	28/6	8/16	NA
Conditioning regimen	TBI in 15	MAC in 31	TBI in 18 RIC in 4	NA
Donor	MSD in 8 UD in 8 MMRD in 4	MRD in 12 UD in 22	MRD in 7 MUD in 2 MMRD in 6 MMUD in 7	NA
Stem cell source	BM in 8 PB in 12	BM in 16 PB in 8 CB in 10	BM in 13 PB in 5 CB in 6	NA
5-year EFS	75%	58%	50%	46%
Progression or relapse	10%	18%	28%	20%
Acute GVHD	II to IV in 8	III to IV in 5	II to IV in 9	NA
Chronic GVHD	Extensive in 4	In 5	NA	NA
TRD	15%	24%	25%	25%

Abbreviations: *HSCT* hematopoietic stem cell transplantation, *ALCL* anaplastic large cell lymphoma, *CR* complete remission, *EFS* event-free survival, *TRD* treatment-related death, *TBI* total body irradiation, *MAC* myeloablative conditioning, *RIC* reduced-intensity conditioning, *MSD* matched sibling donor, *UD* unrelated donor, *MMRD* mismatched related donor, *MRD* matched related donor, *MUD* matched unrelated donor, *MMUD* mismatched unrelated donor, *BM* bone marrow, *PB* peripheral blood, *CB* cord blood, *NA* not available

remission, two patients died of treatment-related complications, and 16 patients experienced progression again. The EFS of 39 children who received the recommended auto-HSCT was 59%. Sixteen patients received allo-HSCT as primary HSCT. Among them, 11 patients stayed in remission, three patients died of treatment-related complications, and two patients experienced progression and died of the disease. Outcomes of patients with bone marrow or CNS involvement, relapse during first-line therapy, or CD3-positive ALCL were poor [53]. The BFM group also reported their experiences with allo-HSCT for children with R/R ALCL. Nine patients received allo-HSCT after the first R/R disease and 11 after multiple relapses. Eight patients received their transplants from matched sibling donors (MSD), eight patients from unrelated donors (UD), and four patients from haploidentical family donors. The conditioning regimen was based on total body irradiation (TBI) in 15 patients. Fifteen patients survived in CR, three patients died of treatment-related complications, and two patients experienced progression and died of the disease. Grade II to IV acute GVHD occurred in four of eight patients who received transplants from MSD and three of eight patients who received transplants from UD. Extensive chronic GVHD was seen in two patient who received a transplant from a matched sibling donor and two from an unrelated donor [55].

The French group reported on 41 children with relapsed ALCL. The OS of all patients was 69%. Among 36 patients who achieved a second remission, 14 patients received auto-HSCT and one patient received allo-HSCT. Nine patients survived in CR, one patient died of treatment-related complications, and five patients experienced subsequent relapses. The DFS for patients treated with HSCT in their second remission was 45%, whereas that for patients treated without HSCT was 52%. Outcomes for patients who developed relapse within 12 months of diagnosis were as poor as a DFS of 28%, regardless of salvage therapy [56]. The French group subsequently reported a series of 34 allo-HSCTs for children with relapsed ALCL registered in the Société Française de Greffe de Moelle et de Thérapie Cellulaire (SFGM-TC) database. At the time of HSCT, 28 patients were in CR, and six patients had detectable disease. Twelve patients received their transplants from HLA-matched related donors, and 22 patients received from UD. Sources of stem cells were BM in 16 patients, PB in eight, and CB in 10. Myeloablative conditioning (MAC) regimens were used in 31 patients. The EFS, cumulative incidence of relapse, and treatment-related mortality were 58%, 18%, and 24%, respectively. The 100-day cumulative incidence of acute GVHD was 73.5%, and grade III to IV acute GVHD occurred in five patients. Five patients harbored chronic GVHD. Among six patients who experienced relapse after allo-HSCT, four patients were alive in CR, including three after vinblastine–corticosteroid therapy and one after donor lymphocyte infusions [57]. As a treatment option other than HSCT, the French group also reported that vinblastine was active as a single agent in relapsed ALCL. Vinblastine-induced CR occurred in 25 of 30 evaluable patients, and nine of these patients remained in CR, with a median follow-up of 7 years from the end of treatment [58].

The Japanese group initially reported consistent results with the series of 26 children with relapsed. Twenty-four patients achieved a second remission. Auto-HSCT and allo-HSCT were performed in six and eight patients, respectively. Relapse-free survival was 53% for chemotherapy alone, 33% for auto-HSCT, and 100% for allo-HSCT [52]. Although these results may suggest an advantage for allo-HSCT in the treatment for R/R ALCL, data from the following two series did not show any statistical difference between auto-HSCT and allo-HSCT.

From the CIBMTR, a series of 36 HSCT procedures for children with R/R ALCL was reported. Twenty-four patients received auto-HSCT and 12 patients received allo-HSCT. The EFS was 35% after auto-HSCT versus 46% after allo-HSCT. Cumulative incidence of progress or relapse was 48% after auto-HSCT versus 20% after allo-HSCT. Although cumulative incidence of progress or relapse seemed lower after allo-HSCT, these differences were not statistically significantly different [17].

The Japanese group subsequently reported data from the series of 47 HSCTs for children with R/R ALCL registered in the Transplant Registry Unified Management Program (TRUMP) system of the Japan Society for Hematopoietic Cell Transplantation. Among 23 patients who received auto-HSCT, 16 patients were in CR, and seven patients had detectable disease at the time of HSCT. TBI/total lymphoid irradiation (TLI)-based conditioning regimens were used in eight patients. The EFS, cumulative incidence of relapse, and treatment-related mortality were 38%, 49%, and 12%, respectively. Among 24 patients who received allo-HSCT, eight patients were in CR, and 16 patients had detectable disease at the time of HSCT. Four of them had received auto-HSCT previously. Seven patients received their transplants from HLA-matched related donors, two patients received from HLA-matched UD, and 13 patients received from HLA-mismatched donors. Sources of stem cells were BM in 13 patients, PB in five, and CB in six. TBI/TLI-based conditioning regimens were used in 18 patients, and reduced-intensity conditioning (RIC) regimens were used in four patients. The EFS, cumulative incidence of relapse, and treatment-related mortality were 50%, 28%, and 25%, respectively. Differences in those values between auto-HSCT and allo-HSCT were not statistically significantly different. Acute GVHD of any grade was observed in 13 patients, including grade III to IV acute GVHD in nine patients. Although three patients who received allo-HSCT using RIC regimens had not been in CR at HSCT, they achieved CR and survived 32–65 months after HSCT [59].

Based on these findings, auto-HSCT may be used to treat children with relapsed ALCL who have achieved subsequent CR by salvage chemotherapy and do not have any poor prognostic factor, such as early relapse. Allo-HSCT may be a preferable treatment option for other children with R/R ALCL, including those who failed to achieve CR by salvage chemotherapy. Although the experiences are limited, allo-HSCT using the RIC regimen may be tolerable and effective. Further study, ideally a prospective study including a large number of patients, will be required to clearly define the indication of HSCT for R/R ALCL.

Recently, the efficacies of molecular-targeted therapy for R/R ALCL have been explored. Brentuximab vedotin, an anti-CD30 antibody–drug conjugate, induced complete responses in 38 of 58 patients in a phase II study of adults [60]. After a median observation period of approximately 6 years, 16 of the 38 patients remained in remission without the start of new therapy other than consolidative HSCT. Of the 16 patients, four patients underwent consolidative auto-HSCT, and four other patients underwent consolidative allo-HSCT. The remaining eight patients remained in remission without consolidative HSCT or any additional anticancer therapy [61]. Crizotinib, an ALK inhibitor, induced complete responses in 21 of 26 patients in a pediatric phase I study with a phase II extension [62, 63]. Although robust and sustained clinical responses to crizotinib have been observed, the optimal duration of therapy remains unclear [64]. These novel treatment strategies may reduce relapse rates and improve outcome for R/R ALCL patients compared with conventional strategies using HSCT. Further studies are required to recognize distinct subgroups of pediatric ALCL with different clinical, phenotypic, and biologic characteristics, which will identify patients who benefit from treatment using HSCT.

HSCT for Rare NHL Occurring in Children

Low-grade or intermediate-grade mature B-cell lymphomas, such as pediatric-type follicular lymphoma and marginal zone lymphoma, and peripheral T-cell lymphoma (PTCL) excluding ALCL are rarely seen in children. Optimal therapy for these rare NHLs is unclear because only case reports and small case series are guides for therapy.

Because the number of pediatric patients with low-grade or intermediate-grade mature B-cell lymphomas is limited and their reported outcomes are generally favorable [65–72], meaningful experiences of HSCT in the treatment for this rare NHL cannot be found.

Although the optimal therapy for children with PTCL is unclear, HSCT may be an option for certain patients. The Japanese group reported retrospective analysis of 21 cases of pediatric PTCL excluding ALCL with a 5-year OS of 85.2%. Among them, two patients received auto-HSCT and nine patients received allo-HSCT in their first-line treatment [73]. In three other retrospective reports, HSCTs were not used in the first-line treatment for PTCL. The United Kingdom Children's Cancer Study Group (UKCCSG) reported on 25 cases of pediatric PTCL treated by standard T-cell LBL or B-cell NHL regimens. Their 5-year survival rate was approximately 50% [74]. The Children's Oncology Group (COG) reported on 20 patients. Eight of 10 patients with low-stage disease achieved long-term DFS compared with only four of 10 patients with high-stage disease [75]. The BFM study group reported 38 cases. Patients with PTCL not otherwise specified treated with mainly ALCL regimens had a 10-year EFS rate of 61%. Patients with NK/T-cell lymphoma had a 10-year EFS rate of 17% [76]. There is insufficient experience to determine the role of HSCT in the treatment for childhood PTCL excluding ALCL.

References

1. Gross TGPS. Malignant non-Hodgkin lymphoma in children. In: Pizzo PAPD, editor. Principles and practice of pediatric oncology. 6th ed. Philadelphia: Lippincott Williams & Wilkins; 2010. p. 663–82.
2. Minard-Colin V, Brugieres L, Reiter A, Cairo MS, Gross TG, Woessmann W, et al. Non-Hodgkin lymphoma in children and adolescents: Progress through effective collaboration, current knowledge, and challenges ahead. J Clin Oncol. 2015;33(27):2963–74.
3. Woessmann W, Seidemann K, Mann G, Zimmermann M, Burkhardt B, Oschlies I, et al. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. Blood. 2005;105(3):948–58.
4. Tsurusawa M, Mori T, Kikuchi A, Mitsui T, Sunami S, Kobayashi R, et al. Improved treatment results of children with B-cell non-Hodgkin lymphoma: a report from the Japanese Pediatric Leukemia/Lymphoma Study Group B-NHL03 study. Pediatr Blood Cancer. 2014;61:1215–21.
5. Gerrard M, Cairo MS, Weston C, Auperin A, Pinkerton R, Lambilliote A, et al. Excellent survival following two courses of COPAD chemotherapy in children and adolescents with resected localized B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study. Br J Haematol. 2008;141(6):840–7.
6. Patte C, Auperin A, Gerrard M, Michon J, Pinkerton R, Sposto R, et al. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. Blood. 2007;109(7):2773–80.
7. Cairo MS, Gerrard M, Sposto R, Auperin A, Pinkerton CR, Michon J, et al. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. Blood. 2007;109(7):2736–43.
8. Minard-Colin V, Auperin A, Pillon M, Burke A, Anderson JR, Barkauskas DA, et al. Results of the randomized Intergroup trial Inter-B-NHL Ritux 2010 for children and adolescents with high-risk B-cell non-Hodgkin lymphoma (B-NHL) and mature acute leukemia (B-AL): evaluation of rituximab (R) efficacy in addition to standard LMB chemotherapy (CT) regimen. J Clin Oncol. 2016;34(15_suppl):10507.

9. Philip T, Hartmann O, Pinkerton R, Zucker JM, Gentet JC, Lamagnere JP, et al. Curability of relapsed childhood B-cell non-Hodgkin's lymphoma after intensive first line therapy: a report from the Societe Francaise d'Oncologie Pediatrique. Blood. 1993;81(8):2003–6.
10. Atra A, Gerrard M, Hobson R, Imeson JD, Hann IM, Pinkerton CR. Outcome of relapsed or refractory childhood B-cell acute lymphoblastic leukaemia and B-cell non-Hodgkin's lymphoma treated with the UKCCSG 9003/9002 protocols. Br J Haematol. 2001;112(4):965–8.
11. Attarbaschi A, Dworzak M, Steiner M, Urban C, Fink FM, Reiter A, et al. Outcome of children with primary resistant or relapsed non-Hodgkin lymphoma and mature B-cell leukemia after intensive first-line treatment: a population-based analysis of the Austrian Cooperative Study Group. Pediatr Blood Cancer. 2005;44(1):70–6.
12. Fujita N, Mori T, Mitsui T, Inada H, Horibe K, Tsurusawa M, et al. The role of hematopoietic stem cell transplantation with relapsed or primary refractory childhood B-cell non-Hodgkin lymphoma and mature B-cell leukemia: a retrospective analysis of enrolled cases in Japan. Pediatr Blood Cancer. 2008;51(2):188–92.
13. Anoop P, Sankpal S, Stiller C, Tewari S, Lancaster DL, Khabra K, et al. Outcome of childhood relapsed or refractory mature B-cell non-Hodgkin lymphoma and acute lymphoblastic leukemia. Leuk Lymphoma. 2012;53(10):1882–8.
14. Jourdain A, Auperin A, Minard-Colin V, Aladjidi N, Zsiros J, Coze C, et al. Outcome of and prognostic factors for relapse in children and adolescents with mature B-cell lymphoma and leukemia treated in three consecutive prospective "Lymphomes Malins B" protocols. A Societe Francaise des Cancers de l'Enfant study. Haematologica. 2015;100(6):810–7.
15. Osumi T, Mori T, Fujita N, Saito AM, Nakazawa A, Tsurusawa M, et al. Relapsed/refractory pediatric B-cell non-Hodgkin lymphoma treated with rituximab combination therapy: a report from the Japanese Pediatric Leukemia/Lymphoma Study Group. Pediatr Blood Cancer. 2016;63(10):1794–9.
16. Ladenstein R, Pearce R, Hartmann O, Patte C, Goldstone T, Philip T. High-dose chemotherapy with autologous bone marrow rescue in children with poor-risk Burkitt's lymphoma: a report from the European Lymphoma Bone Marrow Transplantation Registry. Blood. 1997;90(8):2921–30.
17. Gross TG, Hale GA, He W, Camitta BM, Sanders JE, Cairo MS, et al. Hematopoietic stem cell transplantation for refractory or recurrent non-Hodgkin lymphoma in children and adolescents. Biol Blood Marrow Transplant. 2010;16(2):223–30.
18. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127(20):2375–90.
19. Reiter A, Schrappe M, Ludwig WD, Tiemann M, Parwaresch R, Zimmermann M, et al. Intensive ALL-type therapy without local radiotherapy provides a 90% event-free survival for children with T-cell lymphoblastic lymphoma: a BFM group report. Blood. 2000;95(2):416–21.
20. Burkhardt B, Woessmann W, Zimmermann M, Kontny U, Vormoor J, Doerffel W, et al. Impact of cranial radiotherapy on central nervous system prophylaxis in children and adolescents with central nervous system-negative stage III or IV lymphoblastic lymphoma. J Clin Oncol. 2006;24(3):491–9.
21. Landmann E, Burkhardt B, Zimmermann M, Meyer U, Woessmann W, Klapper W, et al. Results and conclusions of the European Intergroup EURO-LB02 trial in children and adolescents with lymphoblastic lymphoma. Haematologica. 2017;102(12):2086–96.
22. Termuhlen AM, Smith LM, Perkins SL, Lones M, Finlay JL, Weinstein H, et al. Outcome of newly diagnosed children and adolescents with localized lymphoblastic lymphoma treated on Children's Oncology Group trial A5971: a report from the Children's Oncology Group. Pediatr Blood Cancer. 2012;59(7):1229–33.
23. Termuhlen AM, Smith LM, Perkins SL, Lones M, Finlay JL, Weinstein H, et al. Disseminated lymphoblastic lymphoma in children and adolescents: results of the COG A5971 trial: a report from the Children's Oncology Group. Br J Haematol. 2013;162(6):792–801.
24. Sunami S, Sekimizu M, Takimoto T, Mori T, Mitsui T, Fukano R, et al. Prognostic impact of intensified maintenance therapy on children with advanced lymphoblastic lymphoma: a report from the Japanese Pediatric Leukemia/Lymphoma Study Group ALB-NHL03 Study. Pediatr Blood Cancer. 2016;63(3):451–7.
25. Coustan-Smith E, Sandlund JT, Perkins SL, Chen H, Chang M, Abromowitch M, et al. Minimal disseminated disease in childhood T-cell lymphoblastic lymphoma: a report from the children's oncology group. J Clin Oncol. 2009;27(21):3533–9.
26. Mussolin L, Buldini B, Lovisa F, Carraro E, Disaro S, Lo Nigro L, et al. Detection and role of minimal disseminated disease in children with lymphoblastic lymphoma: the AIEOP experience. Pediatr Blood Cancer. 2015;62(11):1906–13.
27. Stark B, Avigad S, Luria D, Manor S, Reshef-Ronen T, Avrahami G, et al. Bone marrow minimal disseminated disease (MDD) and minimal residual disease (MRD) in childhood T-cell lymphoblastic lymphoma stage III, detected by flow cytometry (FC) and real-time quantitative polymerase chain reaction (RQ-PCR). Pediatr Blood Cancer. 2009;52(1):20–5.
28. Bonn BR, Rohde M, Zimmermann M, Krieger D, Oschlies I, Niggli F, et al. Incidence and prognostic relevance of genetic variations in T-cell lymphoblastic lymphoma in childhood and adolescence. Blood. 2013;121(16):3153–60.
29. Burkhardt B, Moericke A, Klapper W, Greene F, Salzburg J, Damm-Welk C, et al. Pediatric precursor T lymphoblastic leukemia and lymphoblastic lymphoma: differences in the common regions with loss of heterozygosity at chromosome 6q and their prognostic impact. Leuk Lymphoma. 2008;49(3):451–61.
30. Bhatia S, Robison LL, Francisco L, Carter A, Liu Y, Grant M, et al. Late mortality in survivors of autologous hematopoietic-cell transplantation: report from the Bone Marrow Transplant Survivor Study. Blood. 2005;105(11):4215–22.
31. Burkhardt B, Reiter A, Landmann E, Lang P, Lassay L, Dickerhoff R, et al. Poor outcome for children and adolescents with progressive disease or relapse of lymphoblastic lymphoma: a report from the berlin-frankfurt-muenster group. J Clin Oncol. 2009;27(20):3363–9.
32. Mitsui T, Mori T, Fujita N, Inada H, Horibe K, Tsurusawa M. Retrospective analysis of relapsed or primary refractory childhood lymphoblastic lymphoma in Japan. Pediatr Blood Cancer. 2009;52(5):591–5.
33. Michaux K, Bergeron C, Gandemer V, Mechinaud F, Uyttebroeck A, Bertrand Y. Relapsed or refractory lymphoblastic lymphoma in children: results and analysis of 23 patients in the EORTC 58951 and the LMT96 protocols. Pediatr Blood Cancer. 2016;63(7):1214–21.
34. Brugieres L, Deley MC, Pacquement H, Meguerian-Bedoyan Z, Terrier-Lacombe MJ, Robert A, et al. CD30(+) anaplastic large-cell lymphoma in children: analysis of 82 patients enrolled in two consecutive studies of the French Society of Pediatric Oncology. Blood. 1998;92(10):3591–8.
35. Seidemann K, Tiemann M, Schrappe M, Yakisan E, Simonitsch I, Janka-Schaub G, et al. Short-pulse B-non-Hodgkin lymphoma-type chemotherapy is efficacious treatment for pediatric anaplastic large cell lymphoma: a report of the Berlin-Frankfurt-Munster Group Trial NHL-BFM 90. Blood. 2001;97(12):3699–706.
36. Mori T, Kiyokawa N, Shimada H, Miyauchi J, Fujimoto J. Anaplastic large cell lymphoma in Japanese children: retrospective analysis of 34 patients diagnosed at the National Research Institute for Child Health and Development. Br J Haematol. 2003;121(1):94–6.
37. Laver JH, Kraveka JM, Hutchison RE, Chang M, Kepner J, Schwenn M, et al. Advanced-stage large-cell lymphoma in children and ado-

lescents: results of a randomized trial incorporating intermediate-dose methotrexate and high-dose cytarabine in the maintenance phase of the APO regimen: a Pediatric Oncology Group phase III trial. J Clin Oncol. 2005;23(3):541–7.
38. Rosolen A, Pillon M, Garaventa A, Burnelli R, d'Amore ES, Giuliano M, et al. Anaplastic large cell lymphoma treated with a leukemia-like therapy: report of the Italian Association of Pediatric Hematology and Oncology (AIEOP) LNH-92 protocol. Cancer. 2005;104(10):2133–40.
39. Lowe EJ, Sposto R, Perkins SL, Gross TG, Finlay J, Zwick D, et al. Intensive chemotherapy for systemic anaplastic large cell lymphoma in children and adolescents: final results of Children's Cancer Group Study 5941. Pediatr Blood Cancer. 2009;52(3):335–9.
40. Pillon M, Gregucci F, Lombardi A, Santoro N, Piglione M, Sala A, et al. Results of AIEOP LNH-97 protocol for the treatment of anaplastic large cell lymphoma of childhood. Pediatr Blood Cancer. 2012;59(5):828–33.
41. Brugieres L, Le Deley MC, Rosolen A, Williams D, Horibe K, Wrobel G, et al. Impact of the methotrexate administration dose on the need for intrathecal treatment in children and adolescents with anaplastic large-cell lymphoma: results of a randomized trial of the EICNHL Group. J Clin Oncol. 2009;27(6):897–903.
42. Attarbaschi A, Mann G, Rosolen A, Williams D, Uyttebroeck A, Marky I, et al. Limited stage I disease is not necessarily indicative of an excellent prognosis in childhood anaplastic large cell lymphoma. Blood. 2011;117(21):5616–9.
43. Wrobel G, Mauguen A, Rosolen A, Reiter A, Williams D, Horibe K, et al. Safety assessment of intensive induction therapy in childhood anaplastic large cell lymphoma: report of the ALCL99 randomised trial. Pediatr Blood Cancer. 2011;56(7):1071–7.
44. Le Deley MC, Rosolen A, Williams DM, Horibe K, Wrobel G, Attarbaschi A, et al. Vinblastine in children and adolescents with high-risk anaplastic large-cell lymphoma: results of the randomized ALCL99-vinblastine trial. J Clin Oncol. 2010;28(25):3987–93.
45. Williams D, Mori T, Reiter A, Woessman W, Rosolen A, Wrobel G, et al. Central nervous system involvement in anaplastic large cell lymphoma in childhood: results from a multicentre European and Japanese study. Pediatr Blood Cancer. 2013;60(10):E118–21.
46. Le Deley MC, Reiter A, Williams D, Delsol G, Oschlies I, McCarthy K, et al. Prognostic factors in childhood anaplastic large cell lymphoma: results of a large European intergroup study. Blood. 2008;111(3):1560–6.
47. Lamant L, McCarthy K, d'Amore E, Klapper W, Nakagawa A, Fraga M, et al. Prognostic impact of morphologic and phenotypic features of childhood ALK-positive anaplastic large-cell lymphoma: results of the ALCL99 study. J Clin Oncol. 2011;29(35):4669–76.
48. Damm-Welk C, Busch K, Burkhardt B, Schieferstein J, Viehmann S, Oschlies I, et al. Prognostic significance of circulating tumor cells in bone marrow or peripheral blood as detected by qualitative and quantitative PCR in pediatric NPM-ALK-positive anaplastic large-cell lymphoma. Blood. 2007;110(2):670–7.
49. Mussolin L, Damm-Welk C, Pillon M, Zimmermann M, Franceschetto G, Pulford K, et al. Use of minimal disseminated disease and immunity to NPM-ALK antigen to stratify ALK-positive ALCL patients with different prognosis. Leukemia. 2013;27(2):416–22.
50. Ait-Tahar K, Damm-Welk C, Burkhardt B, Zimmermann M, Klapper W, Reiter A, et al. Correlation of the autoantibody response to the ALK oncoantigen in pediatric anaplastic lymphoma kinase-positive anaplastic large cell lymphoma with tumor dissemination and relapse risk. Blood. 2010;115(16):3314–9.
51. Damm-Welk C, Mussolin L, Zimmermann M, Pillon M, Klapper W, Oschlies I, et al. Early assessment of minimal residual disease identifies patients at very high relapse risk in NPM-ALK-positive anaplastic large-cell lymphoma. Blood. 2014;123(3):334–7.
52. Mori T, Takimoto T, Katano N, Kikuchi A, Tabuchi K, Kobayashi R, et al. Recurrent childhood anaplastic large cell lymphoma: a retrospective analysis of registered cases in Japan. Br J Haematol. 2006;132(5):594–7.
53. Woessmann W, Zimmermann M, Lenhard M, Burkhardt B, Rossig C, Kremens B, et al. Relapsed or refractory anaplastic large-cell lymphoma in children and adolescents after Berlin-Frankfurt-Muenster (BFM)-type first-line therapy: a BFM-group study. J Clin Oncol. 2011;29(22):3065–71.
54. Fanin R, Ruiz de Elvira MC, Sperotto A, Baccarani M, Goldstone A. Autologous stem cell transplantation for T and null cell CD30-positive anaplastic large cell lymphoma: analysis of 64 adult and paediatric cases reported to the European Group for Blood and Marrow Transplantation (EBMT). Bone Marrow Transplant. 1999;23(5):437–42.
55. Woessmann W, Peters C, Lenhard M, Burkhardt B, Sykora KW, Dilloo D, et al. Allogeneic haematopoietic stem cell transplantation in relapsed or refractory anaplastic large cell lymphoma of children and adolescents--a Berlin-Frankfurt-Munster group report. Br J Haematol. 2006;133(2):176–82.
56. Brugieres L, Quartier P, Le Deley MC, Pacquement H, Perel Y, Bergeron C, et al. Relapses of childhood anaplastic large-cell lymphoma: treatment results in a series of 41 children--a report from the French Society of Pediatric Oncology. Ann Oncol. 2000;11(1):53–8.
57. Strullu M, Thomas C, Le Deley MC, Chevance A, Kanold J, Bertrand Y, et al. Hematopoietic stem cell transplantation in relapsed ALK+ anaplastic large cell lymphoma in children and adolescents: a study on behalf of the SFCE and SFGM-TC. Bone Marrow Transplant. 2015;50(6):795–801.
58. Brugieres L, Pacquement H, Le Deley MC, Leverger G, Lutz P, Paillard C, et al. Single-drug vinblastine as salvage treatment for refractory or relapsed anaplastic large-cell lymphoma: a report from the French Society of Pediatric Oncology. J Clin Oncol. 2009;27(30):5056–61.
59. Fukano R, Mori T, Kobayashi R, Mitsui T, Fujita N, Iwasaki F, et al. Haematopoietic stem cell transplantation for relapsed or refractory anaplastic large cell lymphoma: a study of children and adolescents in Japan. Br J Haematol. 2015;168(4):557–63.
60. Pro B, Advani R, Brice P, Bartlett NL, Rosenblatt JD, Illidge T, et al. Brentuximab vedotin (SGN-35) in patients with relapsed or refractory systemic anaplastic large-cell lymphoma: results of a phase II study. J Clin Oncol. 2012;30(18):2190–6.
61. Pro B, Advani R, Brice P, Bartlett NL, Rosenblatt JD, Illidge T, et al. Five-year results of brentuximab vedotin in patients with relapsed or refractory systemic anaplastic large cell lymphoma. Blood. 2017;130(25):2709–17.
62. Mosse YP, Lim MS, Voss SD, Wilner K, Ruffner K, Laliberte J, et al. Safety and activity of crizotinib for paediatric patients with refractory solid tumours or anaplastic large-cell lymphoma: a Children's Oncology Group phase 1 consortium study. Lancet Oncol. 2013;14(6):472–80.
63. Mosse YP, Voss SD, Lim MS, Rolland D, Minard CG, Fox E, et al. Targeting ALK with Crizotinib in pediatric anaplastic large cell lymphoma and inflammatory Myofibroblastic tumor: a Children's Oncology Group Study. J Clin Oncol. 2017;35(28):3215–21.
64. Gambacorti-Passerini C, Mussolin L, Brugieres L. Abrupt relapse of ALK-positive lymphoma after discontinuation of Crizotinib. N Engl J Med. 2016;374(1):95–6.
65. Louissaint A Jr, Ackerman AM, Dias-Santagata D, Ferry JA, Hochberg EP, Huang MS, et al. Pediatric-type nodal follicular lymphoma: an indolent clonal proliferation in children and adults with high proliferation index and no BCL2 rearrangement. Blood. 2012;120(12):2395–404.
66. Lorsbach RB, Shay-Seymore D, Moore J, Banks PM, Hasserjian RP, Sandlund JT, et al. Clinicopathologic analysis of follicular lymphoma occurring in children. Blood. 2002;99(6):1959–64.

67. Attarbaschi A, Beishuizen A, Mann G, Rosolen A, Mori T, Uyttebroeck A, et al. Children and adolescents with follicular lymphoma have an excellent prognosis with either limited chemotherapy or with a "Watch and wait" strategy after complete resection. Ann Hematol. 2013;92(11):1537–41.
68. Schmidt J, Gong S, Marafioti T, Mankel B, Gonzalez-Farre B, Balague O, et al. Genome-wide analysis of pediatric-type follicular lymphoma reveals low genetic complexity and recurrent alterations of TNFRSF14 gene. Blood. 2016;128(8):1101–11.
69. Oschlies I, Salaverria I, Mahn F, Meinhardt A, Zimmermann M, Woessmann W, et al. Pediatric follicular lymphoma--a clinicopathological study of a population-based series of patients treated within the Non-Hodgkin's Lymphoma--Berlin-Frankfurt-Munster (NHL-BFM) multicenter trials. Haematologica. 2010;95(2):253–9.
70. Agrawal R, Wang J. Pediatric follicular lymphoma: a rare clinicopathologic entity. Arch Pathol Lab Med. 2009;133(1):142–6.
71. Claviez A, Meyer U, Dominick C, Beck JF, Rister M, Tiemann M. MALT lymphoma in children: a report from the NHL-BFM Study Group. Pediatr Blood Cancer. 2006;47(2):210–4.
72. Ronceray L, Abla O, Barzilai-Birenboim S, Bomken S, Chiang AK, Jazbec J, et al. Children and adolescents with marginal zone lymphoma have an excellent prognosis with limited chemotherapy or a watch-and-wait strategy after complete resection. Pediatr Blood Cancer. 2018;65(4). https://doi.org/10.1002/pbc.26932. Epub 2017 Dec 29.
73. Kobayashi R, Yamato K, Tanaka F, Takashima Y, Inada H, Kikuchi A, et al. Retrospective analysis of non-anaplastic peripheral T-cell lymphoma in pediatric patients in Japan. Pediatr Blood Cancer. 2010;54(2):212–5.
74. Windsor R, Stiller C, Webb D. Peripheral T-cell lymphoma in childhood: population-based experience in the United Kingdom over 20 years. Pediatr Blood Cancer. 2008;50(4):784–7.
75. Hutchison RE, Laver JH, Chang M, Muzzafar T, Desai S, Murphy S, et al. Non-anaplastic peripheral t-cell lymphoma in childhood and adolescence: a Children's Oncology Group study. Pediatr Blood Cancer. 2008;51(1):29–33.
76. Kontny U, Oschlies I, Woessmann W, Burkhardt B, Lisfeld J, Salzburg J, et al. Non-anaplastic peripheral T-cell lymphoma in children and adolescents--a retrospective analysis of the NHL-BFM study group. Br J Haematol. 2015;168(6):835–44.

Novel Therapies in Paediatric NHL

26

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Introduction

Current treatments for paediatric NHL result in excellent survival for most groups of children. Burkitt lymphoma (BL) and diffuse large B-cell lymphoma (DLBCL) constitute the major B-cell non-Hodgkin lymphoma (NHL) subtypes, and event-free survival (EFS) of >90% is expected from all major international front-line protocols [1–7]. Relatively short intensive chemotherapy schedules with low long-term but high acute toxicity are the norms. Reduction of acute toxicity is the clinical priority for the majority of children with B-NHL but to date there are no clinic-pathological or biological factors that predict which children may not benefit from therapy reduction.

T-lymphoblastic lymphoma (T-LBL) is the second most common paediatric NHL type behind the mature B-NHLs. T-LBL is treated with acute lymphoblastic leukaemia (ALL)-type therapy with expected event-free survivals exceeding 80% [8–11]. Stage is not a good predictor of outcome and biological risk classifications including molecular markers such as Notch and FBXW7 mutations as well as minimal marrow disease (MMD) and minimal residual disease (MRD) are still being explored.

In order to reduce therapy, either cure must be achieved with less of the conventional agents or new agents with more favourable toxicity profiles will need to be substituted for existing elements of successful regimens. Quite apart from the challenges of developing trials to show equivalence of efficacy of reduced therapy in a disease with a very high event-free survival (EFS) and limited patient numbers, a more urgent problem is the lack of effective treatments for relapsed disease. Relapsed and refractory disease has a very poor outcome with EFS of only ~30% for all B-NHL, only 15–20% for BL and 10–30% for T-LBL [6, 7, 12, 13]. There are only very rare survivors of a second relapse. There is therefore an unmet clinical need for treatments for relapsed paediatric NHL. Once successful salvage therapies are found, strategies to reduce the acute toxicity of first-line therapy can be addressed.

The third most common NHL type in childhood and adolescence, anaplastic large cell lymphoma (ALCL), accounts for about 10–15% of paediatric lymphomas [14]. Outcomes for ALCL hover around EFS of 70% with several different chemotherapy regimens ranging from less to more intensive [14]. Intensification has not shown any efficacy in ALCL. ALCL is defined by strong CD30 expression and recurrent translocation between chromosome 2 and 5 leading to the fusion protein NPM-ALK in greater 90% of paediatric cases [15]. Two targeted therapies (brentuximab vedotin (BV) and crizotinib) are currently being explored in upfront clinical trials.

Because further intensification of therapy is unlikely to lead to additional cures in the upfront setting and retrieval rates are poor with chemotherapy, novel and targeted therapies are needed to further advance the cure rates in childhood, adolescent and young adults (CAYA) NHL. In this chapter, we will consider the large number of novel agents available for treating NHL in adults that have potential application for children. We will consider them based on their mechanism of action and outline a rational approach to prioritization of agents for clinical development. Novel agents of interest in B-NHL and T-NHL are summarized in Tables 26.1 and 26.2.

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Table 26.1 Novel Agents in B-non-Hodgkin Lymphoma (NHL), N/A (Not applicable), refractory/relapsed (r/r), Diffuse large B-cell lymphoma (DLBCL), Rituximab (RTX), Cyclophosphamide, doxorubicin, vincristine, prednisone (CHOP), rituximab +CHOP (R-CHOP), Acute lymphoblastic leukaemia (ALL), Primary mediastinal B-cell lymphoma (PMBCL), Burkitt lymphoma (BL), monoclonal antibody (mAb)

Drug	Mechanism of action	Preclinical data	Efficacy in adult NHL	Safety and toxicity in paediatrics	Paediatric experience in B-NHL
Chemotherapy agents					
Bendamustine	Alkylating agent	N/A	r/r adult low-grade NHL and DLBL in combination with RTX	Acceptable (Cytopenias, pyrexia, nausea/vomiting, diarrhoea)	Phase I/II study in r/r leukaemia (Fraser et al. [16]) Phase I study in combination with clofarabine and etoposide (Jeha et al. [17]
Lenalidomide	Thalidomide derivative with immunomodulatory activity	N/A	Adult follicular lymphoma and DLBCL in combination with RTX or R-CHOP	Well tolerated as single agent	Phase I study as single agent in r/r solid tumours [18]
Bortezomib	Proteasome inhibitor	Able to overcome multi-drug resistance in leukaemia cell lines [19] 100% activity in T-ALL xenografts in preclinical testing programme [20]	Little activity in adult B-NHL	Tolerated in combination with ALL-type induction chemotherapy	Phase II study in combination with ALL-type chemotherapy in relapsed B-ALL [21]
Small-molecule inhibitors					
Ibrutinib	BTK inhibitor	Activity in BL Mouse xenograft model [22]	Activity in adult ABC DLBCL	Ongoing (NCT02703272)	Ongoing (NCT02703272)
Idelalisib	PI3K inhibitor	In vitro activity in RTX and chemotherapy resistant BL cell lines [23]	Activity in adult indolent lymphoma as single agent or in combination with rituximab	None	None
Venetoclax	BCL2 inhibitor	Broad activity in NHL cell lines [24]	Single-agent activity in refractory CLL	Ongoing (NCT03236857)	Ongoing (NCT03236857)
Immunotherapy					
Ipilimumab Pembrolizumab Nivolumab	Checkpoint inhibitors	NA	Activity in adult HL and T-NK lymphoma	Tolerated in phase I study of ipilimumab in paediatric patients with solid tumours [25]	Ongoing paediatric trials
Rituximab	mAB against CD20	NA	Extensive experience in adult B-NHL	Well tolerated with intensive chemotherapy	Activity in paediatric BL, DLBCL and PMBCL International Phase III trial in combination with intensive chemotherapy showed almost 100% survival in paediatric BL and DLBCL [3]
Inotuzumab ozogamicin	Humanized mAB against CD22 conjugated to calicheamicin	Activity in B-NHL xenograft models [26]	Activity in relapsed DLBCL	Tolerated as single agent, sinusoidal obstruction syndrome	68% CR rate in paediatric patients with r/r ALL [27], no experience in paediatric NHL
Blinatumomab	Bispecific T-cell engager (CD3/CD19)	NA	Activity in adult r/r DLBCL	Well tolerated, B-cell aplasia	Approved for the treatment of paediatric patients with relapsed ALL, no experience in paediatric NHL

Table 26.2 Novel Agents in T-non-Hodgkin lymphoma (NHL). Acute lymphoblastic leukaemia (ALL), lymphoblastic lymphoma (LBL), tuberous sclerosis (TS), subependymal giant cell astrocytomas (SEGA), primary mediastinal B-cell lymphoma (PMBCL), refractory/relapsed (r/r), monoclonal antibody (mAb)

Drug	Mechanism of action	Preclinical data	Efficacy in adult NHL	Safety and toxicity	Paediatric experience
T-LBL					
Chemotherapeutic agents					
Bortezomib	Proteasome inhibitor	Able to overcome multi-drug resistance in leukaemia cell lines [19] 100% activity in T-ALL xenografts in preclinical testing programme [20]	NA	Tolerated in combination with ALL-type induction chemotherapy	Phase II study in combination with ALL-type chemotherapy in relapsed B-ALL [21] Phase III study for newly diagnosed paediatric patients with T-ALL or T-LBL (NCT02112916)
Nelarabine	Purine nucleoside analogue	Toxic at micromolar concentration in T-lymphoblasts [28]	NA	Well tolerated in combination with ALL-type chemotherapy [29]	Phase III study for newly diagnosed patients with T-ALL and T-LBL [8]
Small-molecule inhibitors					
Everolimus	mTOR inhibitors	Synergistic activity of sirolimus in mouse childhood tumour xenograft models in combination with several chemotherapeutic drugs [30]	Activity as single agent or in combination with panobinostat in relapsed lymphoma: in combination with rituximab in DLBCL	Well tolerated with ALL-type reinduction	Approved for the treatment of paediatric patients with TS and SEGA Phase I study of everolimus in combination with multi-agent chemotherapy in patients with relapsed ALL [31]
Decitabine Vorinostat Panobinostat	Epigenetic modifiers	Synergistic effects of HDACs and decitabine [32]	Activity in B-NHL [33], promising results if used in combination with standard chemotherapy regimens [34]	Decitabine dose-limiting toxicities as single agent	Phase I trial for r/r NHL with panobinostat (NCT01321346), vorinostat used in autologous HSCT regimens [35]
Tazemetostat	Epigenetic modifier: EZH2 inhibitors	Inhibits proliferation of EZH2 mutant DLBCL cell lines and mice xenografts [36]	Activity in adult B-NHL	Tolerable side effects in adult phase Phase I study [37]	Ongoing paediatric trial (NCT03155620)
Palbociclib Ribociclib Abemaciclib	CDK 4/6 inhibitors	Anti-proliferative effects in multiple cell lines including mantle cell lymphoma, antitumour activity xenografts [38–40]	Activity in adult phase Phase II trial in r/r mantle cell lymphoma [41]. Ongoing trials (NCT01739309, NCT00141297)	Tolerable side effects in adult phase Phase II study [41]	Ongoing paediatric trial (NCT02693535)
Ruxolitinib	JAK inhibitor	Activity in mouse xenograft models of PMBCL [42] Able to overcome glucocorticoid resistance in T-ALL [43]	Ongoing adult studies in PTCL and T/NK lymphoma (NCT02974647) and r/r lymphoma (NCT02613598)	Well tolerated as single agent in paediatric phase Phase I study [44]	Phase I/II study in paediatric ALL and LBL (NCT03117751)
Immunotherapy					
Ipilimumab Pembrolizumab Nivolumab	Checkpoint inhibitors	NA	Extensive experience in adult solid tumours, HL and T-NK lymphoma	Tolerated in phase Phase I study of ipilimumab in paediatric patients with solid tumours [25]	Ongoing paediatric trials
Daratumumab	mAB against CD38	Activity in T-ALL mouse xenograft models [45]	Activity in multiple myeloma	Ongoing (NCT03384654)	Ongoing (NCT03384654)

(continued)

Table 26.2 (continued)

Drug	Mechanism of action	Preclinical data	Efficacy in adult NHL	Safety and toxicity	Paediatric experience
ALCL					
Small-molecule inhibitor					
Crizotinib	Alk inhibitor	NA	Extensive experience in Alk-driven non-small cell lung cancer	Well tolerated as single agent	7/8 CR in relapsed ALCL [46] Ongoing Phase III trial in combination with chemotherapy in newly diagnosed ALCL (NCT01979536)
Immunotherapy					
Brentuximab vedotin	Conjugated mAb against CD30	NA	Phase I study with ORR in 50% in relapsed ALCL and HL [47]	Acceptable toxicity in combination with gemcitabine [48]	Ongoing Phase III trial in combination with chemotherapy in newly diagnosed ALCL (NCT01979536)

Chemotherapy Agents

Few new cytotoxic chemotherapy agents have been investigated for the treatment of paediatric B-NHL over the last few decades. The increase in survival for paediatric B-NHL in the 1980s–2010 has been achieved internationally by adjustments to dosing and scheduling of conventional chemotherapy agents. The recent introduction of rituximab into high-risk paediatric B-NHL schedules will be discussed below. Innovations such as liposomal cytarabine, whilst in theory promising to deliver some advantages, for example, in CNS disease in B-NHL, have not been exploited and its place in the management of childhood B-NHL remains unclear [49].

New Agents for Indolent (Low Grade) Adult B-NHL with no Clear Role in Paediatric NHL

Several new agents have been investigated in adult B-NHL that have found use in indolent B-NHL (including follicular lymphoma and marginal zone lymphoma). Of these, bendamustine, lenalidomide and bortezomib have found established roles. Their efficacy in adult aggressive lymphoma has received some investigation but their place in the management of paediatric B-NHL remains unexplored.

Bendamustine

Bendamustine (a nitrogen mustard alkylating agent) has been shown to be effective in the treatment of relapsed/refractory low-grade NHL combined with rituximab [50]. The same combination has also been shown to be effective treatment for chemotherapy-naïve patients [51] and non-inferior when randomized against standard chemotherapy in front-line treatment of patients with low-grade NHL [52].

In the treatment of DLBCL, bendamustine and rituximab (BR) has applicability in the context of elderly patients with relapsed and refractory disease who do not tolerate more intensive therapy and in those previously untreated for whom more aggressive therapy is not appropriate. Vacirca et al. observed a 45.8% overall response rate (ORR) with complete response (CR) rate of 15.3% with bendamustine and rituximab. The median progression-free survival (PFS) was 3.6 months [53]. Ohmachi et al. demonstrated an ORR of 62.7% and PFS of 6.7 months in relapsed and refractory patients with DLBCL who were ineligible for or who had undergone autologous stem cell transplantation in a multicentre Phase II study. They concluded that BR represented a promising combination in this setting [54]. The Phase II study of Park et al. in elderly patients (median age 74 years) with previously untreated DLBCL showed a response rate of 78% with CR rate of 52%. Survival was however low with PFS of only 5.4 months [55].

Bendamustine has received limited study in paediatric patients. Fraser et al. in a Phase I/II study of relapsed or refractory leukaemia observed some activity of bendamustine monotherapy in patients with acute lymphoblastic leukaemia with a biological activity rate (CR + CR without platelet recovery+ partial response (PR)) of 9.3% [16]. A recent study of bendamustine hydrochloride, clofarabine and etoposide in treating younger patients with relapsed or refractory hematologic malignancies included NHL as an eligible disease [17]. Of 16 patients including two T-NHL, there were 6 CRs, 1 CR without platelet recovery and 3 PRs.

Although there may be some rationale for the study of bendamustine in paediatric B-NHL, this alkylating agent is not likely to receive prioritization in the presence of the other agents discussed in this chapter. Its performance in the relapsed/refractory setting should however be followed with interest.

Lenalidomide

Lenalidomide (a thalidomide derivative with immunomodulatory activity) combined with rituximab has been shown to be superior to lenalidomide alone with similar toxicity in a Phase II randomized study of recurrent follicular lymphoma [56]. Based on this finding, a further Phase II study of lenalidomide and rituximab in untreated follicular lymphoma showed that the combination was highly effective with ORR of 95% and 5-year overall survival (OS) of 100% [57].

In DLBCL, lenalidomide has been shown to have single-agent activity in relapsed and refractory disease in a Phase II single-arm multicentre trial (ORR 19% for DLBCL) [58]. In a larger international Phase II setting, an ORR of 28% was observed for DLBCL [59]. In a study of lenalidomide compared with investigator choice for relapsed DLBCL patients who had experienced ≥2 prior relapses or were ineligible for autologous stem cell transplantation or further combination chemotherapy, lenalidomide showed superior activity compared with investigator choice in the non-germinal centre B-cell like (GCB) subtype of DLBCL [60]. Nowakowski et al. have shown that lenalidomide added to rituximab, cyclophosphamide, doxorubicin, vincristine and prednisone (R-CHOP) overcomes the impact of non-GCB subtype in newly diagnosed patients with DLBCL [61]. In contrast to the historical control group where non-GCB subtype conferred a poorer outcome, there was no difference in 24-month PFS or OS for R-CHOP+ lenalidomide (R^2CHOP) patients on the basis of non-GCB and GCB subtype (60% vs. 59% [$P = 0.83$] and 83% vs. 75% [$P = 0.61$] at 2 years, respectively [61]. A further randomized, double-blind, placebo-controlled, global, Phase III study evaluating the efficacy and safety of R^2CHOP versus placebo-R-CHOP in patients with previously untreated activated B-cell (ABC)-type DLBCL–ROBUST [62] (NCT02285062) is ongoing. Due to the immunomodulatory effects of lenalidomide, there may be a role in combination with checkpoint inhibitors in lymphoma [18, 63].

In the paediatric setting, lenalidomide has been investigated as a single agent in relapsed and refractory solid tumours and myelodysplasia. In this Phase I study, no patients with NHL were recruited and only a single patient with Hodgkin lymphoma (HL) [18]. Lenalidomide at a dose of 70 mg/m^2/day for 21 days was well tolerated with drug clearance being faster in children under the age of 12. Cellular immunity was significantly upregulated. No objective responses were observed. There is an ongoing Phase II study for paediatric patients with relapsed or refractory (R/R) acute myeloid leukaemia (NCT02538965).

There would appear to be little in the literature to support the further investigation of lenalidomide in paediatric B-NHL other than in combination with other novel agents. However, combination therapy is itself complex and the excessive toxicity seen in the adult setting of follicular, and mantle cell lymphoma to the combination of lenalidomide, idelalisib and rituximab is a cautionary lesson [64].

New Agents Active in Adult Indolent and Aggressive Lymphoma with Known or Possible Paediatric NHL Application

In this section, we will discuss several novel agents based on the mechanism of action. In each case, the first or major drug in class will be discussed. Evidence in adult NHL (especially DLBCL) will be presented and any published data for paediatric NHL or known ongoing clinical trials.

Bortezomib

Bortezomib is a proteasome inhibitor that leads to apoptosis through increasing the inhibitory IκB [65] in relapsed adult B-NHL [66, 67]. In relapsed indolent B-NHL, it is more active combined with rituximab than rituximab alone [68], especially in the subgroup of high-risk follicular lymphoma [69]. Combination of bortezomib with bendamustine and rituximab has been shown to be active in R/R follicular lymphoma as well as in combination with rituximab, cyclophosphamide, vincristine and prednisone (RCVP) [70].

The study of Phase II study by Goy et al. of bortezomib in relapsed and refractory B-NHL included 12 patients with DLBCL among whom there was one PR [67]. The unmet clinical needs in the treatment of adult DLBCL are relapsed disease and front-line treatment of DLBCL where the proposed cell of origin is not from the germinal B-cell. Combination of bortezomib with gemcitabine was not shown to be effective for adults with R/R DLBCL in the Phase I/II study of Evens et al. [71]. Furthermore, neither of two randomized Phase II trial comparing R-CHOP with bortezomib substituting for vincristine in the experimental arm showed any benefit for patients with non-GCB DLBCL receiving front-line therapy [72, 73] [64, 65]. In Phase II study in adults with peripheral T-cell lymphoma (PTCL), 30 out of 46 patients (65%) achieved a CR with the combination of bortezomib and a CHOP chemotherapy backbone [74].

To date, no trials of bortezomib in paediatric NHL have been published. Two Phase I studies of bortezomib in R/R solid tumours including lymphoma have been reported, but neither recruited any child with NHL nor showed any objective responses to single-agent bortezomib [75] or combined with the histone deacetylation agent vorinostat [76]. Despite these disappointing early-phase studies, there is considerable enthusiasm for bortezomib in combination therapy. Preclinical data shows that bortezomib is able to overcome resistance to several chemotherapy agents including anthracyclines, alkylators and corticosteroids [77–81]. In the setting of relapsed paediatric acute lymphoblastic leukaemia,

patients with B-precursor ALL who relapsed after 2–3 prior lines of therapy had an 80% response rate to bortezomib combined with vincristine, dexamethasone, pegylated asparaginase and doxorubicin [21]. There is even more interest in bortezomib for T-lymphoblastic lymphoma/leukaemia because the majority are driven through Notch-1 or PI3K/AKT/MTOR pathway signalling and either of those pathways activates NFκB [12]. The current front-line Children's Oncology Group Phase III study for T-ALL and T-LBL is exploring the role of bortezomib in combination with ALL-type chemotherapy (NCT02112916).

However, the relevance of bortezomib to paediatric B-NHL is not easily extrapolated from the available literature. Whilst there is a significant lack of preclinical data in paediatric NHL, a recent report of synergism between bortezomib and an inhibitor of the mitochondrial protein second mitochondria-derived activator of caspase (Smac) in B-NHL cell lines leading to cell death by necroptosis even when apoptosis is inhibited suggests that bortezomib may have a role worth further investigation [82].

Nelarabine

Nelarabine (2-amino-9-B-D-arabinofuranosyl-6-methoxy-9H-purine) is a synthetic deoxyguanosine derivative that is cytotoxic to T-lymphoblasts at micromolar concentrations [83]. A Phase I study of nelarabine in children and adults with refractory haematological malignancies showed an overall response rate of 34%, but in the group of T-ALL/LBL, 9 CRs (23%) and 12 PRs (31%) were reported in the 39 patients [84]. Neurological toxicity was reported in 72% patients including transient somnolence, malaise, fatigue and peripheral motor and sensory neuropathies. A subsequent Phase II study of single-agent nelarabine in refractory T-cell malignancies showed response rates over greater 50% [85]. Subsequent studies showed that nelarabine was well tolerated in combination with intensive chemotherapy [86, 87]. The results of the Children's Oncology Group study AALL0434 for paediatric, adolescent and young adult patients with T-ALL/LBL were presented at the 2018 American Society of Oncology Meeting [8]. The addition of nelarabine significantly improved 4-year disease-free survival (DFS) (88.9 +/− 2.2% versus 83.3 +/− 2.5% (p = 0.0332)) in the T-ALL cohort but not in the high-risk T-LBL cohort with 4-year DFS of 85.0 +/− 5.6% versus 89.0 +/− 4.7% for nelarabine (N = 60) versus no nelarabine (N = 58), p = 0.2788.

Small-Molecule and Pathway Inhibitors

Inhibitors of Bruton's tyrosine kinase (BTK), phosphatidylinositol-4,5-bisphosphate 3-kinase (PI3K) and B-cell CLL/lymphoma 2 (BCL2) have shown activity in adult B-NHL, and all three classes have development in paediatric B-NHL. Because of the upregulation of the Notch and PI3K/mTOR pathways in T-LBL, mTOR and Notch inhibitors are being explored in the treatment of T-LBL. Other agents of interest include epigenetic modulators, CDK4/6 inhibitors, Jak inhibitors and BCL-2 inhibitors. In ALCL, Alk inhibitors are of obvious interest because greater 90% of paediatric and adolescent ALCL are NPM-ALK positive [14].

BTK Inhibitor: Ibrutinib

This first-in-class BTK inhibitor received breakthrough designation by the US Food and Drug Administration in 2013 for the treatment of relapsed and refractory mantle cell lymphoma (MCL). Subsequently, other low-grade NHLs have been added to its indications. Wilson et al. observed single-agent activity of ibrutinib in patients with DLBCL who had an ABC phenotype by gene expression profiling (GEP) [88]. Younes et al. conducted a Phase Ib study of ibrutinib added to R-CHOP, and in all 18 patients who received the recommended Phase II dose, a response was seen. Among those for whom subtyping was conducted, five of seven (71%) with GCB subtype and two (100%) of those with non-GCB subtype had a complete response [89]. Multiple adult studies of ibrutinib including patients with DLBCL are ongoing (https://clinicaltrials.gov).

There is limited preclinical data to suggest that paediatric Burkitt lymphoma may be responsive to BTK inhibition [22]. There is also gene expression data to suggest that B-cell receptor signalling is particularly active in paediatric Burkitt lymphoma suggesting BTK inhibition is an approach for further investigation [90].

A Phase III international randomized open-label controlled study which consists of a pharmacokinetic Part 1 run-in and a randomized Part 2 is underway to assess the safety and efficacy of ibrutinib in paediatric and young adult participants with relapsed or refractory mature B-NHL. In the randomized part, patients receive either physician-choice background chemotherapy alone (rituximab, ifosfamide, carboplatin, etoposide and dexamethasone (RICE) or rituximab, vincristine, ifosfamide, carboplatin, idarubicin and dexamethasone (RVICI)) or combined with ibrutinib (NCT02703272).

PI3K Inhibitor: Idelalisib and Duvelisib

PI3K is a target that sits downstream of the B-cell receptor. Inhibition results in apoptotic cell death. In a Phase II open-label study of 125 patients, Gopal et al. showed activity of idelalisib in heavily pretreated patients with indolent lymphoma

(median 4 prior line of therapy). The overall response rate was 57% with median PFS of 11 months [91]. A more recent Phase II study of idelalisib combined with a spleen tyrosine kinase (SYK) inhibitor entospletinib in relapsed and refractory lymphoma including DLBCL was terminated due to toxicity seen as pneumonitis in 18% of patients with two fatalities [92]. Combination with other agents has been problematic in other settings. In an ongoing Phase III trial of idelalisib or placebo added to bendamustine and rituximab, the first interim result shows increased efficacy of the combination with idelalisib but increased infections and pneumonitis raising concerns [93]. As mentioned above, the combination of idelalisib with lenalidomide and rituximab was found to be too toxic in a Phase I study of relapsed and refractory NHL [64].

To date there have been no large-scale studies of idelalisib in DLBCL, and safety concerns remain which are particularly relevant to investigation of this drug in a paediatric setting although it is not possible to extrapolate safety data from the adult to the paediatric setting.

There is preclinical data to suggest that the PI3K/AKT pathway is important in cell survival in paediatric Burkitt lymphoma [90, 94], and thus from a mechanism of action point of view, this class of drug is worthy of consideration for study.

A Phase I study of duvelisib in patients with cutaneous (CTCL) or peripheral (PTCL) T-cell lymphoma showed overall response rates of 31.6% and 50%, respectively [95].

A Phase Ib trial evaluating idelalisib in children and adolescents with relapsed or refractory DLBCL or mediastinal B-cell lymphoma in combination with rituximab, ifosfamide, carboplatin and etoposide (RICE) is anticipated to start in May 2018 (NCT03349346).

BCL2 Inhibitor: Venetoclax

BCL2 is a pro-apoptotic molecule, and its overexpression is associated with resistance to chemotherapy in a number of settings. Venetoclax has the clinical advantage of less platelets effects over earlier molecules. In a key study by Souers et al., significant antitumour effect of a single dose of venetoclax (ABT199) was seen in three patients with refractory chronic lymphoid leukaemia (CLL) [24]. The Phase I first-in-human study of venetoclax in relapsed and refractory NHL showed an 18% response rate in patients with DLBCL with 12% achieving a CR.

In the paediatric setting, BCL2 inhibition is of interest in the setting of other paediatric tumours such as neuroblastoma and leukaemia based on preclinical data as well as lymphoma. As a result, the first Phase I study of venetoclax in relapsed or refractory paediatric malignancies including NHL is currently recruiting (NCT03236857), and the rationale and design of this study has been described by Place et al. [96].

mTOR Inhibitors

The PI3K/mTOR/AKT pathway is one of the crucial pathways in malignancy [97]. Mammalian target of rapamycin (mTOR) is a serine and threonine protein kinase that signals through two multi-protein complexes, mTORC1 and mTORC2 [98]. Two mTORC1/2 inhibitors everolimus and temsirolimus are currently FDA approved for several adults cancers. Everolimus is also FDA approved for the treatment of paediatric patients with tuberous sclerosis (TS) and subependymal giant cell astrocytomas (SEGA).

In a Phase II study of single-agent everolimus in adults with relapsed lymphoma, overall response rates of 44% (7/16) in PTCL (including two patients with ALCL), 30% (23/77) in B-NHL (30% for DLBCL, 32% for MCL) and 38% in follicular lymphoma [99, 100]. In a Phase I study of everolimus in combination with panobinostat in adults with r/r lymphoma, the ORR was 30% with better responses in patients with Hodgkin lymphoma (HL). Everolimus was also tested in combination with rituximab in heavily pretreated adult patients with DLBCL and an ORR of 38% was reported. In paediatric Phase I study of everolimus in combination with vincristine, prednisone, pegaspargase and doxorubicin in relapsed ALL, 86% achieved a second CR with 68% having minimal residual disease burden <0.01 at the end of reinduction [31].

Temsirolimus was tested in a Phase I COG protocol in combination with intensive chemotherapy in paediatric patients with second or greater relapse of ALL [101]. Whilst 7 out of 15 patients achieved a CR, the addition of temsirolimus to intensive chemotherapy resulted in excessive toxicity.

There are several accruing paediatric clinical protocols with mTOR inhibitors including two Phase I trials for paediatric patients with NHL. Everolimus is tested in combination with nelarabine, cyclophosphamide and etoposide in paediatric patients with R/R T-lymphoblastic leukaemia/lymphoma (NCT03328104). Another Phase I trial is testing the combination of temsirolimus with etoposide and cyclophosphamide in patients with relapsed ALL and NHL (NCT01614197). In addition, the COG paediatric Match trial includes a Phase II arm using the PI3K/mTOR inhibitor LY3023414 in patients with solid tumours, non-Hodgkin lymphoma or histiocytic disorders with TSC or PI3K/MTOR mutations (NCT03213678).

CDK4/6 Inhibitors: Abemaciclib, Palbociclib and Ribociclib

Cell cycle dysregulation is a hallmark of all cancer cells leading to uncontrolled cell proliferation. Cell cycle regulation is a highly controlled process, key components of

which are cyclin-dependent kinases (CDKs). Many malignancies overexpress CDKs making them an attractive therapeutic target [102, 103]. The first-generation CDK inhibitors were nonselective CDK inhibitors that blocked CDK4 but at the same time had significant off-target side effects [103, 104]. Second-generation CDK inhibitors were developed to target selectively the ATP-binding site of the CDK4–cyclin complex as well as CDK6 and showed antiproliferative effects in multiple cell lines including MCL and antitumour activity in human tumour xenografts [38–40]. Tested drugs include palbociclib, ribociclib and abemaciclib, which are FDA approved for the treatment of metastatic hormone receptor-positive breast cancer. CDK4/6 inhibitors have been studied in NHL patients, including MCL in adults. Leonhard et al. treated 17 patients with relapsed disease and found substantial reductions in positron emission tomography (PET)-based tumour metabolism and proliferation. Five patients achieved a progression-free survival time of more than 1 year with one complete and two partial responses [105]. In a Phase II trial, Wang et al. treated 124 patients with relapsed or refractory MCL and observed responses in 81% of patients with 40% achieving CR with a favourable safety profile [41]. CDK4/6 inhibitors are currently further studied in adult NHL patients including patients with MCL (NCT01739309), advanced NHL (NCT00141297) or in the context of molecular profiling studies (NCT03297606). The paediatric Target a Specific Abnormality in a Tumour Gene in People with Advanced Stage Cancer (TAPUR) study also includes an experimental group with palbociclib (NCT02693535).

JAK Inhibitor: Ruxolitinib

Ruxolitinib is a potent and selective ATP-competitive inhibitor of JAK1 and JAK2 kinases. It is used in the treatment of myeloproliferative disease harbouring the JAK2 V617F mutation. Preclinical murine xenograft models have shown activity in Ph-like ALL, HL and PMBCL [42, 106]. Moreover, JAK/STAT pathway inhibition is able to overcome intrinsic glucocorticoid resistance in T-ALL including ETP [43]. In a Phase I trial of paediatric patients with relapsed solid tumours, leukaemia and lymphoma, ruxolitinib was well tolerated [44]. Ruxolitinib is currently being studied as single agent in a Phase II study of adult patients with PTCL and natural killer cell (NK) lymphoma (NCT02974647) and in combination with bortezomib in a Phase I study of R/R lymphoma (NCT02613598). In paediatric patients, the St. Jude's Research Hospital is offering a randomized Phase II/III study for patients with acute lymphoblastic leukaemia/lymphoma (NCT03117751).

PIM Inhibitors

Proviral insertion in murine lymphoma (Pim) family proteins consist of three kinase isoforms Pim-1, Pim-2 and Pim3 and play a role in cell cycle progression, cell survival and tumourigenesis [107]. Pim kinases are essential for hematopoietic lineage cell development [108], and Pim1 has been implicated in chemotherapy resistance in DLBCL [109]. Pim1/Pim2 mRNAs are highly expressed in DLBCL and MCL, and PIM2 is also overexpressed in follicular lymphoma, MALT lymphoma and nodal marginal zone lymphoma [110, 111] and correlate with poor prognosis [107, 112, 113]. Pim kinases are of particular interest because of a potential role as co-regulators of c-MYC-dependent oncogenesis as Pim1 and c-Myc show cooperation during lymphomagenesis in mouse models [114]. SGI-1776, a compound with activity against PIM1, PIM2 and PIM3, has recently been tested in a Phase I trial in adults; however, the trial had to be discontinued due to significant cardiotoxicity (NCT00848601). Novel inhibitors are being currently studied in preclinical settings [115].

Epigenetic Modulators: Decitabine, Vorinostat and Panobinostat

Epigenetic regulation allows for modulating gene expression profiles without modifying the primary sequence of DNA. A complex network of enzymes regulates the post-translational modifications of chromatin or histones utilizing histone methylation, acetylation, ubiquitination and phosphorylation as well as methylation of CpG islands on DNA [116]. Epigenetic modification has been linked to increased expression of essential proteins linked to the development of tumour metastasis and suppression of tumour suppressor genes [116–118]. Epigenetic modifications are executed by specific cellular enzymes including DNA methyltransferases (DNMT) for DNA methylation, histone acetyltransferases (HAT)/histone deacetylases (HDAC) and histone methyltransferases (HMT)/histone demethylases for determining the status of histone acetylation and methylation, respectively [116]. The first drug modifying epigenetic pathways was 5-azacytidine, which targets DNMTs, as well as its deoxy derivative decitabine. In a Phase I trial, 20 patients with CLL and 4 patients with NHL were treated with decitabine. Due to dose-limiting myelosuppression and infectious complications decitabine doses could not be escalated to levels associated with changes in global methylation or gene re-expression [119]. Based on preclinical data showing synergistic effects between HDACs and decitabine [32], trials combining both drugs in adult patients with R/R NHL are currently being carried out (NCT00275080) as well as in combination with other chemotherapeutic

agents (NCT00109824, NCT02846935, NCT03236857). Another class of FDA-approved inhibitor targets HDACs and includes vorinostat and panobinostat. In a Phase II trial, 18 patients with R/R DLBCL were treated with vorinostat with 1 patient achieving a complete response and 1 with stable disease [120]. In a more recent Phase I/II study, vorinostat was incorporated into a rituximab, cyclophosphamide, etoposide and prednisone backbone for the treatment of adult patients with R/R DLBCL, and a 35% CR rate was observed suggesting that combination of standard chemotherapeutic drugs and epigenetic regulators may lead to beneficial outcomes [34]. Vorinostat has been used in children in the setting of autologous hematopoietic stem cell transplantation. Nieto et al. showed that adding vorinostat to a high-dose gemcitabine, busulfan and melphalan regimen for refractory lymphomas let to an event-free and overall survival of 62% and 73%, respectively, at a median follow-up of 25 months [35]. Panobinostat has been studied in a Phase II trial in 40 adult patients with R/R DLBCL with a 28% response rate and a median duration of response of 14.5 months. A Phase I trial is studying the effect of panobinostat in paediatric patients with R/R NHL (NCT01321346).

Epigenetic Modulators: EZH2 Inhibitors—Tazemetostat

Histone modifications including histone acetylation and removal of acetyl groups play an essential role in normal cell development. Enhancer of Zeste Homolog 2 (EZH2) catalyses trimethylation of histone 3 lysine 27 (H3K27), a mark of transcriptional repression, and has been implicated in tumourigenesis either by direct gene overexpression or point mutations [121]. For example, gain-of-function (GOF) mutations in EZH2 are found frequently in follicular lymphoma and GC-DLBCL [122, 123]. A small-molecule inhibitor of EZH2 methyltransferase activity has been shown to effectively inhibit the proliferation of EZH2 mutant DLBCL cell lines and markedly inhibit the growth of EZH2 mutant DLBCL xenografts in mice [36]. Multiple EZH2-directed inhibitors have been developed including Tazemetostat, CPI-1205 and EZH1/2 inhibitor DS-3201. In a first-in-man Phase I study, Italiano et al. showed in 64 patients, 21 with B-cell NHL and 43 with solid tumours, that Tazemetostat has tolerable side effects and an overall response rate in lymphoma patients of 38% with 3 complete responses was observed. Interestingly, responses were achieved irrespective of the EZH2 mutational status; however, 1 patient with EZH2 mutation achieved a durable response for 16 months [37]. A Phase II study is currently being conducted to determine clinical activity in adult patients with DLCBL (NCT03456726) as well as in children (NCT03155620).

Crizotinib

Crizotinib is an oral tyrosine kinase inhibitor (TKI) that targets ALK and MET and was developed for the treatment of ALK-driven non-small cell lung cancer [124]. Additional ALK inhibitors currently in clinical trials and resistance patterns are reviewed by Li et al. [125]. ALK-positive ALCL is characterized by ALK fusion genes. The most common translocation, t(2;5) (p23;q35), fuses the promoter and proximal part of nucleophosmin (NPM) gene on chromosome 5 to the ALK gene on chromosome 2 and is found in >80% of cases [126]. Crizotinib as single agent has shown impressive activity in ALCL with 7/8 relapsed paediatric ALCL patients achieving a CR in a Phase I trial [46]. Relapse in ALCL appears rare whilst on active therapy with crizotinib. Adequate length of therapy is not known [14]. Crizotinib as single agent is well tolerated in the paediatric age group without any development of resistance in the ALCL group [46]. In the relapse setting, it is being used as induction therapy to achieve CR prior to transplant. Crizotinib is also being evaluated as first-line therapy in combination with the ALCL99 backbone by the Children Oncology Group (NCT01979536), and the EICNHL is also planning a trial to evaluate it as first-line therapy in combination with chemotherapy.

Immunotherapies

Immunotherapies either harness natural immune responses to attack cancer cells (checkpoint inhibitors) or are manufactured to elicit new immune responses (monoclonal antibodies, T-cell engager antibodies or chimeric antigen receptor T-cells). These have transformed the landscape in many adult cancers, and their potential for similar impact in childhood cancer remains largely unexplored beyond acute leukaemia. A comprehensive overview of the topic can be found in the review by Majzner et al. [127].

CheckPoint Inhibitors

The immune checkpoint regulatory proteins, cytotoxic T lymphocyte-associated 4 (CTLA-4) and programmed cell death protein 1 (PD-1), are receptors expressed on the surface of cytotoxic T-cells that interact with their ligands cluster of differentiation 80/86 (CD80/86) and programmed death ligand-1 (PDL-1), respectively, on antigen-presenting cells. They are crucial in the maintenance of self-tolerance and prevention against autoimmunity [128]. However, these mechanisms can be exploited by cancer cells. Overexpression of PDL-1 and CD80/86 on tumour cells can lead to T-cell exhaustion enabling cancer cells to evade T-cell-mediated

death [129, 130]. Inhibitors of CTLA-4, PD-1 and PDL-1 have led to significant responses in a variety of adult cancers including melanoma, bladder carcinoma and non-small cell lung cancer [127]. In addition, checkpoint inhibitors have shown activity in heavily pretreated adults with R/R HL [131]. Autoimmune-like syndromes occur in a significant proportion of adults receiving checkpoint inhibitors, most commonly rashes and colitis but immune-related endocrinopathies, haematological and neurological toxicities have also been reported [132]. There is limited experience of checkpoint inhibitors in paediatric patients.

CTLA4 Inhibitor: Ipilimumab

A Phase I study of Ipilimumab in 18 patients with R/R NHL (including three with DLBCL) showed that the drug was well tolerated. There were clinical responses in two patients (one with DLBCL) [133]. As a therapy following relapse from stem cell transplantation, single-agent Ipilimumab did not give any responses in the context on NHL [134]. However, combination with lenalidomide was well tolerated in another study, and there were some durable responses [63].

In the paediatric setting, there has only been one published Phase I study of Ipilimumab in progressive solid tumours (including 12/33 patients with melanoma but no patients with NHL). No objective responses were seen [25]. There are several studies in adult patients with DLBCL using tremelimumab, another CTLA-4 inhibitor [135, 136], but no experience in paediatric patients or in lymphoma has been reported as of yet.

PD-1 Inhibitors: Pembrolizumab and Nivolumab

Primary mediastinal large B-cell lymphoma (PMLBL) is a diffuse large B-cell lymphoma occurring in adults (females>males) but also in children. Relapse of this disease is difficult to cure, and there is an unmet clinical need in this area. In addition for children and adolescents, first-line therapy has not yet been optimized [137]. PMLBL, like Hodgkin lymphoma, has frequent 9p24.1/*PD-L1/PD-L2* mutations making checkpoint inhibitor therapy an attractive prospect. A recent report from an ongoing Phase Ib trial (NCT01953692) reported the result with the cohort of multiply relapsed PMLBL receiving pembrolizumab. A response was observed in 7 of 14 (41%) patients with 2 achieving CR [138]. A recently started Phase II trial of pembrolizumab in R/R grey-zone lymphoma (GZL), primary central nervous system lymphoma (PCNSL) and other extranodal DLBCL (NCT03255018) with a lower inclusion age of 14 years is due to complete in 2022.

NK/T-cell lymphoma has been shown to overexpress PDL-1, and serum PDL-1 levels have been associated with prognosis [139, 140]. In a pilot study of pembrolizumab in seven heavily pretreated refractory patients, five patients achieved a CR and the other two patients had a PR for an ORR of 100% [141]. In addition to NK/T-cell lymphoma, cutaneous T-cell lymphomas have also been shown to overexpress PDL-1 [142]. In a Phase II study, 24 patients with R/R mycosis fungoides and Sezary syndromes were treated with pembrolizumab monotherapy with an overall response rate of 38% (1 CR and 8PRs) [143].

A recent Phase Ib study of nivolumab in R/R haematological malignancy having a median of three prior therapies included 11 patients with DLBCL and 23 patients with R/R T-cell lymphoma. For the DLBCL cohort, an objective response rate of 36% was observed but the T-cell lymphoma cohort only had an overall response rate of 17% without any CRs [144].

The checkmate trials showed that the combination of nivolumab and ipilimumab is superior to nivolumab or ipilimumab as single agent in preclinical studies and solid tumour malignancies. The combination also showed efficacy in adults with lymphoma. ORR of 74%, 20% and 9% were reported in 65 adults with Hodgkin lymphoma [62], B-cell lymphoma [15] and T-cell lymphoma [11], respectively [145].

There are a number of clinical trials of nivolumab alone or in combination open to children with NHL at the time of writing (NCT02304458, NCT02581631, NCT02419417, NCT02813135), but there are no trials of nivolumab or any other checkpoint inhibitor exclusively in paediatric NHL or specifically PMBCL. The Children's Oncology Group in cooperation with one of the adult cooperative groups is currently developing an upfront Phase II trial of pembrolizumab in combination with chemotherapy for paediatric and adult patients with newly diagnosed PMBCL.

PDL-1 Inhibitors

There are several US Food and Drug Administration (FDA) and European Medicines Agency (EMA)-approved PDL-1 inhibitors: atezolizumab for the treatment of metastatic non-small cell lung cancer, durvalumab for the treatment of bladder cancer and avelumab for the treatment of metastatic Merkel cell carcinoma. As of yet, there is no paediatric experience with any PDL-1 inhibitors, but there are ongoing studies in adults with haematological malignancies [130].

Monoclonal Antibodies

CD20: Rituximab and Other B-Cell Antibodies

In 1997, rituximab was approved by the FDA and in 1998 by the EMA for the treatment of R/R indolent lymphoma. Since then, rituximab has transformed the treatment of all adult B-NHL [146]. The study of rituximab in children with lymphoma would wait more than two decades for academic and pharmaceutical interests to coincide aided by changes in regulations mandating drug development in children. Pilot studies of rituximab in paediatric B-NHL demonstrated that it was possible to combine rituximab with existing chemotherapy regimens and suggested improved survival [147–149]. A window study conducted by the Berlin–Frankfurt–Munster (BFM) group demonstrated a meaningful response rate to a single dose of rituximab given before standard chemotherapy [150]. This has paved the way for an ongoing randomized study comparing standard chemotherapy with rituximab given either as a single dose or in repeated doses (NCT0320667). The recently completed Phase III international trial of rituximab added to conventional therapy demonstrated an almost 10% survival advantage for the group randomized to rituximab [3]. However, rituximab added to a prolonged infusional chemotherapy regimen (DA-EPOCH) reported to have outstanding activity in adult patients [151] but has not shown the same efficacy in paediatric and adolescent patients in a recently completed international Phase II study [137].

Rituximab was the first-in-class anti-CD20 monoclonal antibody, and although its use in paediatric lymphoma has only now been confirmed, there are a number of other anti-CD20 antibodies that have yet to be investigated in paediatric lymphoma. Of these, obinutuzumab (GA101), a type II, glycoengineered, humanized anti-CD20 monoclonal antibody, has shown activity patients with rituximab-refractory DLBCL [152]. However, it has not shown superiority over rituximab when combined with CHOP in front-line therapy of DLBCL and is associated with increased toxicity [153]. Nonetheless, there is a current study of the safety of administering obinutuzumab as a single agent alone and in combination with ifosfamide, carboplatin and etoposide (ICE) chemotherapy to determine the response rate of this treatment for children, adolescents and young adults (CAYA) with relapsed CD20 B-NHL (NCT02393157). There is no data relating to other anti-CD20 antibodies (ofatumumab, veltuzumab or ocrelizumab) in paediatric B-NHL, and it is unlikely that their utility will be investigated given the many other classes of novel agent available and the success of rituximab in front-line treatment of high-risk disease. A further challenge is the advent of biosimilar anti-CD20 agents which are approved in US and Europe making paediatric development of proprietary anti-CD20 antibodies unattractive to the pharmaceutical industry.

Antibodies directed against other targets relevant to paediatric B-NHL such as CD22 (epratuzumab [154]) and CD79a (polatuzumab vedotin) have been shown to be effective in R/R adult DLBCL, but their role in paediatric disease remains investigated.

CD38: Daratumumab

Daratumumab, a human immunoglobulin G1κ monoclonal antibody against CD38, is FDA and EMA approved for the treatment of refractory multiple myeloma in combination therapy. It is generally well tolerated [155]. Physiologically, CD38 is highly expressed on plasma cells and to a lesser degree on NK cells and subpopulation of B- and T-cells but it is also found in a variety of non-haematopoietic tissues [156]. In patient-derived T-ALL mouse xenograft models, daratumumab showed efficacy in 14 out of 15 xenografts [45].

There is no clinical experience in lymphoblastic leukaemia/lymphoma yet, but an international Phase II study of daratumumab in addition to standard chemotherapy in paediatric, adolescent and young adult patients with R/R T- or B-lymphoblastic leukaemia/lymphoma has started accrual (NCT03384654). There are also adult case reports of responses to daratumumab in primary effusion lymphoma and NK/T-cell lymphoma [157, 158].

Bispecific T-cell Engager (BiTE) Antibodies

Blinatumomab

Blinatumomab is a bispecific CD19-directed CD3 T-cell engager indicated for adult and paediatric precursor B-cell acute lymphoblastic leukaemia in first or second remission with minimal residual disease (MRD) greater than 0.1%. It is also indicated for relapsed and refractory acute lymphoblastic leukaemia.

The adult Phase I study of Goebeler et al. [159] in relapsed and refractory NHL demonstrated no responses at a continuous infusion rate of less than 15 mcg/m^2/day over a 4–8-week period. There appeared to be a dose–response up to a maximum tolerated dose of 60 mcg/m^2/day with neurological toxicity being the dose-limiting toxicity. This was chosen as the target dose and among patients achieving this, there was a 55% ORR for patients with DLBCL with 4/11 patient obtaining CR/Cru. Single-agent blinatumomab therefore has

activity in adult patients with R/R DLBCL. A subsequent Phase II study in 25 patients with R/R DLBCL confirmed an ORR of 43% in 21 evaluable patients with CR in 19% (4/21). The target dose in this study was at 112 mcg/day for at least 1 week.

There have been no trials of blinatumomab in paediatric NHL to date. However, based on the promising data in adult DLBCL, investigation of this class of antibody is warranted.

Immunoconjugates

CD22: Inotuzumab Ozogamicin

More than 80–100% of adult (reviewed in [160]) and paediatric [161] mature B-cell malignancies express CD22 on the cell surface. Inotuzumab ozogamicin is a humanized immunoglobulin G4 anti-CD22 antibody (G544) with the cytotoxic antibiotic calicheamicin connected by an acetyl butyrate linker. Internalization of CD22 allows for the hydrolysis of the acetyl butyrate linker with intracellular release of calicheamicin. Calicheamicin causes site-specific double-stranded DNA cleavage [162].

A Phase I study of single-agent inotuzumab ozogamicin given once every 3 or 4 weeks showed that the MTD was 1.8 mg/m^2 with thrombocytopenia being a major toxicity. Of the patients with relapsed DLBCL, the ORR was 15% [163]. In combination with rituximab, a further Phase I/II study showed a 74% ORR for relapsed (but not refractory) DLBCL with 50% CR [164]. Combination with R-CVP has been shown to result in a 57% ORR in R/R aggressive lymphoma (including 17 patients with DLBCL and four with MCL) [165]. The retrospective review of 34 heavily pretreated paediatric patients with R/R ALL treated with 1–4 cycles of single-agent inotuzumab ozogamicin was presented at the American Society of Clinical Oncology in 2016 [27]. Of 29 patients with >5% blasts in the bone marrow, 18 (68%) achieved a CR. Inotuzumab ozogamicin was well tolerated in this heavily pretreated patient population. No sinusoidal obstruction syndrome (SOS) was seen during inotuzumab ozogamicin; however, 8 out of 15 patient who received a haematopoietic transplant post-Ino developed SOS. To date, there are no data and no studies of inotuzumab in paediatric B-NHL. The mechanism of action and responses in adult aggressive lymphoma suggest that this immunoconjugate is of interest for investigation in children.

CD30: Brentuximab Vedotin

CD30 is a member of the tumour necrosis factor (TNF) receptor superfamily. Its physiologic expression is restricted to eosinophils and activated T- and B-lymphocytes but is highly expressed in HL and anaplastic large cell lymphoma (ALCL) and variably expressed in paediatric Burkitt lymphoma and DLBCL [166] making it an attractive drug target [167, 168]. SGN-30, a chimeric monoclonal anti-CD30 antibody, showed promising preclinical activity, but in a Phase II study in patients with R/R HL and ALCL, there were no objective responses in the HL cohort and only a 17% objective response rate in the ALCL cohort [169]. To improve activity, a conjugated antibody, brentuximab vedotin (BV), was developed. BV is conjugated to four monomethyl auristatin E molecules. Upon binding to CD30, the conjugated antibody is endocytosed, and the auristatin derivatives are released and act as microtubulin inhibitors. In a Phase I study in adult patients with r/r CD30-positive lymphomas, the maximum tolerated dose (MTD) was reached at 1.8 mg/kg every 3 weeks, and objective responses were observed in 50% of patients at that dose level [47]. It received EMA and FDA approval for R/R HL and ALCL based on two single-arm trials showing ORR of 73% and 86% for HL and ALCL, respectively [170, 171]. The ALCL trials included adolescent patients above the age 14. Single-agent BV has also been explored in B-NHL. Objective responses of 44% were observed in a Phase II study of BV in adults with R/R DLBCL [172], but the ORR was only 13% without any CRs in a Phase II study in patients with R/R PMBCL [173]. BV has been used in combination with chemotherapy in adults with an acceptable toxicity profile with the exception of bleomycin or tubulin inhibitors because of overlapping toxicities [174–177].

The Children's Oncology Group reported the results of Phase I/II study of BV in combination with gemcitabine showing an acceptable toxicity profile and a CR rate of 67% in paediatric patients with R/R HL [48]. This has led to BV being explored in front-line paediatric trials in HL and ALCL in the COG. The current COG front-line study ANHL12P1 randomizes patients between brentuximab vedotin and crizotinib on an ALCL-99 chemotherapy backbone (NCT01979536). Results have not been released as of the writing of this chapter.

Cellular Therapy

Chimeric Antigen Receptor (CAR)-Modified T-cells

CAR T-cells are engineered to confer on the resultant T-cells a new, predefined specificity. In effect, they combine the targeting of a monoclonal antibody with the cellular immune cytotoxicity of a T-cell. The design and optimisation of CAR T-cells is beyond the scope of this chapter, but comprehensive reviews have been published [178, 179]. Whilst for a

variety of human cancers CAR T-cells have provided new promise, they have also brought new toxicities and challenges. Among these, cytokine release syndrome and neurotoxicity are most significant (reviewed in [180]).

CAR T-cells directed against the surface molecule CD19 have transformed the outlook for children and adults with relapsed or refractory acute B-lymphoblastic leukaemia. The recent report from a pivotal global Phase II trial of the Novartis product Tisagenlecleucel showed a response rate to a single infusion of cells of 81% at 3 months with all patients being negative for minimal residual disease by flow cytometry. Acute toxicity was high grade but manageable. Cytokine release syndrome occurred in 77% patients and 48% of these received the anti-interleukin 6 receptor monoclonal antibody tocilizumab [181].

In NHL, CAR T-cells have shown activity across a range of B-cell lymphomas including DLBCL, and durable responses have been observed (the development of CAR T-cell therapy in adult NHL is comprehensively reviewed by Brudno and Kochenderfer [182]. CAR T-cell therapy is T-cell lymphomas is more complicated because selective antigens have to be found that are expressed on blasts but not normal T-cells. CAR T-cells against CD30 have been developed by several groups, and clinical trials are ongoing [182]. Preliminary results show responses in R/R HL and ALCL including brentuximab-resistant disease. In addition, CAR T-cells for CD37, surface antigens expressed in B-cell malignancies and also some peripheral T-cell lymphomas, are in development, but there is no in vivo experience yet [77].

Table 26.3 shows current registered clinical trials of CAR T-cells that are open to patients with paediatric B-NHL.

Epstein–Barr Virus-Specific T-cells

The production of Epstein–Barr virus (EBV)-specific cytotoxic T-lymphocytes (CTL) has become standardized to develop a GMP-grade product [183]. Donor-derived EBV-CTLs have been used prophylactically in hematopoietic stem cell transplantation (HSCT) recipients with EBV viremia or therapeutically in HSCT-related post-transplant lymphoproliferative disease (PTLD). PTLD expresses type III latency EBV antigens including LMP-1 and 2 and EBNA 1, 2 and 3 making it a very immunogenic disease [184]. EBV-CTLs were effective in the prevention of PTLD in 101 patients with EBV viremia, and 24 out of 27 patients with established PTLD achieved a CR [183]. In solid organ transplant (SOT) recipients, PTLD usually arises from recipient cells; recipient and transplant organ are often not HLA matched; and donor-derived PBMCs are not available in cadaver transplants to produce EBV-CTLs making donor-derived EBV-CTLs a poor choice [185]. Autologous EBV-CTLs have been used but production is complicated by the ongoing immunosuppression of the SOT recipients and leads to delays in treatment [183]. They have been used though in immunocompetent HL and NHL patients. In immunocompetent hosts, EBV-positive lymphomas only express type II latency antigens making them less immunogenic, but this can potentially be overcome by targeting LMP-1 and 2 [186]. Of 21 heavily pretreated patients with R/R EBV-positive HL and NHL, 11 patients achieved a CR with another two patients having a PR with a 2-year EFS of 50% [186]. This cohort included five paediatric patients.

In addition, third-party EBV-CTLs cryobanks produced from healthy EBV seropositive donors and covering the most common HLA types are being developed to offer an 'off-the-self' product that is readily available [183, 187–190]. HLA matching at 1–2 loci between CTL product and recipient is sufficient as long as the EBV activity is transmitted through shared alleles. Banks of 30 donors with varied HLA types are sufficient to cover 80%–90% of the population [188, 191, 192]. A multicenter Phase II study of third-party EBV-CTLs in 33 SOT and HSCT recipients including 11 children reported an overall response rate of 52% [193]. Other groups have shown overall responses of 67%–80% [188, 189, 192]. The Children's Oncology Group is currently conducting a Phase II trial using third-party EBV-CTL in paediatric patients with PTLD after SOT (NCT02900976).

Tumour-Associated Antigen T-cells

There are several tumour-associated antigens (TAA) that are not widely expressed in healthy mature tissues including WT1, PRAME and surviving making them potentially attractive targets for cellular therapy [194]. Other potential targets include SSX2, MAGE-A4 and NY-ESO1. Preliminary results were presented by Leen et al. in 2015 and Williams et al. in 2017 at the American Society of Hematology Meeting showing tolerability with <1% product-related side effects and clinical responses in 50–75% of patients [195, 196]. Currently, open protocols for EBV-specific and TAA-cytotoxic T-cells are summarized in Table 26.4.

Rational New Drug Development in Paediatric NHL

The preceding sections describe an array of new therapies available with several being investigated in paediatric NHL. As mentioned in the introduction, the current unmet clinical needs are for salvage therapy for relapsed and refractory B- and T-NHL and reduction of acute toxicity [197]. With many drugs and few patients, there is a need for rational coordinated approaches to novel drug development in NHL. Burkitt lymphoma is the major B-NHL in children with

Table 26.3 CAR T trials relevant to paediatric B-NHL as at 13.4.18

NCT Number	Conditions	Interventions	Sponsor/location
NCT02772198[a]	Pre-B ALL, B-NHL	CD19 CART cells	Sheba Medical Center, China
NCT01853631	NHL, CLL, ALL	CD19 CART cells, Fludarabine, Cyclophosphamide	Baylor College of Medicine, USA
NCT00586391	B NHL, CLL, ALL	CD19CAR-28-zeta T cells, lpilimumab	Baylor College of Medicine, USA
NCT03391726	B-NHL	CART-19 cells	Fujian Medical University, China
NCT02813837	Pre-B ALL, B-NHL	CD19CART	Innovative Cellular Therapeutics Co., Ltd., China
NCT03098355[a]	Pre-B ALL, B-NHL	4SCAR19/22 T cells, Interleukin-2	Zhujiang Hospitall Shenzhen Geno-Immune Medical Institute, China
NCT03281551	Pre-B ALL, B-NHL	PZ01 CAR-T cells	Pinze Lifetechnology Co. Ltd., China
NCT03448393[a]	Pre-B ALL, B-NHL	CD19/CD22 CART-cells, Fludarabine, cyclophosphamide	National Cancer Institute (NCI), USA
NCT02737085	DLBCL	CD19 CAR-T cells; anti-CD20 CAR-T cells	Southwest Hospital, China
NCT02965092	Pre-B ALL, B-NHL	CD19 CAR-T cells	Wuhan Sian Medical Technology Co., Ltd, China
NCT01626495	Pre-B ALL, B-NHL	CART-19[a]	University of Pennsylvania, USA
NCT03118180	NHL	CD19 CAR T cells	Zhejiang University, China
NCT02963038	Pre-B ALL, B-NHL	CD19 CAR T cells	Hebei Senlang Biotechnology Inc., Ltd., China
NCT02728882	NHL	CD19 CAR-T cells	Sinobioway Cell Therapy Co., Ltd., China
NCT03068416	Pre-B ALL, B-NHL	CD19 CAR T cells	Uppsala University, Sweden
NCT02650414[a]	Pre-B ALL, B-NHL	CD22 CART cell transduced with a lentiviral vector to express anti-CD22 scFV TCRz:41BB	University of Pennsylvania, USA
NCT03366350	Pre-B ALL, B-NHL	CD19 CAR-T cells	Wuhan Sian Medical Technology Co., Ltd, China
NCT03366324	Pre-B ALL, B-NHL	CD19 CAR-T cells	Wuhan Sian Medical Technology Co., Ltd, China
NCT03383952	Pre-B ALL, B-NHL	CD19 CAR-T cells	Immune Cell, Inc.,, USA
NCT03398967	Pre-B ALL, B-NHL	Universal dual specificity CD19 and CD20 or CD22 CAR-T cells	Chinese PLA General Hospital, China
NCT03166878	Pre-B ALL, B-NHL	UCART019	Chinese PLA General Hospital, China
NCT02050347	Pre-B ALL, B-NHL, CLL	CD19, CAR-CD28Z T cells	Baylor College of Medicine, USA
NCT01593696[a]	Pre-B ALL, B-NHL	CD19- CART cells	National Cancer Institute (NCI), USA
NCT02431988	DLBCL	CAR19 T-cells, fludarabine, cyclophosphamide	University College, London, UK
NCT00840853	Pre-B ALL, B-NHL, CLL	CD19CAR/virus specific T cells	Baylor College of Medicine, USA

[a]Trials with age of enrolment entirely paediatric or less than 40 years and therefore most likely to recruit paediatric patients

DLBCL being less common making Burkitt lymphoma the predominant relapse disease also. Few of the drugs reviewed above have data in adult Burkitt, and there is very little pre-clinical paediatric data. T-lymphoblastic lymphoma is the predominant T-NHL in childhood. Whilst there are new agents with biological rationale for further exploration, there is little clinical evidence of their efficacy yet. Mechanism of action, toxicity and ability to combine with other agents should be considered in the prioritization of drugs for investigation. Relapse of Burkitt lymphoma is associated with very rapid death in non-responders to relapse therapy with median time to death of 2.5 months in the study of RICE reported by Griffin et al. [198]. Rapid progression and poor retrieval rates are similar in T-lymphoblastic lymphoma. The window of opportunity for salvage is therefore narrow, and new approaches should concentrate on those agents likely to result in rapid disease clearance. Global collaboration is likely to be required as relapses are rare. The ability to investigate more than one agent in an overarching platform study will be necessary if many agents are not going to be left behind.

Table 26.4 Active trials as of 19 September 2018 using Epstein–Barr virus (EBV)-specific or tumour-associated antigen (TAA) cytotoxic T-lymphocytes (CTL)

Clinicaltrials.gov ID	Title	Cellular therapy	Phase	Age	Lead organization
NCT01333046	T-Lymphocytes in treating patients with active or relapsed Hodgkin and non-Hodgkin lymphoma	TAA-specific CTL	I	18 and over	Baylor College of Medicine
NCT02578641	A Phase III trial evaluating chemotherapy and immunotherapy for advanced nasopharyngeal carcinoma (NPC) patients	EBV-specific CTL	III	21 and over	Tessa therapeutics
NCT00062868	Autologous or donor cytotoxic T-lymphocytes in treating patients with Epstein–Barr virus-positive hematologic malignancy	LMP-specific CTL	I/II	0 and over	Baylor College of Medicine
NCT00368082	Autologous or donor cytotoxic T-cells in treating patients with Epstein–Barr virus-positive lymphoma, lymphoepithelioma or lymphoproliferative disorder	TGF-beta-resistant LMP-specific CTL	I	0 and over	Baylor College of Medicine
NCT02239861	Tumour-associated antigen-specific T-cells in treating participants with solid tumours	Donor-derived WT1/PRAME/NY-ESO-1/survivin-specific T-lymphocytes	I	2 to 80	Baylor College of Medicine
NCT01555892	Modified T-cells in treating patients with Epstein–Barr virus-positive lymphoma	LMP/BARF1/EBNA1-specific CTL	I	0 and over	Baylor College of Medicine

References

1. Cairo MS, Gerrard M, Sposto R, Auperin A, Pinkerton CR, Michon J, et al. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. Blood. 2007;109(7):2736–43.
2. Gerrard M, Cairo MS, Weston C, Auperin A, Pinkerton R, Lambilliote A, et al. Excellent survival following two courses of COPAD chemotherapy in children and adolescents with resected localized B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study. Br J Haematol. 2008;141(6):840–7.
3. Minard-Colin V, Auperin A, Pillon M, Burke A, Anderson JR, Barkauskas DA, et al. Results of the randomized Intergroup trial Inter-B-NHL Ritux 2010 for children and adolescents with high-risk B-cell non-Hodgkin lymphoma (B-NHL) and mature acute leukemia (B-AL): Evaluation of rituximab (R) efficacy in addition to standard LMB chemotherapy (CT) regimen. J Clin Oncol. 2016;34(15_suppl):10507.
4. Patte C, Auperin A, Gerrard M, Michon J, Pinkerton R, Sposto R, et al. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. Blood. 2007;109(7):2773–80.
5. Woessmann W, Seidemann K, Mann G, Zimmermann M, Burkhardt B, Oschlies I, et al. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. Blood. 2005;105(3):948–58.
6. Anoop P, Sankpal S, Stiller C, Tewari S, Lancaster DL, Khabra K, et al. Outcome of childhood relapsed or refractory mature B-cell non-Hodgkin lymphoma and acute lymphoblastic leukemia. Leuk Lymphoma. 2012;53(10):1882–8.
7. Jourdain A, Auperin A, Minard-Colin V, Aladjidi N, Zsiros J, Coze C, et al. Outcome of and prognostic factors for relapse in children and adolescents with mature B-cell lymphoma and leukemia treated in three consecutive prospective "Lymphomes Malins B" protocols. A Societe Francaise des Cancers de l'Enfant study. Haematologica. 2015;100(6):810–7.
8. Dunsmore KP, Winter S, Devidas M, Wood BL, Esiashvili N, Eisenberg N, Briegel N, Hayashi RJ, Gastier-Foster JM, Carroll AJ, Heerema NA, Asselin B, Rabin KR, Zweidler-McKay P, Raetz EA, Loh ML, Winick NJ, Carroll WL, Hunger S. COG AALL0434: A randomized trial testing nelarabine in newly diagnosed T-cell malignancy. J Clin Oncol. 2018;36(15_Supplement):10500.
9. Termuhlen AM, Smith LM, Perkins SL, Lones M, Finlay JL, Weinstein H, et al. Disseminated lymphoblastic lymphoma in children and adolescents: results of the COG A5971 trial: a report from the Children's Oncology Group. Br J Haematol. 2013;162(6):792–801.
10. Landmann E, Burkhardt B, Zimmermann M, Meyer U, Woessmann W, Klapper W, et al. Results and conclusions of the European Intergroup EURO-LB02 trial in children and adolescents with lymphoblastic lymphoma. Haematologica. 2017;102(12):2086–96.
11. Mondelaers V, Suciu S, De Moerloose B, Ferster A, Mazingue F, Plat G, et al. Prolonged *versus* standard native E. coli asparaginase therapy in childhood acute lymphoblastic leukemia and non-Hodgkin lymphoma: final results of the EORTC-CLG randomized phase III trial 58951. Haematologica. 2017;102(10):1727–38.
12. Burkhardt B, Mueller S, Khanam T, Perkins SL. Current status and future directions of T-lymphoblastic lymphoma in children and adolescents. Br J Haematol. 2016;173(4):545–59.
13. Atra A, Gerrard M, Hobson R, Imeson JD, Hann IM, Pinkerton CR. Outcome of relapsed or refractory childhood B-cell acute lymphoblastic leukaemia and B-cell non-Hodgkin's lymphoma treated with the UKCCSG 9003/9002 protocols. Br J Haematol. 2001;112(4):965–8.
14. Turner SD, Lamant L, Kenner L, Brugieres L. Anaplastic large cell lymphoma in paediatric and young adult patients. Br J Haematol. 2016;173(4):560–72.
15. Stein H, Foss HD, Durkop H, Marafioti T, Delsol G, Pulford K, et al. CD30(+) anaplastic large cell lymphoma: a review of its histopathologic, genetic, and clinical features. Blood. 2000;96(12):3681–95.
16. Fraser C, Brown P, Megason G, Ahn HS, Cho B, Kirov I, et al. Open-label bendamustine monotherapy for pediatric patients with relapsed or refractory acute leukemia: efficacy and tolerability. J Pediatr Hematol Oncol. 2014;36(4):e212–8.
17. Jeha S, Metzger ML, Crews KR, Campbell P, Ribeiro RC, Cheng C, et al. A phase I trial of bendamustine in combination with

clofarabine and etoposide in pediatric patients with relapsed or refractory hematologic malignancies. Blood. 2016;128(22):1628.
18. Berg SL, Cairo MS, Russell H, Ayello J, Ingle AM, Lau H, et al. Safety, pharmacokinetics, and immunomodulatory effects of lenalidomide in children and adolescents with relapsed/refractory solid tumors or myelodysplastic syndrome: a children's oncology group phase I consortium report. J Clin Oncol. 2011;29(3):316–23.
19. Horton TM, Gannavarapu A, Blaney SM, D'Argenio DZ, Plon SE, Berg SL. Bortezomib interactions with chemotherapy agents in acute leukemia in vitro. Cancer Chemother Pharmacol. 2006;58(1):13–23.
20. Houghton PJ, Morton CL, Kolb EA, Lock R, Carol H, Reynolds CP, et al. Initial testing (stage 1) of the proteasome inhibitor bortezomib by the pediatric preclinical testing program. Pediatr Blood Cancer. 2008;50(1):37–45.
21. Messinger YH, Gaynon PS, Sposto R, van der Giessen J, Eckroth E, Malvar J, et al. Bortezomib with chemotherapy is highly active in advanced B-precursor acute lymphoblastic leukemia: Therapeutic Advances in Childhood Leukemia & Lymphoma (TACL) Study. Blood. 2012;120(2):285–90.
22. Lee S, Yin C, Ayello J, Morris E, O'Connell T, Harrison L, et al. Ibrutinib significantly prolonged survival in a human Burkitt Lymphoma (BL) Xenograft NSG mouse model: ibrutinib may be a potential adjuvant agent in the treatment of BL. Blood. 2015;126(23):5117.
23. Bhatti M, Ippolito T, Mavis C, Gu J, Cairo MS, Lim MS, et al. Pre-clinical activity of targeting the PI3K/Akt/mTOR pathway in Burkitt lymphoma. Oncotarget. 2018;9(31):21820–30.
24. Souers AJ, Leverson JD, Boghaert ER, Ackler SL, Catron ND, Chen J, et al. ABT-199, a potent and selective BCL-2 inhibitor, achieves antitumor activity while sparing platelets. Nat Med. 2013;19(2):202–8.
25. Merchant MS, Wright M, Baird K, Wexler LH, Rodriguez-Galindo C, Bernstein D, et al. Phase I clinical trial of ipilimumab in pediatric patients with advanced solid tumors. Clin Cancer Res. 2016;22(6):1364–70.
26. DiJoseph JF, Popplewell A, Tickle S, Ladyman H, Lawson A, Kunz A, et al. Antibody-targeted chemotherapy of B-cell lymphoma using calicheamicin conjugated to murine or humanized antibody against CD22. Cancer Immunol Immunother. 2005;54(1):11–24.
27. Bhojwani D, Sposto R, Shah N, Rodriguez V, O'Brien MM, McNeer JL, et al. Inotuzumab ozogamicin in pediatric patients with relapsed/refractory acute lymphoblastic leukemia (R/R ALL). J Clin Oncol. 2017;35(15_suppl):10512.
28. Shewach DS, Daddona PE, Ashcraft E, Mitchell BS. Metabolism and selective cytotoxicity of 9-beta-D-arabinofuranosylguanine in human lymphoblasts. Cancer Res. 1985;45(3):1008–14.
29. Winter SS, Dunsmore KP, Devidas M, Eisenberg N, Asselin BL, Wood BL, et al. Safe integration of nelarabine into intensive chemotherapy in newly diagnosed T-cell acute lymphoblastic leukemia: Children's Oncology Group Study AALL0434. Pediatr Blood Cancer. 2015;62(7):1176–83.
30. Houghton PJ, Morton CL, Gorlick R, Lock RB, Carol H, Reynolds CP, et al. Stage 2 combination testing of rapamycin with cytotoxic agents by the pediatric preclinical testing program. Mol Cancer Ther. 2010;9(1):101–12.
31. Place AE, Pikman Y, Stevenson KE, Harris MH, Pauly M, Sulis ML, et al. Phase I trial of the mTOR inhibitor everolimus in combination with multi-agent chemotherapy in relapsed childhood acute lymphoblastic leukemia. Pediatr Blood Cancer. 2018;65(7):e27062.
32. Kalac M, Scotto L, Marchi E, Amengual J, Seshan VE, Bhagat G, et al. HDAC inhibitors and decitabine are highly synergistic and associated with unique gene-expression and epigenetic profiles in models of DLBCL. Blood. 2011;118(20):5506–16.
33. Ogura M, Ando K, Suzuki T, Ishizawa K, Oh SY, Itoh K, et al. A multicentre phase II study of vorinostat in patients with relapsed or refractory indolent B-cell non-Hodgkin lymphoma and mantle cell lymphoma. Br J Haematol. 2014;165(6):768–76.
34. Straus DJ, Hamlin PA, Matasar MJ, Lia Palomba M, Drullinsky PR, Zelenetz AD, et al. Phase I/II trial of vorinostat with rituximab, cyclophosphamide, etoposide and prednisone as palliative treatment for elderly patients with relapsed or refractory diffuse large B-cell lymphoma not eligible for autologous stem cell transplantation. Br J Haematol. 2015;168(5):663–70.
35. Nieto Y, Valdez BC, Thall PF, Ahmed S, Jones RB, Hosing C, et al. Vorinostat combined with high-dose gemcitabine, busulfan, and melphalan with autologous stem cell transplantation in patients with refractory lymphomas. Biol Blood Marrow Transplant. 2015;21(11):1914–20.
36. McCabe MT, Ott HM, Ganji G, Korenchuk S, Thompson C, Van Aller GS, et al. EZH2 inhibition as a therapeutic strategy for lymphoma with EZH2-activating mutations. Nature. 2012;492(7427):108–12.
37. Italiano A, Soria JC, Toulmonde M, Michot JM, Lucchesi C, Varga A, et al. Tazemetostat, an EZH2 inhibitor, in relapsed or refractory B-cell non-Hodgkin lymphoma and advanced solid tumours: a first-in-human, open-label, phase 1 study. Lancet Oncol. 2018;19(5):649–59.
38. Fry DW, Harvey PJ, Keller PR, Elliott WL, Meade M, Trachet E, et al. Specific inhibition of cyclin-dependent kinase 4/6 by PD 0332991 and associated antitumor activity in human tumor xenografts. Mol Cancer Ther. 2004;3(11):1427–38.
39. Tate SC, Cai S, Ajamie RT, Burke T, Beckmann RP, Chan EM, et al. Semi-mechanistic pharmacokinetic/pharmacodynamic modeling of the antitumor activity of LY2835219, a new cyclin-dependent kinase 4/6 inhibitor, in mice bearing human tumor xenografts. Clin Cancer Res. 2014;20(14):3763–74.
40. Sherr CJ, Beach D, Shapiro GI. Targeting CDK4 and CDK6: from discovery to therapy. Cancer Discov. 2016;6(4):353–67.
41. Wang M, Rule S, Zinzani PL, Goy A, Casasnovas O, Smith SD, et al. Acalabrutinib in relapsed or refractory mantle cell lymphoma (ACE-LY-004): a single-arm, multicentre, phase 2 trial. Lancet. 2018;391(10121):659–67.
42. Lee S, Shah T, Yin C, Hochberg J, Ayello J, Morris E, et al. Ruxolitinib significantly enhances in vitro apoptosis in Hodgkin lymphoma and primary mediastinal B-cell lymphoma and survival in a lymphoma xenograft murine model. Oncotarget. 2018;9(11):9776–88.
43. Delgado-Martin C, Meyer LK, Huang BJ, Shimano KA, Zinter MS, Nguyen JV, et al. JAK/STAT pathway inhibition overcomes IL7-induced glucocorticoid resistance in a subset of human T-cell acute lymphoblastic leukemias. Leukemia. 2017;31:2568.
44. Loh ML, Tasian SK, Rabin KR, Brown P, Magoon D, Reid JM, et al. A phase 1 dosing study of ruxolitinib in children with relapsed or refractory solid tumors, leukemias, or myeloproliferative neoplasms: A Children's Oncology Group phase 1 consortium study (ADVL1011). Pediatr Blood Cancer. 2015;62(10):1717–24.
45. Bride KL, Vincent TL, Im SY, Aplenc R, Barrett DM, Carroll WL, et al. Preclinical efficacy of daratumumab in T-cell acute lymphoblastic leukemia. Blood. 2018;131(9):995–9.
46. Mosse YP, Lim MS, Voss SD, Wilner K, Ruffner K, Laliberte J, et al. Safety and activity of crizotinib for paediatric patients with refractory solid tumours or anaplastic large-cell lymphoma: a Children's Oncology Group phase 1 consortium study. The lancet oncology. 2013;14(6):472–80.
47. Younes A, Bartlett NL, Leonard JP, Kennedy DA, Lynch CM, Sievers EL, et al. Brentuximab vedotin (SGN-35) for relapsed CD30-positive lymphomas. N Engl J Med. 2010;363(19):1812–21.
48. Cole PD, McCarten KM, Pei Q, Spira M, Metzger ML, Drachtman RA, et al. Brentuximab vedotin with gemcitabine for paediatric and

young adult patients with relapsed or refractory Hodgkin's lymphoma (AHOD1221): a Children's Oncology Group, multicentre single-arm, phase 1–2 trial. Lancet Oncol. 2018;19(9):1229–38.
49. Hochberg J, Harrison L, Morris E, Militano O, Brand P, Fabricatore S, et al. Safety of liposomal cytarabine CNS prophylaxis in children, adolescent and young adult hematopoietic stem cell transplant recipients with acute leukemia and non-Hodgkin lymphoma. Bone Marrow Transplant. 2016;51(9):1249–52.
50. Rummel MJ, Al-Batran SE, Kim SZ, Welslau M, Hecker R, Kofahl-Krause D, et al. Bendamustine plus rituximab is effective and has a favorable toxicity profile in the treatment of mantle cell and low-grade non-Hodgkin's lymphoma. J Clin Oncol. 2005;23(15):3383–9.
51. Ogura M, Ishizawa K, Maruyama D, Uike N, Ando K, Izutsu K, et al. Bendamustine plus rituximab for previously untreated patients with indolent B-cell non-Hodgkin lymphoma or mantle cell lymphoma: a multicenter phase II clinical trial in Japan. Int J Hematol. 2017;105(4):470–7.
52. Flinn IW, van der Jagt R, Kahl BS, Wood P, Hawkins TE, Macdonald D, et al. Randomized trial of bendamustine-rituximab or R-CHOP/R-CVP in first-line treatment of indolent NHL or MCL: the BRIGHT study. Blood. 2014;123(19):2944–52.
53. Vacirca JL, Acs PI, Tabbara IA, Rosen PJ, Lee P, Lynam E. Bendamustine combined with rituximab for patients with relapsed or refractory diffuse large B cell lymphoma. Ann Hematol. 2014;93(3):403–9.
54. Ohmachi K, Niitsu N, Uchida T, Kim SJ, Ando K, Takahashi N, et al. Multicenter phase II study of bendamustine plus rituximab in patients with relapsed or refractory diffuse large B-cell lymphoma. J Clin Oncol. 2013;31(17):2103–9.
55. Park SI, Grover NS, Olajide O, Asch AS, Wall JG, Richards KL, et al. A phase II trial of bendamustine in combination with rituximab in older patients with previously untreated diffuse large B-cell lymphoma. Br J Haematol. 2016;175(2):281–9.
56. Leonard JP, Jung SH, Johnson J, Pitcher BN, Bartlett NL, Blum KA, et al. Randomized trial of lenalidomide alone versus lenalidomide plus rituximab in patients with recurrent follicular lymphoma: CALGB 50401 (alliance). J Clin Oncol. 2015;33(31):3635–40.
57. Martin P, Jung SH, Pitcher B, Bartlett NL, Blum KA, Shea T, et al. A phase II trial of lenalidomide plus rituximab in previously untreated follicular non-Hodgkin's lymphoma (NHL): CALGB 50803 (Alliance). Ann Oncol. 2017;28(11):2806–12.
58. Wiernik PH, Lossos IS, Tuscano JM, Justice G, Vose JM, Cole CE, et al. Lenalidomide monotherapy in relapsed or refractory aggressive non-Hodgkin's lymphoma. J Clin Oncol. 2008;26(30):4952–7.
59. Witzig TE, Vose JM, Zinzani PL, Reeder CB, Buckstein R, Polikoff JA, et al. An international phase II trial of single-agent lenalidomide for relapsed or refractory aggressive B-cell non-Hodgkin's lymphoma. Ann Oncol. 2011;22(7):1622–7.
60. Czuczman MS, Trneny M, Davies A, Rule S, Linton KM, Wagner-Johnston N, et al. A phase 2/3 multicenter, randomized, open-label study to compare the efficacy and safety of lenalidomide versus investigator's choice in patients with relapsed or refractory diffuse large B-cell lymphoma. Clin Cancer Res. 2017;23(15):4127–37.
61. Nowakowski GS, LaPlant B, Macon WR, Reeder CB, Foran JM, Nelson GD, et al. Lenalidomide combined with R-CHOP overcomes negative prognostic impact of non-germinal center B-cell phenotype in newly diagnosed diffuse large B-Cell lymphoma: a phase II study. J Clin Oncol. 2015;33(3):251–7.
62. Nowakowski GS, Chiappella A, Witzig TE, Spina M, Gascoyne RD, Zhang L, et al. ROBUST: lenalidomide-R-CHOP versus placebo-R-CHOP in previously untreated ABC-type diffuse large B-cell lymphoma. Future Oncol. 2016;12(13):1553–63.
63. Khouri IF, Fernandez Curbelo I, Turturro F, Jabbour EJ, Milton DR, Bassett RL Jr, et al. Ipilimumab plus lenalidomide after allogeneic and autologous stem cell transplantation for patients with lymphoid malignancies. Clin Cancer Res. 2018;24(5):1011–8.
64. Smith SM, Pitcher BN, Jung SH, Bartlett NL, Wagner-Johnston N, Park SI, et al. Safety and tolerability of idelalisib, lenalidomide, and rituximab in relapsed and refractory lymphoma: the alliance for clinical trials in oncology A051201 and A051202 phase 1 trials. Lancet Haematol. 2017;4(4):e176–e82.
65. Fowler N, Kahl BS, Lee P, Matous JV, Cashen AF, Jacobs SA, et al. Bortezomib, bendamustine, and rituximab in patients with relapsed or refractory follicular lymphoma: the phase II VERTICAL study. J Clin Oncol. 2011;29(25):3389–95.
66. Ribrag V, Tilly H, Casasnovas O, Bosly A, Bouabdallah R, Delarue R, et al. Efficacy and toxicity of two schedules of bortezomib in patients with recurrent or refractory follicular lymphoma: a randomised phase II trial from the Groupe d'Etude des Lymphomes de l'Adulte (GELA). Eur J Cancer. 2013;49(4):904–10.
67. Goy A, Younes A, McLaughlin P, Pro B, Romaguera JE, Hagemeister F, et al. Phase II study of proteasome inhibitor bortezomib in relapsed or refractory B-cell non-Hodgkin's lymphoma. J Clin Oncol. 2005;23(4):667–75.
68. Coiffier B, Osmanov EA, Hong X, Scheliga A, Mayer J, Offner F, et al. Bortezomib plus rituximab versus rituximab alone in patients with relapsed, rituximab-naive or rituximab-sensitive, follicular lymphoma: a randomised phase 3 trial. Lancet Oncol. 2011;12(8):773–84.
69. Zinzani PL, Khuageva NK, Wang H, Garicochea B, Walewski J, Van Hoof A, et al. Bortezomib plus rituximab versus rituximab in patients with high-risk, relapsed, rituximab-naive or rituximab-sensitive follicular lymphoma: subgroup analysis of a randomized phase 3 trial. J Hematol Oncol. 2012;5:67.
70. Sehn LH, MacDonald D, Rubin S, Cantin G, Rubinger M, Lemieux B, et al. Bortezomib ADDED to R-CVP is safe and effective for previously untreated advanced-stage follicular lymphoma: a phase II study by the National Cancer Institute of Canada Clinical Trials Group. J Clin Oncol. 2011;29(25):3396–401.
71. Evens AM, Rosen ST, Helenowski I, Kline J, Larsen A, Colvin J, et al. A phase I/II trial of bortezomib combined concurrently with gemcitabine for relapsed or refractory DLBCL and peripheral T-cell lymphomas. Br J Haematol. 2013;163(1):55–61.
72. Leonard JP, Kolibaba KS, Reeves JA, Tulpule A, Flinn IW, Kolevska T, et al. Randomized phase II study of R-CHOP with or without bortezomib in previously untreated patients with non-germinal center B-cell-like diffuse large B-cell lymphoma. J Clin Oncol. 2017;35(31):3538–46.
73. Offner F, Samoilova O, Osmanov E, Eom HS, Topp MS, Raposo J, et al. Frontline rituximab, cyclophosphamide, doxorubicin, and prednisone with bortezomib (VR-CAP) or vincristine (R-CHOP) for non-GCB DLBCL. Blood. 2015;126(16):1893–901.
74. Kim SJ, Yoon DH, Kang HJ, Kim JS, Park SK, Kim HJ, et al. Bortezomib in combination with CHOP as first-line treatment for patients with stage III/IV peripheral T-cell lymphomas: a multicentre, single-arm, phase 2 trial. Eur J Cancer. 2012;48(17):3223–31.
75. Blaney SM, Bernstein M, Neville K, Ginsberg J, Kitchen B, Horton T, et al. Phase I study of the proteasome inhibitor bortezomib in pediatric patients with refractory solid tumors: a Children's Oncology Group study (ADVL0015). J Clin Oncol. 2004;22(23):4804–9.
76. Muscal JA, Thompson PA, Horton TM, Ingle AM, Ahern CH, McGovern RM, et al. A phase I trial of vorinostat and bortezomib in children with refractory or recurrent solid tumors: a Children's Oncology Group phase I consortium study (ADVL0916). Pediatr Blood Cancer. 2013;60(3):390–5.
77. Scarfo I, Ormhoj M, Frigault MJ, Castano AP, Lorrey S, Bouffard AA, et al. Anti-CD37 chimeric antigen receptor T cells are active against B and T cell lymphomas. Blood. 2018;132(23):2527.

78. Mitsiades N, Mitsiades CS, Richardson PG, Poulaki V, Tai YT, Chauhan D, et al. The proteasome inhibitor PS-341 potentiates sensitivity of multiple myeloma cells to conventional chemotherapeutic agents: therapeutic applications. Blood. 2003;101(6):2377–80.
79. Ma MH, Yang HH, Parker K, Manyak S, Friedman JM, Altamirano C, et al. The proteasome inhibitor PS-341 markedly enhances sensitivity of multiple myeloma tumor cells to chemotherapeutic agents. Clin Cancer Res. 2003;9(3):1136–44.
80. Zheng B, Zhou R, Gong Y, Yang X, Shan Q. Proteasome inhibitor bortezomib overcomes P-gp-mediated multidrug resistance in resistant leukemic cell lines. Int J Lab Hematol. 2012;34(3):237–47.
81. Wang CY, Cusack JC Jr, Liu R, Baldwin AS Jr. Control of inducible chemoresistance: enhanced anti-tumor therapy through increased apoptosis by inhibition of NF-kappaB. Nat Med. 1999;5(4):412–7.
82. Bhatti IA, Abhari BA, Fulda S. Identification of a synergistic combination of Smac mimetic and Bortezomib to trigger cell death in B-cell non-Hodgkin lymphoma cells. Cancer Lett. 2017;405:63–72.
83. Reilly KM, Kisor DF. Profile of nelarabine: use in the treatment of T-cell acute lymphoblastic leukemia. Onco Targets Ther. 2009;2:219–28.
84. Kurtzberg J, Ernst TJ, Keating MJ, Gandhi V, Hodge JP, Kisor DF, et al. Phase I study of 506 U78 administered on a consecutive 5-day schedule in children and adults with refractory hematologic malignancies. J Clin Oncol. 2005;23(15):3396–403.
85. Berg SL, Blaney SM, Devidas M, Lampkin TA, Murgo A, Bernstein M, et al. Phase II study of nelarabine (compound 506 U78) in children and young adults with refractory T-cell malignancies: a report from the Children's Oncology Group. J Clin Oncol. 2005;23(15):3376–82.
86. Commander LA, Seif AE, Insogna IG, Rheingold SR. Salvage therapy with nelarabine, etoposide, and cyclophosphamide in relapsed/refractory paediatric T-cell lymphoblastic leukaemia and lymphoma. Br J Haematol. 2010;150(3):345–51.
87. Whitlock J, dalla Pozza L, Goldberg JM, Silverman LB, Ziegler DS, Attarbaschi A, et al. Nelarabine in combination with etoposide and cyclophosphamide is active in first relapse of childhood T-acute lymphocytic leukemia (T-ALL) and T-lymphoblastic lymphoma (T-LL). Blood. 2014;124(21):795.
88. Wilson WH, Young RM, Schmitz R, Yang Y, Pittaluga S, Wright G, et al. Targeting B cell receptor signaling with ibrutinib in diffuse large B cell lymphoma. Nat Med. 2015;21(8):922–6.
89. Younes A, Thieblemont C, Morschhauser F, Flinn I, Friedberg JW, Amorim S, et al. Combination of ibrutinib with rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone (R-CHOP) for treatment-naive patients with CD20-positive B-cell non-Hodgkin lymphoma: a non-randomised, phase 1b study. Lancet Oncol. 2014;15(9):1019–26.
90. Lee S, Day NS, Miles RR, Perkins SL, Lim MS, Ayello J, et al. Comparative genomic expression signatures of signal transduction pathways and targets in paediatric Burkitt lymphoma: a Children's Oncology Group report. Br J Haematol. 2017;177(4):601–11.
91. Gopal AK, Kahl BS, de Vos S, Wagner-Johnston ND, Schuster SJ, Jurczak WJ, et al. PI3Kdelta inhibition by idelalisib in patients with relapsed indolent lymphoma. N Engl J Med. 2014;370(11):1008–18.
92. Barr PM, Saylors GB, Spurgeon SE, Cheson BD, Greenwald DR, O'Brien SM, et al. Phase 2 study of idelalisib and entospletinib: pneumonitis limits combination therapy in relapsed refractory CLL and NHL. Blood. 2016;127(20):2411–5.
93. Zelenetz AD, Barrientos JC, Brown JR, Coiffier B, Delgado J, Egyed M, et al. Idelalisib or placebo in combination with bendamustine and rituximab in patients with relapsed or refractory chronic lymphocytic leukaemia: interim results from a phase 3, randomised, double-blind, placebo-controlled trial. Lancet Oncol. 2017;18(3):297–311.
94. Frys S, Czuczman NM, Mavis C, Rolland DCM, Lim MS, Tiwari A, et al. Deregulation of the PI3K/Akt signal transduction pathway is associated with the development of chemotherapy resistance and can be effectively targeted to improve chemoresponsiveness in burkitt lymphoma pre-clinical models. Blood. 2014;124(21):1769.
95. Horwitz SM, Koch R, Porcu P, Oki Y, Moskowitz A, Perez M, et al. Activity of the PI3K-delta,gamma inhibitor duvelisib in a phase 1 trial and preclinical models of T-cell lymphoma. Blood. 2018;131(8):888–98.
96. Place AE, Goldsmith K, Bourquin JP, Loh ML, Gore L, Morgenstern DA, et al. Accelerating drug development in pediatric cancer: a novel Phase I study design of venetoclax in relapsed/refractory malignancies. Future Oncol. 2018;14(21):2115–29.
97. Cully M, You H, Levine AJ, Mak TW. Beyond PTEN mutations: the PI3K pathway as an integrator of multiple inputs during tumorigenesis. Nat Rev Cancer. 2006;6(3):184–92.
98. Janku F, Yap TA, Meric-Bernstam F. Targeting the PI3K pathway in cancer: are we making headway? Nat Rev Clin Oncol. 2018;15(5):273–91.
99. Witzig TE, Reeder C, Han JJ, LaPlant B, Stenson M, Tun HW, et al. The mTORC1 inhibitor everolimus has antitumor activity in vitro and produces tumor responses in patients with relapsed T-cell lymphoma. Blood. 2015;126(3):328–35.
100. Witzig TE, Reeder CB, LaPlant BR, Gupta M, Johnston PB, Micallef IN, et al. A phase II trial of the oral mTOR inhibitor everolimus in relapsed aggressive lymphoma. Leukemia. 2011;25(2):341–7.
101. Rheingold SR, Tasian SK, Whitlock JA, Teachey DT, Borowitz MJ, Liu X, et al. A phase 1 trial of temsirolimus and intensive re-induction chemotherapy for second or greater relapse of acute lymphoblastic leukaemia: a Children's Oncology Group study (ADVL1114). Br J Haematol. 2017;177(3):467–74.
102. Vijayaraghavan S, Moulder S, Keyomarsi K, Layman RM. Inhibiting CDK in cancer therapy: current evidence and future directions. Target Oncol. 2018;13(1):21–38.
103. Bose P, Simmons GL, Grant S. Cyclin-dependent kinase inhibitor therapy for hematologic malignancies. Expert Opin Investig Drugs. 2013;22(6):723–38.
104. Dickson MA, Schwartz GK. Development of cell-cycle inhibitors for cancer therapy. Curr Oncol. 2009;16(2):36–43.
105. Leonard JP, AS LC, Smith MR, Noy A, Chirieac LR, Rodig SJ, et al. Selective CDK4/6 inhibition with tumor responses by PD0332991 in patients with mantle cell lymphoma. Blood. 2012;119(20):4597–607.
106. Maude SL, Tasian SK, Vincent T, Hall JW, Sheen C, Roberts KG, et al. Targeting JAK1/2 and mTOR in murine xenograft models of Ph-like acute lymphoblastic leukemia. Blood. 2012;120(17):3510–8.
107. Nawijn MC, Alendar A, Berns A. For better or for worse: the role of Pim oncogenes in tumorigenesis. Nat Rev Cancer. 2011;11(1):23–34.
108. An N, Kraft AS, Kang Y. Abnormal hematopoietic phenotypes in Pim kinase triple knockout mice. J Hematol Oncol. 2013;6:12.
109. Cho SH, Goenka S, Henttinen T, Gudapati P, Reinikainen A, Eischen CM, et al. PARP-14, a member of the B aggressive lymphoma family, transduces survival signals in primary B cells. Blood. 2009;113(11):2416–25.
110. Gomez-Abad C, Pisonero H, Blanco-Aparicio C, Roncador G, Gonzalez-Menchen A, Martinez-Climent JA, et al. PIM2 inhibition as a rational therapeutic approach in B-cell lymphoma. Blood. 2011;118(20):5517–27.
111. Cohen AM, Grinblat B, Bessler H, Kristt D, Kremer A, Schwartz A, et al. Increased expression of the hPim-2 gene in human

chronic lymphocytic leukemia and non-Hodgkin lymphoma. Leuk Lymphoma. 2004;45(5):951–5.
112. Hsi ED, Jung SH, Lai R, Johnson JL, Cook JR, Jones D, et al. Ki67 and PIM1 expression predict outcome in mantle cell lymphoma treated with high dose therapy, stem cell transplantation and rituximab: a Cancer and Leukemia Group B 59909 correlative science study. Leuk Lymphoma. 2008;49(11):2081–90.
113. Brault L, Menter T, Obermann EC, Knapp S, Thommen S, Schwaller J, et al. PIM kinases are progression markers and emerging therapeutic targets in diffuse large B-cell lymphoma. Br J Cancer. 2012;107(3):491–500.
114. van Lohuizen M, Verbeek S, Krimpenfort P, Domen J, Saris C, Radaszkiewicz T, et al. Predisposition to lymphomagenesis in pim-1 transgenic mice: cooperation with c-myc and N-myc in murine leukemia virus-induced tumors. Cell. 1989;56(4):673–82.
115. Kreuz S, Holmes KB, Tooze RM, Lefevre PF. Loss of PIM2 enhances the anti-proliferative effect of the pan-PIM kinase inhibitor AZD1208 in non-Hodgkin lymphomas. Mol Cancer. 2015;14:205.
116. Allis CD, Jenuwein T. The molecular hallmarks of epigenetic control. Nat Rev Genet. 2016;17(8):487–500.
117. Kulis M, Esteller M. DNA methylation and cancer. Adv Genet. 2010;70:27–56.
118. Maio M, Covre A, Fratta E, Di Giacomo AM, Taverna P, Natali PG, et al. Molecular pathways: at the crossroads of cancer epigenetics and immunotherapy. Clin Cancer Res. 2015;21(18):4040–7.
119. Blum KA, Liu Z, Lucas DM, Chen P, Xie Z, Baiocchi R, et al. Phase I trial of low dose decitabine targeting DNA hypermethylation in patients with chronic lymphocytic leukaemia and non-Hodgkin lymphoma: dose-limiting myelosuppression without evidence of DNA hypomethylation. Br J Haematol. 2010;150(2):189–95.
120. Crump M, Coiffier B, Jacobsen ED, Sun L, Ricker JL, Xie H, et al. Phase II trial of oral vorinostat (suberoylanilide hydroxamic acid) in relapsed diffuse large-B-cell lymphoma. Ann Oncol. 2008;19(5):964–9.
121. Lue JK, Amengual JE. Emerging EZH2 inhibitors and their application in lymphoma. Curr Hematol Malig Rep. 2018;13(5):369–82.
122. Morin RD, Johnson NA, Severson TM, Mungall AJ, An J, Goya R, et al. Somatic mutations altering EZH2 (Tyr641) in follicular and diffuse large B-cell lymphomas of germinal-center origin. Nat Genet. 2010;42(2):181–5.
123. Bodor C, O'Riain C, Wrench D, Matthews J, Iyengar S, Tayyib H, et al. EZH2 Y641 mutations in follicular lymphoma. Leukemia. 2011;25(4):726–9.
124. Shaw AT, Kim DW, Nakagawa K, Seto T, Crino L, Ahn MJ, et al. Crizotinib versus chemotherapy in advanced ALK-positive lung cancer. N Engl J Med. 2013;368(25):2385–94.
125. Li J, Liu W, Luo H, Bao J. Insight into drug resistance mechanisms and discovery of potential inhibitors against wild-type and L1196 M mutant ALK from FDA-approved drugs. J Mol Model. 2016;22(9):231.
126. Delsol GJ, Falini B, Muller-Hermelink HK, Campo E, Jaffe ES, Gascoyne RD, Stein H, Kinney MC. Anaplastic Large Cell Lymphoma (ALCL), ALK-positive. In: Swerdlow SH, Campo E, Lee Harris N, editors. WHO classification of tumors of the hematopoietic and lymphoid tissues. Lyon: International Agency for Research on Cancer (IARC); 2008. p. 312–6.
127. Majzner RG, Heitzeneder S, Mackall CL. Harnessing the immunotherapy revolution for the treatment of childhood cancers. Cancer Cell. 2017;31(4):476–85.
128. Eyre TA, Collins GP. Immune checkpoint inhibition in lymphoid disease. Br J Haematol. 2015;170(3):291–304.
129. Lulla P, Heslop HE. Checkpoint inhibition and cellular immunotherapy in lymphoma. Hematology Am Soc Hematol Educ Program. 2016;2016(1):390–6.
130. Hude I, Sasse S, Engert A, Brockelmann PJ. The emerging role of immune checkpoint inhibition in malignant lymphoma. Haematologica. 2017;102(1):30–42.
131. Dada R. Program death inhibitors in classical Hodgkin's lymphoma: a comprehensive review. Ann Hematol. 2018;97(4):555–61.
132. Weber JS, Yang JC, Atkins MB, Disis ML. Toxicities of immunotherapy for the practitioner. J Clin Oncol. 2015;33(18):2092–9.
133. Ansell SM, Hurvitz SA, Koenig PA, LaPlant BR, Kabat BF, Fernando D, et al. Phase I study of ipilimumab, an anti-CTLA-4 monoclonal antibody, in patients with relapsed and refractory B-cell non-Hodgkin lymphoma. Clin Cancer Res. 2009;15(20):6446–53.
134. Davids MS, Kim HT, Bachireddy P, Costello C, Liguori R, Savell A, et al. Ipilimumab for patients with relapse after allogeneic transplantation. N Engl J Med. 2016;375(2):143–53.
135. Ribas A, Kefford R, Marshall MA, Punt CJ, Haanen JB, Marmol M, et al. Phase III randomized clinical trial comparing tremelimumab with standard-of-care chemotherapy in patients with advanced melanoma. J Clin Oncol. 2013;31(5):616–22.
136. Comin-Anduix B, Escuin-Ordinas H, Ibarrondo FJ. Tremelimumab: research and clinical development. Onco Targets Ther. 2016;9:1767–76.
137. Burke GAA, Gross TG, Pillon M, Minard-colin V, Delgado RF, Zsíros J, et al. Results of inter-B-NHL Ritux 2010 – phase II study of DA-EPOCH-R for children and adolescents with primary mediastinal large B-cell lymphoma (PMLBL) on behalf of European Intergroup for Childhood Non Hodgkin's Lymphoma (EICNHL) and Children's Oncology Group (COG). Blood. 2017;130(Suppl 1):4124.
138. Zinzani PL, Ribrag V, Moskowitz CH, Michot JM, Kuruvilla J, Balakumaran A, et al. Safety and tolerability of pembrolizumab in patients with relapsed/refractory primary mediastinal large B-cell lymphoma. Blood. 2017;130(3):267–70.
139. Nagato T, Ohkuri T, Ohara K, Hirata Y, Kishibe K, Komabayashi Y, et al. Programmed death-ligand 1 and its soluble form are highly expressed in nasal natural killer/T-cell lymphoma: a potential rationale for immunotherapy. Cancer Immunol Immunother. 2017;66(7):877–90.
140. Wang H, Wang L, Liu WJ, Xia ZJ, Huang HQ, Jiang WQ, et al. High post-treatment serum levels of soluble programmed cell death ligand 1 predict early relapse and poor prognosis in extranodal NK/T cell lymphoma patients. Oncotarget. 2016;7(22):33035–45.
141. Kwong YL, Chan TSY, Tan D, Kim SJ, Poon LM, Mow B, et al. PD1 blockade with pembrolizumab is highly effective in relapsed or refractory NK/T-cell lymphoma failing l-asparaginase. Blood. 2017;129(17):2437–42.
142. Munir S, Andersen GH, Woetmann A, Odum N, Becker JC, Andersen MH. Cutaneous T cell lymphoma cells are targets for immune checkpoint ligand PD-L1-specific, cytotoxic T cells. Leukemia. 2013;27(11):2251–3.
143. AHR MK, Porcu P, Foss FM, Moskowitz AJ, Shustov AR, Shanbhag S, Sokol L, Shine R, Fling SP, Li S, Rabhar Z, Kim J, Yang Y, Yearley J, Chartash EK, Townson SM, Subrahmanyam PB, Maecker H, Alizadeh AA, Dai J, Horwitz SM, Sharon E, Kohrt H, Cheever MA, Kim Y. Pembrolizumab for treatment of relapsed/refractory mycosis fungoides and sezary syndrome: clinical efficacy in a citn multicenter phase 2 study. Blood. 2016;128(22):181.
144. Lesokhin AM, Ansell SM, Armand P, Scott EC, Halwani A, Gutierrez M, et al. Nivolumab in patients with relapsed or refractory hematologic malignancy: preliminary results of a phase Ib study. J Clin Oncol. 2016;34(23):2698–704.
145. Stephen Ansell MEG, Margaret A, Shipp DG, Moskowitz A, Borello I, Popa-Mckiver M, Farsaci B, Zhu L, Lesokhin AM, Armand P. A phase 1 study of Nivolumab in combination with ipilimumab for relapsed or refractory hematologic malignancies (CheckMate 039). Blood. 2016;128(22):183.

146. Salles G, Barrett M, Foa R, Maurer J, O'Brien S, Valente N, et al. Rituximab in B-cell hematologic malignancies: a review of 20 years of clinical experience. Adv Ther. 2017;34(10):2232–73.
147. Barth MJ, Goldman S, Smith L, Perkins S, Shiramizu B, Gross TG, et al. Rituximab pharmacokinetics in children and adolescents with de novo intermediate and advanced mature B-cell lymphoma/leukaemia: a children's oncology group report. Br J Haematol. 2013;162(5):678–83.
148. Goldman S, Smith L, Galardy P, Perkins SL, Frazer JK, Sanger W, et al. Rituximab with chemotherapy in children and adolescents with central nervous system and/or bone marrow-positive Burkitt lymphoma/leukaemia: a children's oncology group report. Br J Haematol. 2014;167(3):394–401.
149. Goldman S, Smith L, Anderson JR, Perkins S, Harrison L, Geyer MB, et al. Rituximab and FAB/LMB 96 chemotherapy in children with stage III/IV B-cell non-Hodgkin lymphoma: a children's oncology group report. Leukemia. 2013;27(5):1174–7.
150. Meinhardt A, Burkhardt B, Zimmermann M, Borkhardt A, Kontny U, Klingebiel T, et al. Phase II window study on rituximab in newly diagnosed pediatric mature B-cell non-Hodgkin's lymphoma and Burkitt leukemia. J Clin Oncol. 2010;28(19):3115–21.
151. Dunleavy K, Pittaluga S, Maeda LS, Advani R, Chen CC, Hessler J, et al. Dose-adjusted EPOCH-rituximab therapy in primary mediastinal B-cell lymphoma. N Engl J Med. 2013;368(15):1408–16.
152. Morschhauser FA, Cartron G, Thieblemont C, Solal-Celigny P, Haioun C, Bouabdallah R, et al. Obinutuzumab (GA101) monotherapy in relapsed/refractory diffuse large b-cell lymphoma or mantle-cell lymphoma: results from the phase II GAUGUIN study. J Clin Oncol. 2013;31(23):2912–9.
153. Vitolo U, Trneny M, Belada D, Burke JM, Carella AM, Chua N, et al. Obinutuzumab or rituximab plus cyclophosphamide, doxorubicin, vincristine, and prednisone in previously untreated diffuse large B-cell lymphoma. J Clin Oncol. 2017;35(31):3529–37.
154. Leonard JP, Coleman M, Ketas JC, Chadburn A, Furman R, Schuster MW, et al. Epratuzumab, a humanized anti-CD22 antibody, in aggressive non-Hodgkin's lymphoma: phase I/II clinical trial results. Clin Cancer Res. 2004;10(16):5327–34.
155. Lokhorst HM, Plesner T, Laubach JP, Nahi H, Gimsing P, Hansson M, et al. Targeting CD38 with daratumumab monotherapy in multiple myeloma. N Engl J Med. 2015;373(13):1207–19.
156. van de Donk N, Richardson PG, Malavasi F. CD38 antibodies in multiple myeloma: back to the future. Blood. 2018;131(1):13–29.
157. Shah NN, Singavi AK, Harrington A. Daratumumab in primary effusion lymphoma. N Engl J Med. 2018;379(7):689–90.
158. Hari P, Raj RV, Olteanu H. Targeting CD38 in refractory extranodal natural killer cell-T-cell lymphoma. N Engl J Med. 2016;375(15):1501–2.
159. Goebeler ME, Knop S, Viardot A, Kufer P, Topp MS, Einsele H, et al. Bispecific T-cell engager (BiTE) antibody construct blinatumomab for the treatment of patients with relapsed/refractory non-Hodgkin lymphoma: final results from a phase I study. J Clin Oncol. 2016;34(10):1104–11.
160. Cesano A, Gayko U. CD22 as a target of passive immunotherapy. Semin Oncol. 2003;30(2):253–7.
161. Perkins SL, Lones MA, Davenport V, Cairo MS. B-cell non-Hodgkin's lymphoma in children and adolescents: surface antigen expression and clinical implications for future targeted bioimmune therapy: a children's cancer group report. Clin Adv Hematol Oncol. 2003;1(5):314–7.
162. Zein N, Sinha AM, McGahren WJ, Ellestad GA. Calicheamicin gamma 1I: an antitumor antibiotic that cleaves double-stranded DNA site specifically. Science. 1988;240(4856):1198–201.
163. Advani A, Coiffier B, Czuczman MS, Dreyling M, Foran J, Gine E, et al. Safety, pharmacokinetics, and preliminary clinical activity of inotuzumab ozogamicin, a novel immunoconjugate for the treatment of B-cell non-Hodgkin's lymphoma: results of a phase I study. J Clin Oncol. 2010;28(12):2085–93.
164. Fayad L, Offner F, Smith MR, Verhoef G, Johnson P, Kaufman JL, et al. Safety and clinical activity of a combination therapy comprising two antibody-based targeting agents for the treatment of non-Hodgkin lymphoma: results of a phase I/II study evaluating the immunoconjugate inotuzumab ozogamicin with rituximab. J Clin Oncol. 2013;31(5):573–83.
165. Ogura M, Tobinai K, Hatake K, Davies A, Crump M, Ananthakrishnan R, et al. Phase I study of inotuzumab ozogamicin combined with R-CVP for relapsed/refractory CD22+ B-cell non-Hodgkin lymphoma. Clin Cancer Res. 2016;22(19):4807–16.
166. Cheng J, Zhu H, Choi JK. CD30 expression in pediatric neoplasms, study of 585 cases. Pediatr Dev Pathol. 2017;20(3):191–6.
167. Falini B, Pileri S, Pizzolo G, Durkop H, Flenghi L, Stirpe F, et al. CD30 (Ki-1) molecule: a new cytokine receptor of the tumor necrosis factor receptor superfamily as a tool for diagnosis and immunotherapy. Blood. 1995;85(1):1–14.
168. Durkop H, Latza U, Hummel M, Eitelbach F, Seed B, Stein H. Molecular cloning and expression of a new member of the nerve growth factor receptor family that is characteristic for Hodgkin's disease. Cell. 1992;68(3):421–7.
169. Forero-Torres A, Leonard JP, Younes A, Rosenblatt JD, Brice P, Bartlett NL, et al. A phase II study of SGN-30 (anti-CD30 mAb) in Hodgkin lymphoma or systemic anaplastic large cell lymphoma. Br J Haematol. 2009;146(2):171–9.
170. Gravanis I, Tzogani K, van Hennik P, de Graeff P, Schmitt P, Mueller-Berghaus J, et al. The European medicines agency review of brentuximab vedotin (Adcetris) for the treatment of adult patients with relapsed or refractory CD30+ Hodgkin lymphoma or systemic anaplastic large cell lymphoma: summary of the scientific assessment of the committee for medicinal products for human use. Oncologist. 2016;21(1):102–9.
171. de Claro RA, McGinn K, Kwitkowski V, Bullock J, Khandelwal A, Habtemariam B, et al. U.S. food and drug administration approval summary: brentuximab vedotin for the treatment of relapsed Hodgkin lymphoma or relapsed systemic anaplastic large-cell lymphoma. Clin Cancer Res. 2012;18(21):5845–9.
172. Jacobsen ED, Sharman JP, Oki Y, Advani RH, Winter JN, Bello CM, et al. Brentuximab vedotin demonstrates objective responses in a phase 2 study of relapsed/refractory DLBCL with variable CD30 expression. Blood. 2015;125(9):1394–402.
173. Zinzani PL, Pellegrini C, Chiappella A, Di Rocco A, Salvi F, Cabras MG, et al. Brentuximab vedotin in relapsed primary mediastinal large B-cell lymphoma: results from a phase 2 clinical trial. Blood. 2017;129(16):2328–30.
174. Younes A, Connors JM, Park SI, Fanale M, O'Meara MM, Hunder NN, et al. Brentuximab vedotin combined with ABVD or AVD for patients with newly diagnosed Hodgkin's lymphoma: a phase 1, open-label, dose-escalation study. Lancet Oncol. 2013;14(13):1348–56.
175. Fanale MA, Horwitz SM, Forero-Torres A, Bartlett NL, Advani RH, Pro B, et al. Brentuximab vedotin in the front-line treatment of patients with CD30+ peripheral T-cell lymphomas: results of a phase I study. J Clin Oncol. 2014;32(28):3137–43.
176. Kumar A, Casulo C, Yahalom J, Schoder H, Barr PM, Caron P, et al. Brentuximab vedotin and AVD followed by involved-site radiotherapy in early stage, unfavorable risk Hodgkin lymphoma. Blood. 2016;128(11):1458–64.
177. Connors JM, Jurczak W, Straus DJ, Ansell SM, Kim WS, Gallamini A, et al. Brentuximab Vedotin with chemotherapy for stage III or IV Hodgkin's lymphoma. N Engl J Med. 2018;378(4):331–44.
178. Lim WA, June CH. The principles of engineering immune cells to treat cancer. Cell. 2017;168(4):724–40.

179. Fesnak AD, June CH, Levine BL. Engineered T cells: the promise and challenges of cancer immunotherapy. Nat Rev Cancer. 2016;16(9):566–81.
180. Neelapu SS, Tummala S, Kebriaei P, Wierda W, Gutierrez C, Locke FL, et al. Chimeric antigen receptor T-cell therapy – assessment and management of toxicities. Nat Rev Clin Oncol. 2018;15(1):47–62.
181. Maude SL, Laetsch TW, Buechner J, Rives S, Boyer M, Bittencourt H, et al. Tisagenlecleucel in children and young adults with B-cell lymphoblastic leukemia. N Engl J Med. 2018;378(5):439–48.
182. Brudno JN, Kochenderfer JN. Chimeric antigen receptor T-cell therapies for lymphoma. Nat Rev Clin Oncol. 2018;15(1):31–46.
183. Bollard CM, Rooney CM, Heslop HE. T-cell therapy in the treatment of post-transplant lymphoproliferative disease. Nat Rev Clin Oncol. 2012;9(9):510–9.
184. Bollard CM. Improving T-cell therapy for epstein-barr virus lymphoproliferative disorders. J Clin Oncol. 2013;31(1):5–7.
185. Wistinghausen B, Gross TG, Bollard C. Post-transplant lymphoproliferative disease in pediatric solid organ transplant recipients. Pediatr Hematol Oncol. 2013;30(6):520–31.
186. Bollard CM, Gottschalk S, Torrano V, Diouf O, Ku S, Hazrat Y, et al. Sustained complete responses in patients with lymphoma receiving autologous cytotoxic T lymphocytes targeting Epstein-Barr virus latent membrane proteins. J Clin Oncol. 2014;32(8):798–808.
187. Bollard CM, Gottschalk S, Helen Huls M, Leen AM, Gee AP, Rooney CM. Good manufacturing practice-grade cytotoxic T lymphocytes specific for latent membrane proteins (LMP)-1 and LMP2 for patients with Epstein-Barr virus-associated lymphoma. Cytotherapy. 2011;13(5):518–22.
188. Leen AM, Bollard CM, Mendizabal AM, Shpall EJ, Szabolcs P, Antin JH, et al. Multicenter study of banked third-party virus-specific T cells to treat severe viral infections after hematopoietic stem cell transplantation. Blood. 2013;121(26):5113–23.
189. Vickers MA, Wilkie GM, Robinson N, Rivera N, Haque T, Crawford DH, et al. Establishment and operation of a good manufacturing practice-compliant allogeneic Epstein-Barr virus (EBV)-specific cytotoxic cell bank for the treatment of EBV-associated lymphoproliferative disease. Br J Haematol. 2014;167(3):402–10.
190. Heslop HE, Slobod KS, Pule MA, Hale GA, Rousseau A, Smith CA, et al. Long-term outcome of EBV-specific T-cell infusions to prevent or treat EBV-related lymphoproliferative disease in transplant recipients. Blood. 2010;115(5):925–35.
191. Naik S, Nicholas SK, Martinez CA, Leen AM, Hanley PJ, Gottschalk SM, et al. Adoptive immunotherapy for primary immunodeficiency disorders with virus-specific T lymphocytes. J Allergy Clin Immunol. 2016;137(5):1498–505. e1
192. O'Reilly RJ, Prockop S, Hasan AN, Koehne G, Doubrovina E. Virus-specific T-cell banks for 'off the shelf' adoptive therapy of refractory infections. Bone Marrow Transplant. 2016;51(9):1163–72.
193. Haque T, Wilkie GM, Jones MM, Higgins CD, Urquhart G, Wingate P, et al. Allogeneic cytotoxic T-cell therapy for EBV-positive posttransplantation lymphoproliferative disease: results of a phase 2 multicenter clinical trial. Blood. 2007;110(4):1123–31.
194. Grant M, Bollard CM. Developing T-cell therapies for lymphoma without receptor engineering. Hematology Am Soc Hematol Educ Program. 2017;2017(1):622–31.
195. Leen A, Tzannou I, Bilgi M, Liu H, Vera JF, Gerdemann U, et al. Immunotherapy for lymphoma using T cells targeting multiple tumor associated antigens. Blood. 2015;126(23):186.
196. Williams KM, Hanley P, Grant M, Fortiz MF, Stanojevic M, Ismail M, et al. Complete remissions post infusion of multiple tumor antigen specific T cells for the treatment of high risk leukemia and lymphoma patients after HCT. Blood. 2017;130(Suppl 1):4516.
197. Bollard CM, Lim MS, Gross TG, Committee COGN-HL. Children's Oncology Group's 2013 blueprint for research: non-Hodgkin lymphoma. Pediatr Blood Cancer. 2013;60(6):979–84.
198. Griffin TC, Weitzman S, Weinstein H, Chang M, Cairo M, Hutchison R, et al. A study of rituximab and ifosfamide, carboplatin, and etoposide chemotherapy in children with recurrent/refractory B-cell (CD20+) non-Hodgkin lymphoma and mature B-cell acute lymphoblastic leukemia: a report from the Children's Oncology Group. Pediatr Blood Cancer. 2009;52(2):177–81.

27 Childhood and Adolescence Non-Hodgkin Lymphomas in Low- and Middle-Income Countries

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Introduction

The non-Hodgkin lymphomas (NHL) of childhood and adolescence represent an intriguing group of malignancies. Characterized by some of the highest cure rates among all pediatric cancers in high-income countries, their typically excellent response to chemotherapy usually prevails over the often drastic and severe initial clinical presentations. There is still much to learn about the epidemiology of pediatric NHL in low- and middle-income countries (LMIC), as well as a critical need to develop treatment protocols adapted to local settings. Nonetheless, despite varying limitations in resources across LMIC, this group of malignancies represents an ideal focal point upon which advances in curative outcomes for pediatric oncology in LMIC may be achieved. This unique opportunity arises from a combination of factors including the relatively high incidence of NHL in children worldwide, their ability to be cured with chemotherapy alone (without a need for complex surgical resections or radiation therapy), as well as the relatively short duration of most treatment protocols. Important challenges exist as well though, as the treatment protocols delivering favorable curative outcomes in high-income countries are based on chemotherapy backbones that include multi-agent intensive chemotherapy regimens often consisting of high-dose methotrexate and at times, high-dose cytarabine, requiring formidable provision of multidisciplinary supportive care (Table 27.1). This chapter will focus on NHL in sub-Saharan Africa (SSA) and Central and South America as a representation of the nuances of the epidemiology and management of pediatric NHL in LMIC.

Table 27.1 Opportunities for and challenges against improving curative outcomes for childhood and adolescent NHL in LMIC

Opportunities	Challenges
1. Relatively high incidence of pediatric NHL in LMIC 2. Excellent outcomes already established in high-income countries 3. Short duration of most treatment protocols 4. Complex surgical resections and radiation therapy not required	1. Requirement of intensive multi-agent chemotherapy regimens 2. Frequent use for high-dose methotrexate 3. Requirement of comprehensive supportive care practices

NHL non-Hodgkin lymphoma, *LMIC* low- and middle-income countries

Epidemiology of Childhood and Adolescent NHL in LMIC

While the four most common pediatric NHL—Burkitt lymphoma (BL), lymphoblastic lymphoma, diffuse large B-cell lymphoma (DLBCL), and anaplastic large cell lymphoma (ALCL)—account for the vast majority of diagnoses worldwide, the spectrum and distribution of disease can vary based on geography (Table 27.2) [1]. The most obvious of such distinctions is epitomized by the epidemiological variants of BL. Although sporadic BL is the most common NHL of childhood throughout the world, endemic BL occurs specifically in the malaria-endemic regions of Africa, where it is by far the most common childhood cancer overall [2–4]. On the other hand, a few rare NHL diagnoses appear to occur almost exclusively in individuals of specific ethnic origins. For example, some Epstein–Barr virus (EBV)-associated lymphomas have a distinct predilection for individuals of Asian or indigenous Central and South American descent, including the systemic EBV-positive T-cell lymphoma of childhood, extranodal NK-/

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Table 27.2 Distinct geographical associations of childhood and adolescent NHL in LMIC

Geographical region	Lymphoma	Infectious links
Equatorial Africa	Endemic Burkitt lymphoma	Epstein–Barr virus, malaria
Central and South America plus East Asia	Systemic EBV-positive T-cell lymphoma of childhood	Epstein–Barr virus
	Hydroa vacciniforme-like lymphoproliferative disorder	
	Extranodal NK-/T-cell lymphoma, nasal type	
East Asia, Caribbean Islands, Central and South America, Central/Southern Africa	Adult T-cell leukemia/lymphoma	Human T-cell lymphotropic virus type 1

NHL non-Hodgkin lymphoma, *LMIC* low- and middle-income countries, *EBV* Epstein–Barr virus

T-cell lymphoma, nasal type, and hydroa vacciniforme-like lymphoproliferative disorder [5–7]. Additionally, although rarely occurring in children, adult T-cell leukemia/lymphoma occurs in regions with endemic human T-cell lymphotropic virus type 1 (HTLV-1) infection including Japan and other countries in East Asia, the Caribbean islands, Central and South America, and central and southern Africa [8]. And while data have yet to be established, one may hypothesize that there is a greater incidence of human herpesvirus-8 (HHV-8)-associated lymphomas/lymphoproliferative disorders in central and eastern Africa, the region of the world with the highest prevalence of HHV-8 infection [9].

Furthermore, in the face of the HIV epidemic, the incidence of NHL will continue to evolve in SSA, with reports suggesting varying effects on overall disease burden as high as a 13-fold increase in adult NHL incidence in certain countries [10]. A comparison of international cancer registry databases revealed that SSA has the highest rates of NHL incidence in the world, while it is estimated that approximately 90% of all childhood and adolescent NHL occur in LMIC [1, 11]. This disproportionate disease burden highlights the importance of developing an improved understanding of NHL trends worldwide and working together as a global community to achieve improved curative outcomes for children and adolescents with lymphoma throughout the world, no matter how poor the country they live in.

Childhood and Adolescent NHL in Sub-Saharan Africa

The stark disparity in outcomes for childhood cancer between high- and low-income countries is well encapsulated by the story of Burkitt lymphoma (BL). Originally identified by Denis Burkitt and colleagues in the 1960s in Uganda, it was eventually realized that BL was the most common childhood NHL worldwide [12]. Due to its overall predominance as the most common childhood cancer in the region, the unmistakably unique and eye-catching clinical presentation with a rapidly enlarging jaw mass, as well as the incredible aura of history that surrounds it, endemic BL has almost become synonymous with NHL in equatorial Africa. This misconception unfortunately leads to both clinical- and pathology-based diagnostic bias to label patients as having BL even in atypical scenarios [13, 14]. Data from the International Network for Cancer Treatment and Research (INCTR) showed that BL accounted for 82% of all NHL in a multicenter study across SSA, and due to limitations in pathological diagnosis, other forms of NHL are significantly under-reported as their true incidence in Africa remains unknown [15]. Severe limitations exist in pathology laboratory resources—including lack of immunohistochemical stains, flow cytometry, and cytogenetic analyses, resulting in an over-reliance on morphology-based diagnoses from biopsy and fine needle aspirate samples [16]. Knowing the well-documented overlap in the morphological appearance of BL and the other common NHL of childhood, it is understandable that without the ability to identify the characteristic disease translocations, for example, clinicopathologic bias could easily produce errors of overcalling diagnoses of BL.

Endemic Burkitt Lymphoma in Sub-Saharan Africa

Endemic BL occurs in equatorial Africa as well as in Papua New Guinea [2–4]. Distribution of the disease corresponds to areas of holoendemic malaria infection and early acquisition of EBV infection [3, 4, 17]. This distinct geographic distribution informed the categorization of "endemic BL." More than 90% of BL in SSA is also associated with EBV infection [18]. This is in contrast to sporadic BL which is present in the rest of the world, including the non-malarial regions of SSA, where the association with EBV is typically 20–30% [19]. Estimates indicate that the incidence of endemic BL is approximately 30–60 per million children in equatorial Africa [3]. Children living in areas with high incidence of endemic BL acquire primary EBV infection at a much younger age (often before 3 years of age) [20]. It is hypothesized that younger age at time of acquisition of infection (resulting in poor control of viral infection) and co-infection with malaria are associated with higher frequencies of EBV reactivation and prolonged episodes of viremia [18, 21, 22]. EBV was in fact discovered in BL tumor samples from Ugandan children in collaborative work between Denis Burkitt and Anthony Epstein and is well known to be associated with

several malignancies with an array of established and postulated oncogenic mechanisms [5, 23, 24].

HIV, like EBV, has been implicated in different malignancies. Unlike in other parts of the world, the role of HIV infection in BL in SSA remains unclear, and similar frequencies have been reported in both HIV-infected and non-infected children [3, 16]. However, a higher incidence of non-BL NHL, predominantly DLBCL, has been reported in children who are HIV-positive [25, 26].

Pathology of Endemic Burkitt Lymphoma

The hallmark of BL is the occurrence of a translocation between the *c-myc* oncogene on chromosome 8, and the enhancer regions of immunoglobulin (Ig) genes on chromosomes 14, 2, or 22. A unique biological distinction between endemic and sporadic BL lies in the patterns of chromosomal breakpoint locations, with the vast majority of endemic cases and only a small subset of sporadic cases exhibiting a breakpoint outside of the *Hin*dIII fragment that encompasses the *c-myc* gene [27]. Other biological distinctions between endemic and sporadic disease have also been explored via genomic and proteomic analyses [28–33]. The general pathological characteristics of BL are described in detail in previous chapters on mature B-cell lymphomas.

Although the associations between EBV, malaria, and BL have been long established, their role in the pathogenesis of the disease, and specifically how these infections are linked to the deregulation of C-MYC, is not well defined. However, newer insights suggest that *P. falciparum* infection induces the expression of activation-induced cytidine deaminase (AID)—a DNA-mutating enzyme, mainly expressed in the germinal center of B cells where it is involved in somatic hypermutation and class-switch recombination, both of which are important steps in the generation of antibodies against pathogens [34]. The overexpression of this enzyme can increase the risk of leukemias and lymphomas and has been shown to induce *c-myc*/Ig gene translocations in BL mouse models [35–37].

This aberrant expression of AID alone does not explain the pathogenesis of BL since deregulation of C-MYC in B-cells also alters apoptotic pathways. It has been proposed that EBV-infected B-cells are able to inhibit apoptosis of cells with *c-myc*/Ig gene translocations through their expression of latency phase proteins. Although the precise factors underlying EBV-driven oncogenesis in BL are still unclear, it is typically thought that through disruption of apoptotic pathways, EBV antigens indirectly enable proliferation of lymphocytes that have undergone *c-myc* translocation and malignant transformation [38–44]. Furthermore, malaria infection increases the number of B-cells infected with latent EBV through viral reactivation and increased viral load, a decrease in EBV-specific cytotoxic T-cell response, and induction of *c-myc* translocation through overexpression of AID [45–47].

Clinical Presentation of Endemic Burkitt Lymphoma

Although the classic description of children with endemic BL typically portrays rapidly progressing jaw masses, abdominal masses are an equally important and common clinical presentation in equatorial Africa [16, 48–50]. Jaw masses can cause mass effect, distorting the oral gingiva and displacing the associated teeth, creating the appearance of "floating teeth" within the oral cavity. A significant number of patients present with both facial and abdominal masses [16, 50, 51]. Abdominal masses can arise from any intra-abdominal organ including retroperitoneal nodes, ovaries, intestines, kidneys, and liver. Central nervous system involvement is not infrequent, and up to 20% of patients with BL may present with sudden-onset paraplegia [52, 53]. Peak incidence occurs at 6 years of age, and there is a male-to-female ratio of 2:1 [17].

Sporadic BL in SSA is seen outside the lymphoma belt of Africa (which consists predominantly of east, west, and central Africa) and is often associated with relatively older age at presentation, abdominal masses in the absence of jaw masses, and a higher propensity for bone marrow involvement [19, 25]. Abdominal mass is the predominant presentation for sporadic BL, followed by lymphadenopathy, while jaw involvement is uncommon [54]. Patients can present with severe abdominal pain and small bowel obstruction from intussusception secondary to involvement of the ileocecal valve. Bone marrow involvement has been reported in up to 25% of children with sporadic BL in South Africa—including patients presenting with extensive marrow disease with >25% involvement [25]. Less common sites of presentation of BL include the mediastinum, heart, appendix, testis, breasts, and thyroid glands [17].

Diagnosis of Endemic Burkitt Lymphoma

The presence of a facial mass alone is not sufficient to diagnose BL. Although jaw masses are often associated with BL, other malignancies in childhood including rhabdomyosarcoma can mimic this presentation. Ideally, the diagnosis of BL requires confirmation by morphological and cytogenetic methods to document the *c-myc*/Ig gene translocations. Unfortunately, diagnostic resources are lacking in most centers in SSA, and this imparts significant restrictions on the ability to accurately diagnose BL in these regions [16]. In the absence of pathology resources, a reasonable approach has been to empirically initiate chemotherapy in a patient with a rapidly growing jaw mass (duration of presentation is almost never more than 2 months); because BL is typically extremely sensitive to chemotherapy, a brisk response to chemotherapy may reaffirm a clinical diagnosis; however, this is not always accurate and other diagnoses may be missed.

In cases where clinical suspicion is accompanied by cytological examination through fine needle aspiration (FNA),

restraints on diagnostic accuracy still exist. Diagnostic evaluation via FNA does not provide sufficient tissue for confirmation with immunohistochemical stains, and therefore relies entirely on evaluation of cell morphology. This contrasts with centers in higher income countries where excisional biopsies or imaging-guided core needle biopsies are favored, and morphological diagnosis is confirmed with an array of tests including immunohistochemistry, flow cytometry, and cytogenetic and molecular analyses [55]. A major limitation to FNA-based cytological diagnoses is that a variety of pediatric solid tumors may mimic the appearance of BL with numerous sheets of small–intermediate-sized round blue cells on cytology. Furthermore, there is a striking similarity in the morphological appearance of BL and other non-BL NHL including DLBCL and lymphoblastic lymphoma [16]. The latter requires a different treatment intensity and duration from BL and DLBCL, and outcomes are poor without appropriate therapy [56, 57].

The standard staging approach applied to BL is the St. Jude pediatric NHL staging system [58]. This staging workup should include a bone marrow and cerebrospinal fluid evaluation. Their results inform decisions to intensify the treatment regimen for patients with advanced-stage disease in higher income countries, as higher doses of methotrexate as well as high-dose cytarabine are incorporated into treatment regimens for patients with CNS involvement and/or >25% involvement of the bone marrow. Standard diagnostic imaging in high-income countries is based on computed tomography (CT) to define areas of disease involvement according to the St. Jude staging system. Other radiology modalities such as abdominal ultrasonography, magnetic resonance imaging, and positron emission tomography may also be utilized, but CT imaging is the current gold standard.

Management of Endemic Burkitt Lymphoma

Several iterations of studies ranging from single agent to multidrug combinations have been trialed for endemic BL in SSA. These are summarized in Table 27.3. The over-arching conclusions from these efforts are that while single-agent chemotherapy may be an ineffective treatment approach, the application of contemporary multi-agent intensive regimens similar to what is used in high-income countries is met with remarkably high rates of treatment-related toxicity and even mortality. These experiences are discussed in more detail below and please note that the majority of the contemporary publications utilized intrathecal chemotherapy in conjunction with the systemic chemotherapy regimens described.

Cyclophosphamide Monotherapy

Following the initial descriptions of BL by Denis Burkitt over 50 years ago, survival after single-agent cyclophosphamide had been documented in several reports [59, 60]. Based on multiple studies in a variety of countries, survival outcomes with cyclophosphamide monotherapy have been reported to range from 30% to 50%, with the higher survival rates occurring in cohorts with a greater proportion of patients with limited-stage I/II disease [59–65]. These results are strikingly inferior to contemporary cure rates in high-

Table 27.3 Outcomes of childhood and adolescent Burkitt lymphoma treatment regimens in Sub-Saharan Africa

Location(s)	Years	Sample size, *n*	% HIV+	Survival analysis	Cohort survival	Survival by stage				TRM
						I	I I	I I I	I V	
Cyclophosphamide monotherapy regimens										
Uganda	1967–1970	57	NA	OS	30%	67%		24%	0%	18%
Ghana	1968–1972	110	NA	2-year OS	44%	44%	48%	31%	20%	NR
Malawi	1991–1997	73	NR	OS	55%	64%		33%		NR
GFAOP 6	2005–2008	178	2%	1-year EFS	33%	44%	49%	30%	16%	8%
Cameroon	2006–2008	95	NR	1-year EFS	35%	44%		33%		7%
Cameroon	2008–2009	129	2%	1-year EFS	61%	100%	85%	60%	27%	9%
Cyclophosphamide, vincristine, ± low-dose methotrexate regimens										
Uganda	1985–2005	1217	4%	NR	Mortality risk: stage III/IV HR 4.04, facial tumor only HR 0.33					NR
INCTR	2004–2009	356	5%	1-year EFS/OS	54/67%	Not provided				13%
Malawi	2010–2012	70	3%	1-year EFS	48%	100%	83%	24%	32%	3%
Cyclophosphamide-based regimens plus anthracyclines										
Kenya	2003–2011	428	1%	1-year EFS	31%	Not provided				NR
Malawi	2011–2013	74	4%	18-month OS	29%	49%		26%		8%
Malawi	2012–2014	58	6%	1-year DFS	69%	100%	56%	66%		12%
Malawi	2013–2015	73	3%	18-month OS	29%	51%		28%	17%	16%
Regimens containing high-dose methotrexate										
Malawi	2000–2002	42	2%	1-year EFS	33%	50%	50%	24%	25%	33%
GFAOP 3	2001–2004	187	<1%	3-year OS	56%	64%		52%	36%	21%

income countries that exceed 90% [66]. These data have repeatedly shown that the patients most likely to achieve cure with cyclophosphamide-only chemotherapy regimens are those with stage I/II disease limited to the jaw, and this regimen does not sufficiently induce prolonged disease remission in patients with more advanced-stage disease, which, depending on the cohort, can account for the majority of all patients [49].

Cyclophosphamide, Vincristine, and Low-Dose Methotrexate (COm)

Building upon the experience with single-agent cyclophosphamide, the addition of vincristine and low-dose methotrexate (COm) failed to result in any significant improvement for patients with advanced-stage disease [49]. A multicenter study reported by the INCTR with a regimen combining cyclophosphamide, vincristine, and low-dose (75 mg/m^2) methotrexate resulted in a 1-year event-free survival (EFS) of 54%, with a 1-year overall survival (OS) of 67% [67]. A second-line chemotherapy regimen (including ifosfamide, etoposide, and cytarabine) was built in for those that failed COm, which is reflective of the OS in that study. Additionally, it was limited by inconsistencies in duration of follow-up reported, creating uncertainty regarding the durability of the treatment response and whether the outcomes reported represented short-term response versus long-term remission. Similarly, a different study combining vincristine with the cyclophosphamide backbone in Blantyre, Malawi, demonstrated a 1-year EFS of 48%. Patients with stage III and IV disease experienced EFS of 24% and 32%, respectively [53].

Combination Chemotherapy with Cyclophosphamide plus Anthracyclines

Subsequent attempts to intensify cyclophosphamide monotherapy with the addition of anthracyclines (i.e., with CHOP and CHOP-like regimens containing cyclophosphamide, doxorubicin, vincristine, and prednisone) have not also yielded significant improvements in survival outcomes [16, 51, 68, 69]. Results from a study in Blantyre, Malawi, reported a 66% 2-year disease-free survival (DFS) using a 28-day regimen that added two doses of doxorubicin to the backbone chemotherapy regimen containing cyclophosphamide, vincristine, and prednisone [69]. However, the results may have been limited by selection bias, as a significant proportion of patients who died before initiation of treatment were excluded from the analysis. Furthermore, these outcomes have not been reproduced in other experiences in the region, including a prospective study of six cycles of CHOP in neighboring Lilongwe, Malawi, which reported an 18-month OS of 29% [51]. A larger retrospective cohort of 428 children in Kenya analyzed a regimen similar to that used in Blantyre, except it additionally continued with 24 months of maintenance chemotherapy consisting of low-dose cyclophosphamide and vincristine given monthly [68]. One-year EFS in the Kenyan cohort was 31%. The treatment-related mortality (TRM) of these regimens including anthracyclines was generally higher than the cyclophosphamide monotherapy regimens, ranging from 8% to 22%. This, however, was not high enough to explain the suboptimal outcomes. In a retrospective cohort reported in Lilongwe utilizing CHOP for patients with stage III/IV disease, the TRM was 8%, but the 18-month OS was only 29% [16]. Ultimately, many patients are dying from systemic disease relapse despite the various strategies to intensify the cyclophosphamide chemotherapy backbone [70].

Regimens with High-Dose Methotrexate

The Groupe Franco-Africain d'Oncologie Pédiatrique (GFAOP), a collaboration between Francophone countries in North and West Africa, has attempted incorporating high-dose methotrexate into their BL treatment protocols. The GFAOP experience with high-dose methotrexate (3000 mg/m^2) in combination with cyclophosphamide, vincristine, and prednisone resulted in a 3-year OS of 61%, broken down by a 3-year OS of 56% in the combined cohorts from Cameroon, Madagascar, and Senegal and a 3-year OS of 75% in the combined cohorts from Morocco, Algeria, and Tunisia [71]. This study was initially characterized by high TRM rates; however, through improvements in supportive care, the group demonstrated improved safety outcomes and decreased toxic deaths with each year of the study (TRM first year 26%, second year 19%, and third year 12%). The improvement in supportive care measures resulted in an increase in OS from 54% in the first year to 73% in the third year [71]. As their experience with high-dose methotrexate matures, it may very well set the standard of care for optimizing the chemotherapeutic approach for BL in low-income sites in Africa. The feasibility of delivering high-dose methotrexate (at doses of even 1000 mg/m^2 and above) in low-resource settings is challenged by limitations to provide optimal supportive care including hyperhydration with intravenous fluids, bicarbonate-based oral or intravenous supplementation, capacity to obtain timely and frequent assessments of renal function, and limitations in being able to appropriately monitor drug levels and the trajectory of methotrexate clearance. These challenges were underscored by the discouraging experience incorporating high-dose methotrexate at 2000 mg/m^2 in Blantyre, Malawi, where an exceptionally high TRM of 33% resulted in suboptimal survival outcomes with a 33% 1-year EFS [72].

Rituximab for NHL in LMIC

The Intergroup Study for Children and Adolescents with mature B-cell NHL (B-NHL), a cooperative study between centers in Europe and North America, randomized high-risk B-NHL patients (including BL) to the standard French–

American–British/Lymphomes Malins B (FAB/LMB) backbone regimen with or without the anti-CD20 monoclonal antibody rituximab. Interim analysis demonstrated a 1-year EFS of 94.2% in the rituximab arm versus 81.5% in the control arm, prompting the study to be halted prematurely (investigator letter issued by the Children's Oncology Group and Gustave Roussy, November 2015). Following this observation, rituximab in addition to FAB/LMB or Berlin–Frankfurt–Münster (BFM) backbone chemotherapy is now standard of care for patients with BL in high-income countries. These results reaffirm the published pilot study that initially described the combination of rituximab with combination chemotherapy [73]. Thus, incorporation of rituximab in combination with chemotherapy regimens may represent an exciting treatment option for B-NHL in LMIC.

It should be noted, though, that rituximab as a single agent may not be likely to provide adequate therapy for BL in Africa. Based on the BFM experience delivering a 5-day rituximab-only window phase to newly diagnosed children and adolescents with mature B-NHL (subsequently followed by treatment with the standard BFM regimen), the overall response rate to rituximab was 41% [74]. Broken down further, patients with BL had a 40% response rate compared to 47% for DLBCL, while those with solid tumor lesions had a response rate of 33% compared to 67% for those with bone marrow involvement. Of those patients with a disease response evaluated by a solid lymphoma lesion, only one achieved a complete remission with rituximab [74]. These data suggest that the optimal role for rituximab in LMIC would potentially be in combination with multi-agent chemotherapy, ideally enabling dose reductions of cytotoxic agents to render it more feasible to provide the necessary supportive care.

Critical Role for Adequate Supportive Care

The observations above buttress the crucial impact that the availability of adequate supportive services plays in defining outcomes of childhood cancer patients in LMIC. It would appear that the first step toward improving outcomes in children with NHL and childhood cancer in general in low-resource settings should focus on capacity building and establishment of adequate supportive services, e.g., improved nursing care and pharmacy services, pathology laboratories, blood bank services, nutritional support, and subspecialty support in the form of critical care, palliative care, and surgical expertise among others. Without these basic fundamental pillars of support, it is challenging to build the complex paradigms that contemporary treatment protocols from high-income countries require to optimize curative outcomes.

Reconciling Discrepancies in Outcome Data

A seemingly disparate range of survival outcomes has been reported for similar regimens throughout the region; however, the overall trend across studies illustrates the challenges in curing patients with advanced-stage disease amid the resource limitations in SSA (Table 27.3). Theories that may explain such heterogeneous outcomes include selection bias and variation in staging techniques leading to potentially over-staging patients. While the St. Jude staging system for pediatric NHL is based on CT imaging, abdominal ultrasonography is commonly utilized to determine stage in African centers [58]. Pitfalls in using abdominal ultrasound to "upstage" patients without evidence of abdominal mass on physical exam are illustrated by a study from Cameroon, in which patients rendered stage III by virtue of abdominal involvement identified solely on ultrasound had a 1-year EFS of 64%, contrasting with the 23% EFS for patients defined as stage III by clinical evidence of abdominal involvement [65]. This dramatic difference suggests that there are significant limitations in the use of ultrasonography to define advanced-stage disease presentations.

Non-Burkitt NHL in Sub-Saharan Africa

The spectrum of other childhood and adolescent NHL in SSA is not well described. As descriptions of endemic BL dominate the published literature, there is a paucity of published information describing pathological and clinical characteristics of other NHL as well as overall treatment outcomes. As previously discussed, clinician and pathologist bias may very well be a major contributing factor to the predominance of BL in the epidemiology of NHL in equatorial Africa. A collaborative study between pathologists from the Netherlands and Uganda revealed that although the diagnoses rendered by Ugandan pathologists often agreed with clinical diagnoses, confirmatory pathology analyses performed in the Netherlands (with access to expanded pathology laboratory resources) conflicted in the majority of cases [13]. Ultimately, it is clear that improvements in pathology laboratory resources are a minimum requirement to strengthen the integrity of lymphoma diagnoses in SSA [75]. Starting with a basic spectrum of immunohistochemical stains would provide a critical first step in determining hematological malignancies versus solid tumors and help to differentiate between mature B-NHL (i.e., BL and DLBCL), T-cell lymphoblastic lymphoma, and Hodgkin lymphoma. Additionally, cytogenetic analyses for karyotype or even fluorescent in situ hybridization for the *c-myc*/Ig gene translocation would have a critical impact on strengthening the accuracy of BL diagnoses. Flow cytometry technology would also improve diagnostic accuracy. As fundamental changes occur in strengthening pathology-based resources and expertise in SSA, so will improvements in understanding the true spectrum of disease pathology.

The experience in Lilongwe, Malawi with childhood and adolescent lymphoma describes the diagnostic and therapeutic challenges for patients with NHL [16]. In a cohort of 114 pediatric lymphomas, 65% were BL, 15% lymphoblastic lymphoma, 11% DLBCL, and 18% Hodgkin lymphoma. Access to pathology resources increased over the study period and pathology confirmation of the diagnoses increased from 29% of cases to 60%. This, however, did not have much practical impact on the distribution of lymphoma diagnoses, except for enabling an increase in the proportion of DLBCL diagnoses (from 2% to 10%) [16]. Those patients in the latter period of the study with pathology-confirmed diagnoses of DLBCL were previously being labeled as BL on clinical grounds. Ultimately, the most critical differentiation though is between mature B-NHL and lymphoblastic lymphoma, as the therapeutic approach differs dramatically. In a region where the treatment outcomes for children with acute lymphoblastic leukemia are still dismal, it is logical that patients with lymphoblastic lymphoma experienced unfavorable outcomes while receiving CHOP-based chemotherapy. Tellingly, the 18-month OS for mature B-NHL was 33%, while for lymphoblastic lymphoma there were no survivors, and 92% of those with lymphoblastic disease died from the underlying malignancy [16]. In addition to improving the diagnostic capacity for childhood NHL in the region, continued work to develop disease-specific and risk-stratified treatment protocols for the various NHL diagnoses is paramount to improve overall survival.

The ultimate impact of HIV on non-Burkitt NHL in SSA remains to be seen [76]. Data from South Africa and Malawi have demonstrated that there appears to be an increase in pediatric non-Burkitt NHL diagnoses in the HIV epidemic era, with DLBCL being the disease singled out as increasing in incidence [25, 26]. However, an important question focuses on the impact of HIV on HHV-8 associated hematological malignancies, especially in eastern and central Africa, regions that are of specific interest because they carry the world's highest rates of HHV-8 prevalence [9, 77]. Kaposi sarcoma (KS), the most common HHV-8-associated malignancy, is by far the most common HIV-associated malignancy in children and adults in SSA, accounting for ~90% of all HIV-associated cancers in children in eastern and central Africa [78, 79]. Considering the consequence of high prevalence of both HHV-8 and HIV on the incidence of KS in children in the region, one may hypothesize that other HHV-8-associated malignancies also occur. The most common HHV-8-associated lymphomas/lymphoproliferative disorders include multicentric Castleman disease, primary effusion lymphoma, and plasmablastic lymphoma [80]. As improvements in pathology laboratory infrastructure in Malawi have progressed, a small cohort of adults with HIV-associated multicentric Castleman disease has been discovered [81]. These findings suggest that HHV-8-associated hematological malignancies may be identified in the pediatric population as well.

Childhood and Adolescent NHL in Central and South America

Although a common linguistic and historical backdrop unifies many of the countries in Central and South America, the region is heterogeneous in many aspects and the countries within it range from lower middle-income to high-income levels according to the World Bank classification. The majority of countries in Central and South America are categorized as middle-income and therefore do not encounter the same severe limitations experienced in SSA currently. The medical infrastructure across the region is generally more developed than that established in SSA, and clinical treatment programs for childhood cancer have been initiated much earlier [82]. However, the challenges inherent to treating populations living in extreme poverty are similarly encountered in both Central and South America as well as SSA, and although the limitations in societal and medical resources may not be as drastic, they certainly represent significant obstacles to achieving the curative outcomes currently reported in the United States and Western Europe [82]. The detrimental impact of malnutrition on the treatment of childhood cancer, for example, has been described in a report from Nicaragua [83].

The incidence of childhood and adolescent NHL in Central and South America, although not as high as the rates in SSA, is generally higher than rates reported from the United States and Europe [11]. Epidemiological data from Central America reports that NHL is among the top-five most common childhood malignancies [82]. Although data describing the precise breakdown of pediatric NHL histologies in Central and South America are lacking, reports on NHL in adults from the region demonstrate a similar distribution in comparison with North America [84]. The exceptionally high frequency of extranodal NK/T-cell lymphoma, nasal type (ENKTL), is distinctive and, in a global comparison, renders the frequencies of NHL subtypes more similar to that seen in Asia [84, 85]. Other unique features of NHL in Central and South America include high rates of chronic active EBV, EBV-associated lymphomas, and lymphoproliferative disorders, as well as an intermediate rate of EBV involvement in BL (Table 27.2) [86–90]. This section of the chapter will focus on these unique disease entities that comprise childhood and adolescent NHL in Central and South America as well as feature some of the reported treatment outcomes for pediatric NHL in general.

EBV-Associated Lymphomas in Central and South America

As discussed in the section on endemic BL in SSA, the role of EBV is critical to enabling lymphomagenesis and one that has been extensively explored. One way in which EBV infection in LMIC contrasts with high-income settings is the age at which primary infection occurs. Data from Argentina reveal that nearly 70% of children have experienced primary EBV infection by the age of 2 years, and >80% by age 6 [91]. This contrasts with data from the United States, with an EBV prevalence of only 50% among children between the ages of 6 and 8 years [92]. Although the predominance of endemic BL and its consistent association with EBV in SSA often overshadow the experience in other regions of the world, it is important to point out the distinct role of EBV in a wide variety of childhood lymphomas in Central and South America, including BL. Based on the significant association between EBV and younger children (<10 years of age) with lymphoma, it has been hypothesized that the early age of EBV seroconversion may drive the unique characteristics of EBV-associated NHL in the Central and South America [91].

EBV-Associated Burkitt Lymphoma

Although virtually all cases of endemic BL in SSA are associated with EBV, this association has been reported to be as low as 13% in the United States [93]. In Central and South America, it appears that the association with EBV and BL is at an intermediate level between these two extremes. Data from Brazil revealed that 72–87% of pediatric BL and 53% of combined pediatric and adult BL were found to be EBV positive [94–97]. However, a report from Argentina demonstrated EBV expression in only 37% of pediatric BL cases [98]. Of interest, EBV sequencing profiles from BL tumor specimens comparing patients from Ghana, Brazil, and Argentina documented shared novel nucleotide-base changes in the latent membrane protein-1 promoter and gene across all regions [99]. Ultimately, although it appears that epidemiological variation exists across geographical regions in the world, there may be some common biological aspects to EBV viral oncogenesis in pediatric BL in LMIC.

EBV-Associated T- and NK-cell Lymphomas

Perhaps the most distinctive feature of childhood and adolescent NHL in Central and South America is the disproportionately high occurrence of rare EBV-associated T- and NK-cell lymphomas, a pattern similarly observed in East Asia as well [5]. The three specific EBV-associated malignancies include hydroa vacciniforme-like lymphoproliferative disorder (HV-like LPD), the systemic EBV-positive T-cell lymphoma of childhood, and ENKTL, nasal type [5]. Both HV-like LPD and the systemic EBV-positive T-cell lymphoma of childhood have been associated with chronic active EBV infection, another disease entity that is more prevalent in Central and South America as well as East Asia [86–90]. The explanation for this fascinating geographic and ethnic predilection of disease has never been determined, but the trends have firmly been established. As the clinical experience and translational research for these diseases evolve, hope for improved therapeutic strategies beckons. Currently, these diseases represent a subset of lymphomas with an extremely poor curative prognosis in comparison with other pediatric NHL [5].

Hydroa Vacciniforme-Like Lymphoproliferative Disorder

The understanding of HV-like LPD has evolved over the past decade, including a name change from HV-like lymphoma to HV-like lymphoproliferative disorder [5]. The clinical presentation necessarily involves characteristic skin lesions that may progress from vesiculopapular to ulcerated and to crusted typically leaving scars. Well described in cohorts from Mexico and Peru, skin lesions classically occur on sun-exposed areas of the skin but can involve unexposed areas as well [100–102]. Severe hypersensitivity to mosquito bites can occur as well. Facial edema is typically present, and in some cases, systemic symptoms including fever, lymphadenopathy, and hepatosplenomegaly may be found. The differential diagnosis is broad and includes several different cutaneous lymphomas; but what separates HV-like LPD from the rest is its association with EBV as a cutaneous form of chronic active EBV [5]. The clinical course is often indolent and protracted, and in an illustrative series of 20 children from Mexico, the mean duration of disease at time of clinical presentation to tertiary-level care was 2.4 years [100]. The majority of HV-like LPD cases demonstrate a T-cell phenotype, typically $CD8^+$, but approximately one-third will be NK-cell in derivation [5, 100, 101]. The treatment course is remitting and relapsing, and patients often develop transient disease control with anti-inflammatory or immune-modulating therapies including steroids or thalidomide [100–102]. Long-term complete remission has only been described in the setting of an allogeneic hematopoietic stem cell transplant [103].

Systemic EBV-Positive T-Cell Lymphoma of Childhood

The systemic EBV-positive T-cell lymphoma of childhood is a relatively new diagnostic entity that was first established in the 2008 World Health Organization classification of lymphoid neoplasms [104]. It is characterized by an acute-onset fulminant clinical course presenting with features of an exaggerated systemic inflammatory response including fever, hepatosplenomegaly, pancytopenia, liver dysfunction, and, at times, multi-organ failure [7]. The clinical presentation often fulfills criteria for hemophagocytic lymphohistiocytosis (HLH) in the setting of EBV viremia [105]. Diagnosis can be challenging based on the rarity of the disease, but a

clonal T-cell population can be identified through molecular analysis of the T-cell receptor gene, and EBV is always positive in the infiltrating T-cells on biopsy [5]. The prognosis is dismal, and very few patients achieve sustained disease control let alone cure [7, 105]. The published experience with this disease entity arises from East Asia plus a number of cases occurring in children of Central and South American descent in the United States [7, 90, 105, 106]. As a relatively new diagnostic entity and a pathologic diagnosis that is difficult to determine without subspecialized pathology expertise, it is understandable that published reports of this disease have yet to arise from within Central and South America. However, based upon the experience with children of immigrants in the United States and the overlapping spectrum of EBV-related lymphomas shared by Asia and Latin America, it may be hypothesized that the systemic EBV-positive T-cell lymphoma of childhood is an unrecognized disease entity occurring in Central and South America.

Extranodal NK/T-Cell Lymphoma, Nasal Type

ENKTL is the third EBV-related T/NK-cell lymphoma displaying this distinct epidemiological pattern of occurring in East Asia plus Central and South America. Characterized by a mass presenting in the nasal cavity, nasopharynx, paranasal sinuses, or palate, patients often present with clinical features arising from localized involvement including nasal obstruction, nosebleeds, or persistent discharge. Unlike descriptions of HV-like LPD and the systemic EBV-positive T-cell lymphoma of childhood, which occur primarily in children, ENKTL appears to occur predominately in adults. Data published from Brazil detailing the clinicopathologic features of 122 patients with ENKTL included only three pediatric cases, while a similar report from Peru describing 32 cases of ENKTL also had only three patients under the age of 18 [107, 108]. Although the prognosis for ENKTL is not as dismal as the other two EBV-related T- and NK-cell lymphomas featured in this chapter, survival rates are low for patients with advanced-stage disease [6]. The combination of chemotherapy and radiation therapy is the mainstay of the therapeutic approach [6, 109].

Treatment Outcomes for Pediatric NHL in Central and South America

Comprehensive pediatric oncology programs have been providing treatment for childhood and adolescent NHL in many countries in Central and South America for over 20 years [82]. Although the results vary from country to country based upon a multitude of contributing factors, the evolution of treatment regimens for pediatric NHL in the region has resulted in great progress over the past two decades. Some of the recent efforts to provide curative treatment for NHL in children and adolescents in this region are described and highlight some of the important developments achieved over time (Table 27.4). In comparison to the experience in SSA, the advances in Central and South America are partially attributable to successfully adapting treatment protocols from high-income countries in an effort to optimize treatment response while minimizing toxicity and, at the same time, being able to provide the supportive care required to avoid morbidity and mortality with the adapted regimens.

One of the earliest reports from South America describes the experience in northeastern Brazil from 1980 to 1987 treating predominantly BL, with the majority of patients having advanced-stage disease. Multi-agent chemotherapy regimens were utilized, most often the LSA_2L_2 regimen; however, their analyses demonstrated that the chemotherapy regimen did not have significant effect on EFS [95]. They reported a 5-year EFS of 75% for patients with stage I/II disease, and 42% for those with advanced-stage III/IV disease. Only 39% of patients achieved long-term survival due to high rates of treatment-related mortality from sepsis (25%) as well as treatment abandonment (10%) [95]. Data from Honduras also demonstrated high rates of toxic death utilizing an unmodified LMB89 protocol, with 45% treatment-related mortality in their preliminary experience [110]. Development of modified treatment regimens adapted to match local capacity was a critical adjustment required to improve overall outcomes.

Examples of successfully adapted regimens are highlighted in data from Brazil, Argentina, and Nicaragua. In a single-center study from Rio de Janeiro, Klumb et al. reported outcomes treating mature B-NHL with a modified BFM86/90 protocol in which they reduced high-dose methotrexate from 5 g/m^2 to 2 g/m^2. In a cohort of 53 patients in which 87% presented with advanced-stage disease, they achieved a 78% EFS with a median follow-up time of 35 months [111]. Notably, there was only one sepsis-related death. Similarly, in Argentina, a modified BFM90 regimen also reduced high-dose methotrexate from 5 g/m^2 to 2 g/m^2. Additionally, they reduced high-dose cytarabine from 3 g/m^2/dose to 2 g/m^2/dose for patients that were BFM risk stratified as R4 (highest risk based on CNS involvement or being stage III/IV with elevated lactate dehydrogenase levels). A 5-year EFS of 79% for a cohort of 57 patients was reported [112]. Finally, in Nicaragua, a modified multi-agent regimen was used to treat pediatric NHL including BL, lymphoblastic lymphoma, and DLBCL. Incorporating high-dose methotrexate at 1 g/m^2 for patients with BL, and delivering a 10-week induction phase followed by maintenance therapy to complete 12–18 months (18 for those with lymphoblastic disease), they achieved a 9-year EFS of 53% for a cohort of 53 patients (including 26 with BL) [113]. While these survival rates do not match the approximately 90% EFS currently achieved for patients with mature B-NHL in the

Table 27.4 Outcomes of childhood and adolescent NHL treatment regimens in Central and South America

Location	Years	Sample size, *n*	NHL breakdown	Regimen basis	Key dose adjustments	Survival analysis	Cohort survival	Survival by stage	TRM
Brazil	1980–1987	98	94% BL	Various	Not applicable	Long-term OS	39%	EFS stages I/II 75%, stages III/IV 42%	25%
Brazil	1998–2003	53	89% mature B-NHL	BFM86/90	hdMTX from 5 to 2 g/m^2	~3-year EFS	78%	Stages I/II 100%, stages III/IV 74%	2%
Argentina	1994–1999	57	Mature B-NHL only	BFM90	hdMTX from 5 to 2 g/m^2	5-year EFS	79%	R1 100%, R2 86%, R3 82%, R4 50%	9%
Nicaragua	1996–2003	53	49% BL, 11% DLBCL	Unspecified	hdMTX at 1 g/m^2	9-year EFS	53%	Stage I 100%, II 71%, III 55%, IV 17%	2%
Venezuela	1995–2002	96	Mature B-NHL only	LMB89	Unmodified	2-year EFS	75%	Group A 100%, B 76%, C 56%	9%
AHOPCA	2008–2012	1313	B- and T-cell ALL only	BFM ALL	hdMTX at 2 g/m^2 for SR/IR	3-year EFS	59%	SR 69%, IR 62%, HR 48%	6%
AHOPCA	2000–2013	31	ALCL only	Various	APO versus more intensive	5-year EFS	67%	Localized 100%, advanced 66%	19%

NHL non-Hodgkin lymphoma, *TRM* treatment-related mortality, *BL* Burkitt lymphoma, *OS* overall survival, *EFS* event-free survival, *B-NHL* B-cell NHL, *BFM* Berlin–Frankfurt–Münster consortium, *hdMTX* high-dose methotrexate, *R1-R4* BFM risk stratification groups, *DLBCL* diffuse large B-cell lymphoma, *LMB* French Lymphomes Malins B protocol, *AHOPCA* Asociacion de Hemato-Oncologia de Centroamerica, *ALL* acute lymphoblastic leukemia, *SR/IR/HR* standard, intermediate, and high-risk ALL risk stratification groups, *ALCL* anaplastic large cell lymphoma, *APO* doxorubicin, prednisone, and vincristine

United States and Europe, they nonetheless provide excellent hope for survival for patients in Central and South America as well as benchmark of success for aspiring programs in SSA looking to establish adapted treatment regimens that are deliverable amid local limitations.

Advances in the delivery of complex oncology care to high-risk pediatric NHL patients are highlighted by a few specific publications from Central and South America. Precedence was established in Venezuela to deliver an unmodified LMB89 B-NHL protocol with outstanding success. In this cohort of 96 patients, 83% were stratified as intermediate-risk Group B, and 9% were high-risk Group C. The 2-year EFS was 75% overall, and there was a 9% TRM [114]. In the absence of rasburicase, nine patients developed severe tumor lysis syndrome requiring dialysis, four of whom died from the complications. This complex, high-intensity, multi-agent chemotherapy regimen was delivered with modest toxicity and excellent curative rates despite treating a significant proportion of intermediate-high-risk patients [114].

Another example of significant advances in achieving improved curative outcomes arises from the Asociacion de Hemato-Oncologia de Centroamerica (AHOPCA) collaborative group of five countries in Central America. They devised a risk-stratified regimen for the treatment of acute lymphoblastic leukemia (ALL) adapted from the BFM ALL protocol, including patients with T-cell disease as well. Although patients with lymphoblastic lymphoma were not included, because the treatment approach in high-income countries is similar for ALL and lymphoblastic lymphoma depending on risk stratification and the underlying immunophenotype (i.e., B versus T-cell), the results of this study are very relevant for application to patients with lymphoblastic lymphoma. They delivered a risk-stratified protocol that included induction and consolidation phases, followed by a cycle with four consecutive rounds of high-dose methotrexate (2 g/m^2 for standard and intermediate-risk patients and 5 g/m^2 for high-risk) given every 2 weeks, followed by re-induction and then maintenance therapy. The 3-year EFS was reported as 59%, and the toxic mortality was extrapolated off of the number of early deaths (3%) plus the deaths in first complete remission (3.3%) [115]. Similar to the example from Venezuela, although the curative outcomes are modest in comparison to the approximately 85% cure rate achieved for ALL in high-income countries, this experience establishes an important precedent that complex multi-agent protocols can be delivered safely and effectively for patients with lymphoblastic lymphoma. This contrasts drastically with the experience of treating ALL in SSA, where the EFS is typically <30% [116–118].

A third example of advances in the treatment of childhood and adolescent NHL in Central and South America also comes from the AHOPCA consortium through their experience treating ALCL. The paucity of published data on the treatment of ALCL in LMIC once again reflects the challenges in making a definitive diagnosis as well as in delivering disease-specific therapy. This retrospective analysis of the AHOPCA experience described outcomes based on treatment with one of three regimens—the less intensive APO (doxorubicin, prednisone, and vincristine) regimen, a modified BFM protocol, and a compressed multi-agent T-cell lymphoma protocol developed in Guatemala. While the 5-year EFS was relatively favorable at 67%, increased TRM on the more intensive regimens was reported [119]. However, despite the challenges encountered in treating ALCL in Central America, these data establish an important precedent that disease-specific and risk-stratified regimens can be delivered in LMIC with survival outcomes that approach those achieved in high-income countries.

Conclusions

In conclusion, NHL accounts for a significant proportion of all childhood and adolescent malignancies worldwide. Although improvements have been achieved in Central and South America over the past two decades, there still exists a significant gap in survival rates in comparison with the United States and Europe. Meanwhile, in SSA, the region of the world with by far the highest incidence of childhood and adolescent NHL, the vast majority of children with endemic BL and other lymphomas still experience death from their disease. The disparity in mortality rates for children and adolescents with cancer living in LMIC has prevailed for too long. Focus on improving outcomes for children and adolescents with NHL could serve as the vehicle to improve the overall standard of care and treatment for pediatric cancers in low-income settings and, potentially, help address the severe disparities in socioeconomic and health standards in LMIC.

References

1. Gross TG, Biondi A. Paediatric non-Hodgkin lymphoma in low and middle income countries. Br J Haematol. 2016;173(4):651–4.
2. Burkitt DP. Epidemiology of Burkitt's lymphoma. Proc R Soc Med. 1971;64(9):909–10.
3. Magrath I. Epidemiology: clues to the pathogenesis of Burkitt lymphoma. Br J Haematol. 2012;156(6):744–56.
4. van den Bosch CA. Is endemic Burkitt's lymphoma an alliance between three infections and a tumour promoter? Lancet Oncol. 2004;5(12):738–46.
5. Dojcinov SD, Fend F, Quintanilla-Martinez L. EBV-positive lymphoproliferations of B- T- and NK-cell derivation in non-immunocompromised hosts. Pathogens. 2018;7(1):28.
6. Haverkos BM, Pan Z, Gru AA, Freud AG, Rabinovitch R, Xu-Welliver M, et al. Extranodal NK/T cell lymphoma, nasal type (ENKTL-NT): an update on epidemiology, clinical presentation,

and natural history in North American and European cases. Curr Hematol Malig Rep. 2016;11(6):514–27.
7. Quintanilla-Martinez L, Kumar S, Fend F, Reyes E, Teruya-Feldstein J, Kingma DW, et al. Fulminant EBV(+) T-cell lymphoproliferative disorder following acute/chronic EBV infection: a distinct clinicopathologic syndrome. Blood. 2000;96(2):443–51.
8. Iwanaga M, Watanabe T, Yamaguchi K. Adult T-cell leukemia: a review of epidemiological evidence. Front Microbiol. 2012;3:322.
9. Dollard SC, Butler LM, Jones AM, Mermin JH, Chidzonga M, Chipato T, et al. Substantial regional differences in human herpesvirus 8 seroprevalence in sub-Saharan Africa: insights on the origin of the "Kaposi's sarcoma belt". Int J Cancer. 2010;127(10):2395–401.
10. Orem J, Otieno MW, Remick SC. AIDS-associated cancer in developing nations. Curr Opin Oncol. 2004;16(5):468–76.
11. Linet MS, Brown LM, Mbulaiteye SM, Check D, Ostroumova E, Landgren A, et al. International long-term trends and recent patterns in the incidence of leukemias and lymphomas among children and adolescents ages 0-19 years. Int J Cancer. 2016;138(8):1862–74.
12. Burkitt D, O'Conor GT. Malignant lymphoma in African children. I. A clinical syndrome. Cancer. 1961;14:258–69.
13. Orem J, Sandin S, Weibull CE, Odida M, Wabinga H, Mbidde E, et al. Agreement between diagnoses of childhood lymphoma assigned in Uganda and by an international reference laboratory. Clin Epidemiol. 2012;4:339–47.
14. Ogwang MD, Zhao W, Ayers LW, Mbulaiteye SM. Accuracy of Burkitt lymphoma diagnosis in constrained pathology settings: importance to epidemiology. Arch Pathol Lab Med. 2011;135(4):445–50.
15. Naresh KN, Raphael M, Ayers L, Hurwitz N, Calbi V, Rogena E, et al. Lymphomas in sub-Saharan Africa--what can we learn and how can we help in improving diagnosis, managing patients and fostering translational research? Br J Haematol. 2011;154(6):696–703.
16. El-Mallawany NK, Mutai M, Mtete I, Gopal S, Stanley CC, Wasswa P, et al. Beyond Endemic Burkitt Lymphoma: navigating challenges of differentiating childhood lymphoma diagnoses amid limitations in pathology resources in Lilongwe, Malawi. Glob Pediatr Health. 2017;4:2333794X17715831.
17. Molyneux EM, Rochford R, Griffin B, Newton R, Jackson G, Menon G, et al. Burkitt's lymphoma. Lancet. 2012;379(9822):1234–44.
18. Moormann AM, Bailey JA. Malaria - how this parasitic infection aids and abets EBV-associated Burkitt lymphomagenesis. Curr Opin Virol. 2016;20:78–84.
19. Stefan DC, Lutchman R. Burkitt lymphoma: epidemiological features and survival in a South African centre. Infect Agents Cancer. 2014;9:19.
20. Piriou E, Asito AS, Sumba PO, Fiore N, Middeldorp JM, Moormann AM, et al. Early age at time of primary Epstein-Barr virus infection results in poorly controlled viral infection in infants from Western Kenya: clues to the etiology of endemic Burkitt lymphoma. J Infect Dis. 2012;205(6):906–13.
21. Reynaldi A, Schlub TE, Chelimo K, Sumba PO, Piriou E, Ogolla S, et al. Impact of Plasmodium falciparum coinfection on longitudinal Epstein-Barr virus kinetics in Kenyan children. J Infect Dis. 2016;213(6):985–91.
22. Lam KM, Syed N, Whittle H, Crawford DH. Circulating Epstein-Barr virus-carrying B cells in acute malaria. Lancet. 1991;337(8746):876–8.
23. Epstein MA, Achong BG, Barr YM. Virus particles in cultured lymphoblasts from Burkitt's lymphoma. Lancet. 1964;1(7335):702–3.
24. Epstein A. Burkitt lymphoma and the discovery of Epstein-Barr virus. Br J Haematol. 2012;156(6):777–9.
25. Davidson A, Hendricks M. Experience with B-cell lymphoma at a South African centre in the HIV Era. Transfus Apher Sci. 2013;49(1):31–9.
26. Mutalima N, Molyneux EM, Johnston WT, Jaffe HW, Kamiza S, Borgstein E, et al. Impact of infection with human immunodeficiency virus-1 (HIV) on the risk of cancer among children in Malawi - preliminary findings. Infect Agents Cancer. 2010;5(5):1–6.
27. Shiramizu B, Barriga F, Neequaye J, Jafri A, Dalla-Favera R, Neri A, et al. Patterns of chromosomal breakpoint locations in Burkitt's lymphoma: relevance to geography and Epstein-Barr virus association. Blood. 1991;77(7):1516–26.
28. Piccaluga PP, De Falco G, Kustagi M, Gazzola A, Agostinelli C, Tripodo C, et al. Gene expression analysis uncovers similarity and differences among Burkitt lymphoma subtypes. Blood. 2011;117(13):3596–608.
29. Lenze D, Leoncini L, Hummel M, Volinia S, Liu CG, Amato T, et al. The different epidemiologic subtypes of Burkitt lymphoma share a homogenous micro RNA profile distinct from diffuse large B-cell lymphoma. Leukemia. 2011;25(12):1869–76.
30. Kaymaz Y, Oduor CI, Yu H, Otieno JA, Ong'echa JM, Moormann AM, et al. Comprehensive transcriptome and mutational profiling of endemic Burkitt lymphoma reveals EBV type-specific Differences. Mol Cancer Res. 2017;15(5):563–76.
31. Abate F, Ambrosio MR, Mundo L, Laginestra MA, Fuligni F, Rossi M, et al. Distinct viral and mutational spectrum of endemic Burkitt lymphoma. PLoS Pathog. 2015;11(10):e1005158.
32. Amato T, Abate F, Piccaluga P, Iacono M, Fallerini C, Renieri A, et al. Clonality analysis of immunoglobulin gene rearrangement by next-generation sequencing in endemic Burkitt lymphoma suggests antigen drive activation of BCR as opposed to sporadic Burkitt lymphoma. Am J Clin Pathol. 2016;145(1):116–27.
33. El-Mallawany NK, Day N, Ayello J, Van de Ven C, Conlon K, Fermin D, et al. Differential proteomic analysis of endemic and sporadic Epstein-Barr virus-positive and negative Burkitt lymphoma. Eur J Cancer. 2015;51(1):92–100.
34. Thorley-Lawson D, Deitsch KW, Duca KA, Torgbor C. The link between Plasmodium falciparum malaria and endemic Burkitt's lymphoma-new insight into a 50-year-old enigma. PLoS Pathog. 2016;12(1):e1005331.
35. Park SR. Activation-induced cytidine deaminase in B Cell immunity and cancers. Immune Netw. 2012;12(6):230–9.
36. Ramiro AR, Jankovic M, Eisenreich T, Difilippantonio S, Chen-Kiang S, Muramatsu M, et al. AID is required for c-myc/IgH chromosome translocations in vivo. Cell. 2004;118(4):431–8.
37. Ramiro AR, Jankovic M, Callen E, Difilippantonio S, Chen HT, McBride KM, et al. Role of genomic instability and p53 in AID-induced c-myc-Igh translocations. Nature. 2006;440(7080):105–9.
38. Clybouw C, McHichi B, Mouhamad S, Auffredou MT, Bourgeade MF, Sharma S, et al. EBV infection of human B lymphocytes leads to down-regulation of Bim expression: relationship to resistance to apoptosis. J Immunol. 2005;175(5):2968–73.
39. Kelly GL, Milner AE, Baldwin GS, Bell AI, Rickinson AB. Three restricted forms of Epstein-Barr virus latency counteracting apoptosis in c-myc-expressing Burkitt lymphoma cells. Proc Natl Acad Sci U S A. 2006;103(40):14935–40.
40. Kelly GL, Milner AE, Tierney RJ, Croom-Carter DS, Altmann M, Hammerschmidt W, et al. Epstein-Barr virus nuclear antigen 2 (EBNA2) gene deletion is consistently linked with EBNA3A, -3B, and -3C expression in Burkitt's lymphoma cells and with increased resistance to apoptosis. J Virol. 2005;79(16):10709–17.
41. Kennedy G, Komano J, Sugden B. Epstein-Barr virus provides a survival factor to Burkitt's lymphomas. Proc Natl Acad Sci U S A. 2003;100(24):14269–74.

42. Komano J, Maruo S, Kurozumi K, Oda T, Takada K. Oncogenic role of Epstein-Barr virus-encoded RNAs in Burkitt's lymphoma cell line Akata. J Virol. 1999;73(12):9827–31.
43. Nanbo A, Inoue K, Adachi-Takasawa K, Takada K. Epstein-Barr virus RNA confers resistance to interferon-alpha-induced apoptosis in Burkitt's lymphoma. EMBO J. 2002;21(5):954–65.
44. Ruf IK, Rhyne PW, Yang C, Cleveland JL, Sample JT. Epstein-Barr virus small RNAs potentiate tumorigenicity of Burkitt lymphoma cells independently of an effect on apoptosis. J Virol. 2000;74(21):10223–8.
45. Njie R, Bell AI, Jia H, Croom-Carter D, Chaganti S, Hislop AD, et al. The effects of acute malaria on Epstein-Barr virus (EBV) load and EBV-specific T cell immunity in Gambian children. J Infect Dis. 2009;199(1):31–8.
46. Whittle HC, Brown J, Marsh K, Greenwood BM, Seidelin P, Tighe H, et al. T-cell control of Epstein-Barr virus-infected B cells is lost during P. falciparum malaria. Nature. 1984;312(5993):449–50.
47. Moormann AM, Chelimo K, Sumba OP, Lutzke ML, Ploutz-Snyder R, Newton D, et al. Exposure to holoendemic malaria results in elevated Epstein-Barr virus loads in children. J Infect Dis. 2005;191(8):1233–8.
48. Ogwang MD, Bhatia K, Biggar RJ, Mbulaiteye SM. Incidence and geographic distribution of endemic Burkitt lymphoma in northern Uganda revisited. Int J Cancer. 2008;123(11):2658–63.
49. Orem J, Mulumba Y, Algeri S, Bellocco R, Mangen FW, Mbidde EK, et al. Clinical characteristics, treatment and outcome of childhood Burkitt's lymphoma at the Uganda Cancer Institute. Trans R Soc Trop Med Hyg. 2011;105(12):717–26.
50. Offor UT, Akyea RK, Neequaye JE, Renner LA, Segbefia CI. The changing clinical pattern of endemic Burkitt lymphoma in Western Africa: experience from a tertiary center in Ghana. Pediatr Blood Cancer. 2018;65:e27275.
51. Stanley CC, Westmoreland KD, Heimlich BJ, El-Mallawany NK, Wasswa P, Mtete I, et al. Outcomes for paediatric Burkitt lymphoma treated with anthracycline-based therapy in Malawi. Br J Haematol. 2016;173(5):705–12.
52. Walusansa V, Okuku F, Orem J. Burkitt lymphoma in Uganda, the legacy of Denis Burkitt and an update on the disease status. Br J Haematol. 2012;156(6):757–60.
53. Depani S, Banda K, Bailey S, Israels T, Chagaluka G, Molyneux E. Outcome is unchanged by adding vincristine upfront to the Malawi 28-day protocol for endemic Burkitt lymphoma. Pediatr Blood Cancer. 2015;62(11):1929–34.
54. El-Mallawany NK, Cairo MS. Advances in the diagnosis and treatment of childhood and adolescent B-cell non-Hodgkin lymphoma. Clin Adv Hematol Oncol. 2015;13(2):113–23.
55. Miles RR, Arnold S, Cairo MS. Risk factors and treatment of childhood and adolescent Burkitt lymphoma/leukaemia. Br J Haematol. 2012;156(6):730–43.
56. Anderson JR, Jenkin RD, Wilson JF, Kjeldsberg CR, Sposto R, Chilcote RR, et al. Long-term follow-up of patients treated with COMP or LSA2L2 therapy for childhood non-Hodgkin's lymphoma: a report of CCG-551 from the Childrens Cancer Group. J Clin Oncol Off J Am Soc Clin Oncol. 1993;11(6):1024–32.
57. Tubergen DG, Krailo MD, Meadows AT, Rosenstock J, Kadin M, Morse M, et al. Comparison of treatment regimens for pediatric lymphoblastic non-Hodgkin's lymphoma: a Childrens Cancer Group study. J Clin Oncol Off J Am Soc Clin Oncol. 1995;13(6):1368–76.
58. Murphy SB. Classification, staging and end results of treatment of childhood non-Hodgkin's lymphomas: dissimilarities from lymphomas in adults. Semin Oncol. 1980;7(3):332–9.
59. Burkitt D, Hutt MS, Wright DH. The African lymphoma: preliminary observations on response to therapy. Cancer. 1965;18:399–410.
60. Ziegler JL, Morrow RH Jr, Fass L, Kyalwazi SK, Carbone PP. Treatment of Burkitt's tumor with cyclophosphamide. Cancer. 1970;26(2):474–84.
61. Nkrumah FK, Perkins IV. Burkitt's lymphoma: a clinical study of 110 patients. Cancer. 1976;37(2):671–6.
62. Olweny CL, Katongole-Mbidde E, Otim D, Lwanga SK, Magrath IT, Ziegler JL. Long-term experience with Burkitt's lymphoma in Uganda. Int J Cancer. 1980;26(3):261–6.
63. Kazembe P, Hesseling PB, Griffin BE, Lampert I, Wessels G. Long term survival of children with Burkitt lymphoma in Malawi after cyclophosphamide monotherapy. Med Pediatr Oncol. 2003;40(1):23–5.
64. Traore F, Coze C, Atteby JJ, Andre N, Moreira C, Doumbe P, et al. Cyclophosphamide monotherapy in children with Burkitt lymphoma: a study from the French-African Pediatric Oncology Group (GFAOP). Pediatr Blood Cancer. 2011;56(1):70–6.
65. Marjerrison S, Fernandez CV, Price VE, Njume E, Hesseling P. The use of ultrasound in endemic Burkitt lymphoma in Cameroon. Pediatr Blood Cancer. 2012;58(3):352–5.
66. Cairo MS, Sposto R, Gerrard M, Auperin A, Goldman SC, Harrison L, et al. Advanced stage, increased lactate dehydrogenase, and primary site, but not adolescent age (≥ 15 years), are associated with an increased risk of treatment failure in children and adolescents with mature B-cell non-Hodgkin's lymphoma: results of the FAB LMB 96 study. J Clin Oncol Off J Am Soc Clin Oncol. 2012;30(4):387–93.
67. Ngoma T, Adde M, Durosinmi M, Githang'a J, Aken'Ova Y, Kaijage J, et al. Treatment of Burkitt lymphoma in equatorial Africa using a simple three-drug combination followed by a salvage regimen for patients with persistent or recurrent disease. Br J Haematol. 2012;158(6):749–62.
68. Buckle G, Maranda L, Skiles J, Ong'echa JM, Foley J, Epstein M, et al. Factors influencing survival among Kenyan children diagnosed with endemic Burkitt lymphoma between 2003 and 2011: a historical cohort study. Int J Cancer. 2016;139(6):1231–40.
69. Molyneux E, Schwalbe E, Chagaluka G, Banda K, Israels T, Depani S, et al. The use of anthracyclines in the treatment of endemic Burkitt lymphoma. Br J Haematol. 2017;177(6):984–90.
70. Westmoreland KD, El-Mallawany NK, Kazembe P, Stanley CC, Gopal S. Dissecting heterogeneous outcomes for paediatric Burkitt lymphoma in Malawi after anthracycline-based treatment. Br J Haematol. 2017;181:853–4.
71. Harif M, Barsaoui S, Benchekroun S, Bouhas R, Doumbe P, Khattab M, et al. Treatment of B-cell lymphoma with LMB modified protocols in Africa--report of the French-African Pediatric Oncology Group (GFAOP). Pediatr Blood Cancer. 2008;50(6):1138–42.
72. Hesseling P, Broadhead R, Mansvelt E, Louw M, Wessels G, Borgstein E, et al. The 2000 Burkitt lymphoma trial in Malawi. Pediatr Blood Cancer. 2005;44(3):245–50.
73. Goldman S, Smith L, Anderson JR, Perkins S, Harrison L, Geyer MB, et al. Rituximab and FAB/LMB 96 chemotherapy in children with Stage III/IV B-cell non-Hodgkin lymphoma: a Children's Oncology Group report. Leukemia. 2013;27(5):1174–7.
74. Meinhardt A, Burkhardt B, Zimmermann M, Borkhardt A, Kontny U, Klingebiel T, et al. Phase II window study on rituximab in newly diagnosed pediatric mature B-cell non-Hodgkin's lymphoma and Burkitt leukemia. J Clin Oncol Off J Am Soc Clin Oncol. 2010;28(19):3115–21.
75. Montgomery ND, Liomba NG, Kampani C, Krysiak R, Stanley CC, Tomoka T, et al. Accurate real-time diagnosis of lymphoproliferative disorders in Malawi through clinicopathologic teleconferences: a model for pathology services in sub-Saharan Africa. Am J Clin Pathol. 2016;146(4):423–30.

76. Rees CA, Keating EM, Lukolyo H, Danysh HE, Scheurer ME, Mehta PS, et al. Mapping the epidemiology of Kaposi Sarcoma and non-Hodgkin lymphoma among children in sub-Saharan Africa: a review. Pediatr Blood Cancer. 2016;63(8):1325–31.
77. Martro E, Bulterys M, Stewart JA, Spira TJ, Cannon MJ, Thacher TD, et al. Comparison of human herpesvirus 8 and Epstein-Barr virus seropositivity among children in areas endemic and non-endemic for Kaposi's sarcoma. J Med Virol. 2004;72(1):126–31.
78. Tukei V, Kekitiinwa A, Beasley R. Prevalence and outcome of HIV-associated malignancis among children. AIDS. 2011;25:1789–93.
79. El-Mallawany NK, Wasswa P, Mtete I, Mutai M, Stanley CC, Mtunda M, et al. Identifying opportunities to bridge disparity gaps in curing childhood cancer in Malawi: malignancies with excellent curative potential account for the majority of diagnoses. Pediatr Hematol Oncol. 2017;34(5):261–74.
80. Chadburn A, Said J, Gratzinger D, Chan JK, de Jong D, Jaffe ES, et al. HHV8/KSHV-positive lymphoproliferative disorders and the spectrum of plasmablastic and plasma cell neoplasms: 2015 SH/EAHP workshop report-part 3. Am J Clin Pathol. 2017;147(2):171–87.
81. Gopal S, Liomba NG, Montgomery ND, Moses A, Kaimila B, Nyasosela R, et al. Characteristics and survival for HIV-associated multicentric Castleman disease in Malawi. J Int AIDS Soc. 2015;18:20122.
82. Valsecchi MG, Tognoni G, Bonilla M, Moreno N, Baez F, Pacheco C, et al. Clinical epidemiology of childhood cancer in Central America and Caribbean countries. Ann Oncol. 2004;15(4):680–5.
83. Pribnow AK, Ortiz R, Baez LF, Mendieta L, Luna-Fineman S. Effects of malnutrition on treatment-related morbidity and survival of children with cancer in Nicaragua. Pediatr Blood Cancer. 2017;64(11):e26590.
84. Laurini JA, Perry AM, Boilesen E, Diebold J, Maclennan KA, Muller-Hermelink HK, et al. Classification of non-Hodgkin lymphoma in Central and South America: a review of 1028 cases. Blood. 2012;120(24):4795–801.
85. Perry AM, Molina-Kirsch H, Nathwani BN, Diebold J, Maclennan KA, Muller-Hermelink HK, et al. Classification of non-Hodgkin lymphomas in Guatemala according to the World Health Organization system. Leuk Lymphoma. 2011;52(9):1681–8.
86. Cohen JI, Kimura H, Nakamura S, Ko YH, Jaffe ES. Epstein-Barr virus-associated lymphoproliferative disease in non-immunocompromised hosts: a status report and summary of an international meeting, 8-9 September 2008. Ann Oncol. 2009;20(9):1472–82.
87. Cohen JI, Jaffe ES, Dale JK, Pittaluga S, Heslop HE, Rooney CM, et al. Characterization and treatment of chronic active Epstein-Barr virus disease: a 28-year experience in the United States. Blood. 2011;117(22):5835–49.
88. Kimura H, Cohen JI. Chronic active Epstein-Barr virus disease. Front Immunol. 2017;8:1867.
89. Bollard CM, Cohen JI. How I treat T cell chronic active Epstein barr virus disease. Blood. 2018;131:2899.
90. Ohshima K, Kimura H, Yoshino T, Kim CW, Ko YH, Lee SS, et al. Proposed categorization of pathological states of EBV-associated T/natural killer-cell lymphoproliferative disorder (LPD) in children and young adults: overlap with chronic active EBV infection and infantile fulminant EBV T-LPD. Pathol Int. 2008;58(4):209–17.
91. Chabay PA, Preciado MV. EBV primary infection in childhood and its relation to B-cell lymphoma development: a mini-review from a developing region. Int J Cancer. 2013;133(6):1286–92.
92. Balfour HH Jr, Sifakis F, Sliman JA, Knight JA, Schmeling DO, Thomas W. Age-specific prevalence of Epstein-Barr virus infection among individuals aged 6-19 years in the United States and factors affecting its acquisition. J Infect Dis. 2013;208(8):1286–93.
93. Mbulaiteye SM, Pullarkat ST, Nathwani BN, Weiss LM, Rao N, Emmanuel B, et al. Epstein-Barr virus patterns in US Burkitt lymphoma tumors from the SEER residual tissue repository during 1979-2009. APMIS. 2014;122(1):5–15.
94. Araujo I, Foss HD, Bittencourt A, Hummel M, Demel G, Mendonca N, et al. Expression of Epstein-Barr virus-gene products in Burkitt's lymphoma in Northeast Brazil. Blood. 1996;87(12):5279–86.
95. Sandlund JT, Fonseca T, Leimig T, Verissimo L, Ribeiro R, Lira V, et al. Predominance and characteristics of Burkitt lymphoma among children with non-Hodgkin lymphoma in northeastern Brazil. Leukemia. 1997;11(5):743–6.
96. Queiroga EM, Gualco G, Weiss LM, Dittmer DP, Araujo I, Klumb CE, et al. Burkitt lymphoma in Brazil is characterized by geographically distinct clinicopathologic features. Am J Clin Pathol. 2008;130(6):946–56.
97. Klumb CE, Hassan R, De Oliveira DE, De Resende LM, Carrico MK, De Almeida Dobbin J, et al. Geographic variation in Epstein-Barr virus-associated Burkitt's lymphoma in children from Brazil. Int J Cancer. 2004;108(1):66–70.
98. Lara J, Cohen M, De Matteo E, Aversa L, Preciado MV, Chabay P. Epstein-Barr virus (EBV) association and latency profile in pediatric Burkitt's lymphoma: experience of a single institution in Argentina. J Med Virol. 2014;86(5):845–50.
99. Lei H, Li T, Li B, Tsai S, Biggar RJ, Nkrumah F, et al. Epstein-Barr virus from Burkitt Lymphoma biopsies from Africa and South America share novel LMP-1 promoter and gene variations. Sci Rep. 2015;5:16706.
100. Quintanilla-Martinez L, Ridaura C, Nagl F, Saez-de-Ocariz M, Duran-McKinster C, Ruiz-Maldonado R, et al. Hydroa vacciniforme-like lymphoma: a chronic EBV+ lymphoproliferative disorder with risk to develop a systemic lymphoma. Blood. 2013;122(18):3101–10.
101. Rodriguez-Pinilla SM, Barrionuevo C, Garcia J, Martinez MT, Pajares R, Montes-Moreno S, et al. EBV-associated cutaneous NK/T-cell lymphoma: review of a series of 14 cases from peru in children and young adults. Am J Surg Pathol. 2010;34(12):1773–82.
102. Barrionuevo C, Anderson VM, Zevallos-Giampietri E, Zaharia M, Misad O, Bravo F, et al. Hydroa-like cutaneous T-cell lymphoma: a clinicopathologic and molecular genetic study of 16 pediatric cases from Peru. Appl Immunohistochem Mol Morphol. 2002;10(1):7–14.
103. El-Mallawany NK, Geller L, Bollard CM, Wistinghausen B, Mussai F, Wayne AS, et al. Long-term remission in a child with refractory EBV(+) hydroa vacciniforme-like T-cell lymphoma through sequential matched EBV(+)-related allogeneic hematopoietic SCT followed by donor-derived EBV-specific cytotoxic T-lymphocyte immunotherapy. Bone Marrow Transplant. 2011;46(5):759–61.
104. Jaffe ES, Harris NL, Stein H, Isaacson PG. Classification of lymphoid neoplasms: the microscope as a tool for disease discovery. Blood. 2008;112(12):4384–99.
105. Smith MC, Cohen DN, Greig B, Yenamandra A, Vnencak-Jones C, Thompson MA, et al. The ambiguous boundary between EBV-related hemophagocytic lymphohistiocytosis and systemic EBV-driven T cell lymphoproliferative disorder. Int J Clin Exp Pathol. 2014;7(9):5738–49.
106. Sevilla DW, El-Mallawany NK, Emmons FN, Alexander S, Bhagat G, Alobeid B. Spectrum of childhood Epstein-Barr virus-associated T-cell proliferations and bone marrow findings. Pediatr Dev Pathol. 2011;14(1):28–37.
107. Gualco G, Domeny-Duarte P, Chioato L, Barber G, Natkunam Y, Bacchi CE. Clinicopathologic and molecular features of 122 Brazilian cases of nodal and extranodal NK/T-cell lymphoma,

nasal type, with EBV subtyping analysis. Am J Surg Pathol. 2011;35(8):1195–203.
108. Quintanilla-Martinez L, Franklin JL, Guerrero I, Krenacs L, Naresh KN, Rama-Rao C, et al. Histological and immunophenotypic profile of nasal NK/T cell lymphomas from Peru: high prevalence of p53 overexpression. Hum Pathol. 1999;30(7):849–55.
109. Yamaguchi M, Suzuki R, Oguchi M. Advances in the treatment of extranodal NK/T-cell lymphoma, nasal type. Blood. 2018;131(23):2528–40.
110. Howard SC, Ortiz R, Baez LF, Cabanas R, Barrantes J, Fu L, et al. Protocol-based treatment for children with cancer in low income countries in Latin America: a report on the recent meetings of the Monza International School of Pediatric Hematology/Oncology (MISPHO)--part II. Pediatr Blood Cancer. 2007;48(4):486–90.
111. Klumb CE, Schramm MT, De Resende LM, Carrico MK, Coelho AM, de Meis E, et al. Treatment of children with B-cell non-Hodgkin's lymphoma in developing countries: the experience of a single center in Brazil. J Pediatr Hematol Oncol. 2004;26(7):462–8.
112. Chantada G, Casak S, Alderete D, Zubizarreta P, Gallo G, Muriel FS. Treatment of B-cell malignancies in children with a modified BFM-NHL 90 protocol in Argentina. Med Pediatr Oncol. 2003;41(5):488–90.
113. Baez F, Pillon M, Manfredini L, Ocampo E, Mendez G, Ortiz R, et al. Treatment of pediatric non-Hodgkin lymphomas in a country with limited resources: results of the first national protocol in Nicaragua. Pediatr Blood Cancer. 2008;50(1):148–52.
114. Acquatella G, Insausti CL, Garcia R, Gomez R, Hernandez M, Carneiro M, et al. Outcome of children with B cell lymphoma in Venezuela with the LMB-89 protocol. Pediatr Blood Cancer. 2004;43(5):580–6.
115. Navarrete M, Rossi E, Brivio E, Carrillo JM, Bonilla M, Vasquez R, et al. Treatment of childhood acute lymphoblastic leukemia in central America: a lower-middle income countries experience. Pediatr Blood Cancer. 2014;61(5):803–9.
116. Chagaluka G, Carey P, Banda K, Schwab C, Chilton L, Schwalbe E, et al. Treating childhood acute lymphoblastic leukemia in Malawi. Haematologica. 2013;98(1):e1–3.
117. Kersten E, Scanlan P, Dubois SG, Matthay KK. Current treatment and outcome for childhood acute leukemia in Tanzania. Pediatr Blood Cancer. 2013;60(12):2047–53.
118. Rubagumya F, Xu MJ, May L, Driscoll C, Uwizeye FR, Shyirambere C, et al. Outcomes of low-intensity treatment of acute lymphoblastic leukemia at Butaro Cancer Center of Excellence in Rwanda. J Glob Oncol. 2018;2018(4):1–11.
119. Ceppi F, Ortiz R, Antillon F, Vasquez R, Gomez W, Gamboa J, et al. Anaplastic large cell lymphoma in Central America: a report from the Central American Association of Pediatric Hematology Oncology (AHOPCA). Pediatr Blood Cancer. 2016;63(1):78–82.

28 Long-Term Outcomes in Survivors of Childhood and Adolescent Non-Hodgkin Lymphoma

Paul C. Nathan, Karin P. S. Langenberg-Ververgaert, and Noelle Cullinan

Introduction

Among children and adolescents (0–19 years old) treated for Non-Hodgkin lymphoma (NHL) in the United States, 5-year survival has increased from 45% in those diagnosed between 1975 and 1977 to 89% in those diagnosed between 2007 and 2013 [1]. Consequently, there is a growing population of long-term survivors of NHL. In 2011, there were an estimated 23,708 survivors of childhood NHL alive in the United States, 60% of whom had survived 15 or more years from their primary cancer [2].

Unfortunately, the very therapies that have resulted in improved survival of NHL can cause long-term physical, psychological, and psychosocial morbidities. These "late effects" of cancer therapy can develop during cancer treatment or may only manifest years to decades after the initial cancer. As a consequence, all survivors of childhood NHL require lifelong medical care that is adapted to the specific risks stemming from their cancer treatment. The causes of late effects in NHL survivors are multifactorial. The multimodal cancer therapies used to treat NHL are the primary cause of late morbidity. Among these, radiation is frequently implicated because it impacts not only tumor tissue but also the surrounding healthy tissue, potentially affecting growth, organ development and function, and increasing the risk for subsequent cancers. Several classes of chemotherapy agents (e.g., anthracyclines, alkylating agents, and epipodophyllotoxins) have specific long-term toxicities—these are described in detail below. Occasionally, the location of the primary tumor, or the surgery needed to biopsy or resect it, can lead to late morbidities. In the few patients with NHL who require hematopoietic stem cell transplantation (HSCT), the intense conditioning regimen can compound the late effects arising from their prior therapy. Beyond the physical sequelae of lymphoma therapy, the cancer experience can have a long-term impact on mental health and psychosocial outcomes. Increasingly, the financial impact of being treated for and subsequently living with the long-term effects of cancer therapy has been recognized. Once survivors of childhood NHL reach adulthood, this financial toxicity can include high out-of-pocket medical costs for ongoing care or delay of required medical care because of cost, asset depletion and bankruptcy, limitations or inability to work, and job lock, where a survivor is forced to stay in their current job in order to retain their insurance coverage [3].

In this chapter, we review the existing research regarding late effects in survivors of childhood NHL. We will explore the most common and morbid late effects seen in these patients and discuss the lifelong care and support that all survivors require in order to minimize their long-term morbidities and maximize their health-related quality of life (HRQoL).

Evolution of Therapy for Children with NHL

Marked improvements in long-term survival of NHL have been accomplished with the use of multimodal therapy, tailored to the histologic subtype and clinical stage of disease. International collaborative clinical trials have focused not only on improving outcome but also on reducing long-term morbidities in survivors of childhood NHL by omitting radiotherapy, modifying central nervous system (CNS)-directed therapy to reduce or eliminate cranial radiation,

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and decreasing cumulative doses of alkylating agents, anthracyclines, and methotrexate.

Therapy for Mature B-Cell Non-Hodgkin Lymphoma

Mature B-cell NHL (Burkitt and Burkitt-like lymphoma/leukemia, diffuse large B-cell lymphoma, and primary mediastinal B-cell lymphoma) is aggressive and requires an intensive treatment regimen. As survival rates have improved since the mid-1980s, routine use of radiotherapy has been omitted, and the duration of chemotherapy treatment has been shortened. More recently, the anti-CD20 monoclonal antibody, rituximab, has been added to therapeutic regimens. Table 28.1 displays the chemotherapy agents and doses used in many of the more recent clinical trials [4–6].

Therapy for Anaplastic Large Cell Lymphoma (ALCL)

Since ALCL was not recognized as a distinct entity until the late 1980s, most early treatment protocols were based on those used for mature B-cell NHL lymphomas. Therapy can include surgical resection (in low stage disease) and chemotherapy regimens that incorporate corticosteroids, anthracyclines, alkylating agents, epipodophyllotoxins, and methotrexate (MTX) (Table 28.2). CNS disease at diagnosis is exceptionally rare. When needed, CNS-directed therapy involves multiagent chemotherapy, including high-dose MTX (HD-MTX), cytarabine, and intrathecal treatment. Short-pulse HD-MTX without intrathecal injections has been established as the optimal way to deliver MTX in order to minimize toxicity [7–9]. There is no routine role for cranial radiation [10].

Therapy for Lymphoblastic Lymphoma

The World Health Organization (WHO) classifies lymphoblastic lymphoma as the same disease entity as acute lymphoblastic leukemia (ALL) [11]. A consensus about optimal therapy for lymphoblastic lymphoma is lacking, and outcomes have not changed significantly since the 1980s [11–13]. Most successful treatment strategies for advanced-stage pediatric lymphoblastic lymphoma are derived from regimens designed for children with high-risk ALL, consisting of intensive chemotherapy followed by maintenance therapy for a total duration of 2 years. Numerous drugs are given including corticosteroids, anthracyclines, cyclophosphamide, and MTX (Table 28.3). As with ALL, cranial radiation has been omitted over time [12–14], and CNS prophylaxis relies on HD-MTX and/or intrathecal injections [15, 16].

Treatment of Relapsed Disease

There is no standard treatment option for patients with recurrent or progressive NHL. Re-induction regimens use novel chemotherapy combinations such as ifosfamide, carboplatin, and etoposide (ICE) [17]. In most cases, myeloablative chemotherapy with either autologous or allogeneic HSCT may offer the best option for cure [18–22]. In T-cell malignancies, nelarabine has been shown to be effective as a single agent [23]. Depending on the presence of cell surface markers, monoclonal antibodies may be added to the regimen: the anti-CD20 antibody rituximab, anti-CD30 agent brentuximab vedotin [24–28], or bispecific antibodies (anti-CD20/anti-CD3 or CD19/CD3 T-cell engager blinatumomab) [6, 17, 18, 29–34]. Targeted therapies for ALK-positive ALCL include crizotinib, a kinase inhibitor that blocks the activity of the NPM-ALK fusion protein [24]. The long-term effects of these novel therapies in childhood cancer survivors are poorly characterized [24, 35–41].

Radiation Therapy

Based on several clinical trials, prophylactic cranial radiation has been omitted for pediatric patients with NHL. It has also been eliminated for patients with anaplastic large cell lymphoma and B-cell NHL who present with CNS disease [12, 13, 42, 43]. For patients with lymphoblastic lymphoma, low-dose radiation therapy (1800 cGy) is often used to treat overt CNS disease (e.g., cranial nerve palsies, intracerebral tumor extension, paraplegia). However, long-term survivors of NHL treated in prior eras might have been exposed to variable doses and fields of radiation, significantly increasing the risk for subsequent malignant neoplasms (SMNs), endocrine dysfunction, and neurocognitive difficulties [44].

Late Effects in Childhood NHL Survivors

Contemporary childhood NHL therapy differs from that used decades ago with regard to treatment intensity and recent omission of irradiation. Differences in treatment between histologic NHL subtypes have resulted in heterogeneous exposures in survivors. However, despite these variations in combination and cumulative exposure, the same chemotherapeutic agents continue to form the backbone of current treatment regimens. Several international cohorts have reported late outcomes in adult survivors of childhood NHL. A study of 362 adult survivors of childhood NHL treated between 1964 and 2002 at St. Jude Children's Research Hospital revealed that by a median age of 34 years (range 20–58), survivors had a significant

Table 28.1 Treatment protocols for mature Pediatric B-cell lymphoma

	Year (Ref)	Anthracyclines			Epipodophyllotoxins	Alkylating agents		CNS-directed therapy	Radiation	Rituximab
		Daunorubicin	Doxorubicin	MTX	Etoposide	Cyclophosphamide	Ifosfamide			
		mg/m^2	mg/m^2	g/m^2	g/m^2	g/m^2	g/m^2			
LMB 89	1989–1996 [124]	None	120–240	0–40	0–2.5	3–6.8	None	0–10 TIT	None	None
FAB LMB 96	1996–2001 [42, 84]	None	120–240	0–40	0–2.5	3–6.8	None	0–10 TIT	None	None
BFM 90	1990–1995 [125]	None	50–150	1–30	0.2–0.6	2–4	4–12	3–7 TIT	None	None
BFM 95	1996–2001 [126]	None	50–100	2–20	0.2–1.4	2–2.4	4–8	3–7 TIT	None	None
Inter B NHL Ritux 2010	Since 2011 [4]	None	120–180	15–32	0–1.6	3.3–5.8	None	10–12 TIT	None	2250 mg/m^2

Table 28.2 Treatment protocols for pediatric anaplastic large cell lymphoma

		Anthracyclines			Epipodophyllotoxins		Alkylating agents			
	Year (Ref)	Daunorubicin	Doxorubicin	MTX	Etoposide	Teniposide	Cyclophosphamide	Ifosfamide	CNS-directed therapy	Radiation
		mg/m^2	mg/m^2	g/m^2	g/m^2	g/m^2	g/m^2	g/m^2		
POG-9219	1983–1991 [127]	None	120	0.6 (PO)	None	None	2.25	None	MTX or TIT	Involved field radiation with a total dose of 27 Gy in 18 fractions during a period of 3 1/2 weeks (1983–1987)
BFM 86–90	1986–1995 [125]	None	50–150	1.5–3	600–1300	400	1.4–3.4	8–12	4–11	None
SFOP HM89–91	1989–1997 [128]	None	360	18	1200–1800	None	7.3–10.3	None	None	*Patients with CNS disease excluded*
UKCCSG 9001–9003	1990–1997 [129]	None	180–240	4–24	0–2500	None	5.8–6.8	None	10	None
POG-9315	1994–2000 [130]	None	300	0.9–8.9	None	None	None	None	MTX*6–11	None
ALCL99	1999–2006 [9]	None	150	6–18	0.6	None	3.4	12	TIT*7	None

Table 28.3 Treatment protocols for pediatric lymphoblastic lymphoma

		Anthracyclines			Epipodophyllotoxins		Alkylating agents			
	Year (Ref)	Daunorubicin mg/m^2	Doxorubicin mg/m^2	MTX g/m^2	Etoposide g/m^2	Teniposide g/m^2	Cyclophosphamide g/m^2	Ifosfamide g/m^2	CNS-directed therapy	Radiation
LSA2L2	1971–1990 [131]	240–300	None	None	None	None	15.6 gram (pre-1980) 8.4 gr (post 1980)	None	None	Bulky disease: 20–55 Gray before 1977 20 Gray 1977–1989
LMT-81	1981–1989 [132]	240–300	None	HD*10	None	None	8.4	None	None	Cranial if CNS involvement
POG T3	1986–1992 [133]	None	400	None	None	3.6	6.2	None	TIT*17	2400 cGy if WBC >50
NHL13	1992–2002 [13]	50	None	42	13.8	None	10.2	None	15–22 TIT	Eliminated
CCG 5941	1994–1997 [134]	60	180	7	2.8	None	8.4	None	*12	1800 cGy cranial and 600 cGy spinal at end of therapy if CNS positive at diagnosis
POG 9404	1996–2001 [15]	None	450	0–20	None	None	None	None	TIT11–13 or amended	1800 cGy cranial
COG A5971	2000–2005 [16]	120	None	0–20	None	None	3–5	None	13–23 MTX + AraC	1800 cGy (>2); 1200 cGy (1–2y)
AALL1231	Since 2014	100–130	75–150	1–11	0–0.5	None	0.3–3.2	0–4	20–28	Testicular radiation if positive at the end of induction (2400 cGy) IR or VHR and CNS 3: 1800 cGy at start of maintenance

burden of chronic health conditions [45]. Most notable was the high prevalence of cardiovascular risk factors common to the metabolic syndrome: overweight/obesity (65%), elevated fasting glucose (37%), high total cholesterol (35%), and hypertension (25%), even in individuals who were not exposed to cranial or abdominal radiation. Survivors exposed to cardiotoxic therapies were at risk for cardiomyopathy (19/164 exposed), and survivors treated with higher doses of alkylating agents and/or gonadal radiation were at risk for gonadal dysfunction, including oligospermia/azoospermia among male survivors. Survivors who received neck or thyroid radiation were at risk for primary hypothyroidism. The risk for SMN was sixfold that in the general population, with all SMNs occurring in previously radiated survivors. Among participants who underwent comprehensive neurocognitive assessments (n = 171), 68% experienced at least mild impairment in executive function, attention, and/or memory. In another study of the same cohort, slower processing speed and poorer self-reported executive function were associated with symptoms of depression [46]. Survivors with neurocognitive impairment were at risk for lower educational attainment, unemployment, and reduced occupational status. A study of 103 young survivors of high-grade NHL treated between 1973 and 1993 as per treatment protocols of the Pediatric Oncology Branch of the National Cancer Institute revealed cardiotoxicity in 26% of patients who had received doxorubicin and SMN in 2% [47]. Endocrine impairments were detected in 16% of all evaluated patients with hypopituitarism, thyroid dysfunction, and hypogonadism reported as the most common abnormalities.

An assessment of cause-specific mortality and SMN incidence among 1082 survivors of NHL in the North American Childhood Cancer Survivor Study who were treated between 1970 and 1986 observed 87 late deaths that occurred at a median age of 24 years and a median of 13 years after NHL diagnosis. This was approximately fourfold that expected in the general population. Causes of death included disease recurrence, secondary solid tumors and leukemia, cardiac disease, and pneumonia. The risk for death remained elevated beyond 20 years after NHL, with female sex and chest irradiation noted as significant risk factors. Increased risk of death in survivors of childhood NHL has also been reported in several international cohorts [48–50]. However, a statistically significant decline in excess mortality was observed in the British Childhood Cancer Survivor Study among those with a diagnosis of childhood NHL between 1990 and 2006 compared with survivors with a diagnosis before 1970 [51], confirming other reports that suggest that changes in therapy over time have decreased the risk for late mortality [52, 53].

Specific Late Effects in Survivors of Childhood Non-Hodgkin Lymphoma

Cardiotoxicity

Childhood cancer survivors are at higher risk than the general population for the development of cardiovascular and cerebrovascular disease [54, 55]. Children treated with cardiotoxic therapies such as anthracycline chemotherapy and radiation to a field that includes the heart are at elevated risk for cardiomyopathy [56] and coronary artery disease [57]. Anthracyclines, such as daunorubicin, doxorubicin, epirubicin, and idarubicin, have been incorporated in treatment protocols for NHL for decades. These agents impact cardiac function in a dose-dependent manner, with patients treated with >250–300 mg/m^2 of doxorubicin-equivalent anthracycline therapy at particular risk for cardiomyopathy. Younger age at treatment, particularly therapy as an infant or toddler, increases this risk [56]. While therapeutic radiation to the chest and mediastinum inevitably involves dose to the heart, radiation incorporating the spine (including total body irradiation (TBI) in the setting of hematopoietic stem cell transplantation), left flank or left upper quadrant may also cause significant radiation exposure to cardiac tissue. Radiation contributes to the development of cardiomyopathy, coronary artery disease, valvular heart disease, pericardial disease, and arrhythmias. With changes in NHL protocols over time, there has been a trend toward the use of lower anthracycline doses with a goal of reducing cardiovascular risk. Advancements in radiation planning techniques have also reduced the cardiac radiation dose in patients with thoracic disease [58].

However, cardiovascular disease remains an important cause of late mortality in NHL survivors. Death due to circulatory causes in NHL patients is 4.2–7.3 times higher than that of the general population. Although subsequent malignant neoplasms (SMNs) are the leading cause of late death in childhood cancer survivors overall, as these survivors pass 60 years of age, death from cardiovascular diseases overtakes deaths from new cancers [51]. Studies of survivors of childhood lymphoma have demonstrated higher proportions of body fat, indicating reduced lean body mass in survivors compared with healthy controls [59]. Survivors have also been shown to be at elevated risk for hypertension, dyslipidemia, and overweight, all of which may contribute to cardiovascular risk; smoking may further exacerbate this risk [60]. These modifiable cardiovascular risk factors provide targets for preventive interventions in survivors who have been exposed to cardiotoxic therapies.

When evaluating cardiovascular risk in a survivor of childhood NHL, it is important to document both the cumulative anthracycline dose and any cardiac radiation exposure. Annual clinical evaluation should include a comprehensive

history and physical assessment with documentation and evaluation of symptoms of dyspnea (on exertion and/or at rest), orthopnea, chest pain, and palpitations. Physical examination should incorporate a comprehensive cardiac evaluation including evaluation of blood pressure, auscultation of heart sounds, and monitoring for early signs of cardiac dysfunction. It is imperative that physicians be alert to symptoms of chest pain and/or exertional intolerance in younger patients who have had exposure to anthracyclines and/or radiation. It is also important to recognize that atypical presentations such as complaints of abdominal pain in a young adult may be cardiac in nature. Such symptoms warrant comprehensive cardiac evaluation with a low threshold to proceed to cardiology referral in those with a history of anthracycline or radiation exposure [61]. General health measures for all childhood NHL survivors should include adopting a healthy diet, participation in regular exercise, and avoidance of smoking and drugs that can affect cardiac function (e.g., cocaine, diet pills, etc.). Extreme isometric exercise (e.g., heavy weight lifting) should be avoided. Long-term follow-up guidelines published by the Children's Oncology Group (available at www.survivorshipguidelines.org) suggest that survivors exposed to an anthracycline or cardiac radiation have a baseline echocardiogram at entry to long-term follow-up and then a repeat echocardiogram every 1–5 years depending on age at treatment, radiation exposure, and cumulative anthracycline dose. Regular surveillance for cardiac disease and lifestyle changes to mitigate cardiovascular risk may reduce the incidence of cardiovascular complications and improve outcomes in survivors of NHL exposed to anthracyclines and/or chest-directed radiation therapy.

Subsequent Malignant Neoplasms (SMNs)

Childhood cancer survivors are at substantial risk of developing an SMN [51, 54, 62–64]. Among all survivors, the cumulative incidence of developing an SMN by 30 years from their primary childhood cancer diagnosis is 7–10% [48, 62, 65, 66]. Mortality rates due to SMN exceed that of all other causes including primary disease recurrence [67]. Beyond age 35 years, survivors of childhood malignancy experience a marked and disproportionate increase in cumulative incidence of malignant neoplasms compared with siblings [64]. SMN subsequent to treatment for NHL tend to occur earlier than those subsequent to treatment for solid tumors [68]. Many studies have documented the types of SMNs typically seen following treatment for NHL. These include secondary myelodysplastic syndromes (MDS)/acute myeloid leukemias (AML), primarily caused by epipodophyllotoxins and alkylating agents, and an increased risk of solid tumors, particularly thyroid cancer, primarily caused by radiation exposure. Cumulative rates of SMN at 30 years from NHL diagnosis vary from 2.5% to 8% in international cohorts [48, 62, 69]. A study of 1150 survivors of childhood NHL diagnosed in the United States between 1973 and 2002 noted a ratio of observed to expected cancers of 5.3 and a notable increase in instances of leukemia and breast cancer following NHL treatment [70]. An international study of 2563 survivors of childhood Non-Hodgkin lymphoma observed a cumulative incidence of SMN of 2.5% within 30 years of treatment, with a particularly elevated risk of thyroid cancer and brain tumors [69]. More recent studies, including reports from the Childhood Cancer Survivor Study, have shown non-melanoma skin cancer, thyroid cancer, and leukemias to be the most prevalent SMN [62].

Risk factors for SMN include treatment exposures and host genetic factors. These include treatment with anthracyclines, epipodophyllotoxins, or alkylating agents, exposure to therapeutic radiation, and/or hematopoietic stem cell transplantation [71]. Chemotherapy-related SMNs include acute leukemias (typically MDS/AML), likely secondary to therapy with topoisomerase inhibitors and alkylating agents, which usually occur in the first 10 years after cancer treatment, and solid tumors which typically present 10–20 years after cancer treatment. Thyroid and breast cancers are two more common SMN observed in the setting of radiation to the neck or chest. Advances in radiation techniques and improved radiation planning strategies have reduced radiation dose to healthy tissues in some clinical situations compared with mantle-type radiation historically employed for mediastinal disease [58]. Genetic assessment should include documentation of a family history of malignancy (e.g., lymphoma affecting other family members, notable family history of other malignancies), eliciting clinical clues to the existence of an underlying cancer predisposition syndrome (e.g., presence of an underlying immunodeficiency, dysmorphic features, or congenital anomalies), and the presence of consanguinity [72, 73]. SMN risk after treatment with some of the newer agents employed in the treatment of NHL is less well established and will require long-term follow-up of NHL cohorts over time. Emerging data related to rituximab use in survivors of diffuse large B-cell lymphoma and primary mediastinal B-cell lymphoma suggests increased risks of AML, thyroid cancer, and melanoma as well as other cancers [39], although with some conflicting reports [38, 40, 41]. For survivors with a history of resistant, refractory or relapsed disease, long-term effects of therapy and SMN risk related to hematopoietic stem cell transplantation (especially secondary MDS and AML) [74, 75] are a significant concern, although there is no evidence for the use of routine complete blood counts to screen for MDS/AML. The Children's Oncology Group guidelines suggest that survivors exposed to radiation warrant particular attention to SMN surveillance in irradiated fields. This includes annual

dermatological evaluation to assess for skin cancers, annual thyroid exam examination, and screening with mammography/breast MRI and colonoscopy where appropriate.

Gonadotoxicity and Fertility

At the time of diagnosis of a life-threatening malignancy, the long-term effects of therapy on the reproductive potential of young patients are often far from the minds of those facing treatment and their family members. However, the ability to have children is a major concern among adult survivors of childhood cancer. Infertility in NHL survivors is closely linked to the cumulative dose of specific chemotherapy agents (particularly alkylating agents), exposure to abdominal irradiation (in males and females), and/or direct or indirect radiation to the testes [76]. Cranial radiation (with dose to the hypothalamus/pituitary axis) and total body irradiation (in the setting of hematopoietic stem cell transplantation) have also been implicated. Although any dose of alkylating agent can impact gonadal function, a cyclophosphamide-equivalent dose in excess of 6–8 g/m^2 is generally considered the threshold above which the risk rises [77]. Rates of gonadotoxicity are lower in survivors of NHL compared to survivors of Hodgkin lymphoma (HL), as significantly more patients with HL require radiation or treatment with gonadotoxic drugs such as procarbazine [78, 79]. Some pubertal males have reduced sperm counts at presentation of their NHL or will develop them during or after chemotherapy—many will gradually recover their sperm counts to achieve normospermic levels [80]. However, pelvic radiotherapy and higher alkylating agent exposures are associated with permanent reductions in sperm count. A study of 757 patients of all ages treated for aggressive NHL documented a cumulative incidence of infertility of 29% in females and 18% in males at 15 years from treatment, while another study reported azoospermia in 11% of male survivors treated for childhood NHL [81]. Risk factors included autologous stem cell transplant, abdominal radiation, alkylating agents, and need for salvage treatments in the setting of recurrent/relapsed disease. Kiserud and colleagues reported on 129 male patients with NHL, of whom 50% had abnormal gonadal hormone levels. Of these 129 patients, 25% had exocrine hypogonadism with high follicle stimulating hormone (FSH) and normal testosterone and luteinizing hormone (LH). Endocrine hypogonadism with low testosterone and/or elevated LH was present in 24% of all NHL survivors. Fifty percent of the patients with endocrine hypogonadism had undergone autologous stem cell rescue after conditioning with either high-dose chemotherapy with BEAM (carmustine, etoposide, cytarabine, and melphalan) or total body irradiation plus cyclophosphamide [82].

Evolution of NHL treatment protocols over time has resulted in a reduction in the cumulative doses of alkylating agents such as cyclophosphamide and the elimination of CCNU, reducing the risk of gonadotoxicity [83, 84]. Additionally, advancements in fertility preservation techniques offer many patients the opportunity to preserve fertility, an option that was not available to historic cohorts. This includes sperm banking in male patients and oocyte retrieval and cryopreservation in females. For prepubertal patients, emerging technologies such as testicular tissue cryopreservation in young male patients and ovarian tissue cryopreservation in females, though still at an experimental stage, offer hope for reproductive potential in the future [85, 86]. Such techniques, however, are only available in limited number of institutions. For health professionals involved in survivor care, there is an increasing need for appropriate education to facilitate provision of fertility and sexual health assessments and counseling at clinic visits. Survivors may have concerns that extend beyond their likelihood of conceiving children, such as fear of rejection in the setting of infertility, hesitancy to share infertility concerns with partners, and fear of transmitting cancer to their offspring [87]. Access to input from services such as psychology, endocrinology, urology, and gynecology is important to optimize care in this critical area of survivor care.

Schooling and Education

School is an important component of the normal childhood and adolescence experience, not just from an educational perspective but also for various aspects of psychosocial, emotional, and personal development. Across institutions and cancer diagnoses, physicians encourage children, when feeling well, to attend school during cancer therapy. Despite this, many children miss significant portions of their school terms as a consequence of their disease, its treatment, and complications of therapy. Consequently, children may require additional supports when they transition back to full-time education. Some children experience significant anxiety and have difficulties readjusting to school life, while others experience psychosocial and emotional difficulties upon reintegration with peers. Children who have received CNS-directed chemotherapy and/or cranial irradiation as part of their treatment are at particular risk [88]; they can develop deficits in attention and concentration as well as in higher-order functions such as memory and information processing, with resultant impacts on learning ability [89–91]. Other risk factors for poorer outcomes include younger age at diagnosis and female sex [92]. In a study of survivors enrolled in the Childhood Cancer Survivor Study [93], survivors of NHL ($n = 908$) were significantly less likely to finish high school compared with siblings (odds ratio [OR] 1.8).

Recently, Ehrhardt and colleagues described outcomes in adult survivors of childhood NHL [46]. Although they noted normal intelligence and attention in survivors, memory, executive function, processing speed, and academics were impaired compared with the general population. Over two thirds of survivors experienced at least mildly impaired executive function, attention and/or memory, and neurocognitive impairment was associated with lower educational attainment and lower HRQoL. The impact of lower educational attainment on HRQoL has been noted in other studies, particularly when survivors have unmet school needs or limited access to needed support services [94–96]. Optimization of neurocognitive, educational, and psychosocial outcomes must be a focus in long-term follow-up clinics. Despite the elimination of cranial irradiation from most NHL treatment regimens, survivors may remain at risk for neurocognitive impairment due to CNS-directed chemotherapies that continue to be incorporated in treatment protocols. Survivorship programs must engage multidisciplinary teams and school services, incorporating open communication between schools and medical teams, as well as the implementation of neurocognitive interventions and school re-entry programs, with accommodations for exams and assignments where appropriate [89, 96, 97].

Quality of Life and Psychosocial Outcomes

The care of survivors of NHL must extend beyond surveillance and intervention for physical late effects. Unfortunately, some survivors are at risk for psychological sequelae of their cancer therapy. Patients treated during adolescence appear to be particularly vulnerable to having increased mental health needs, even years after the conclusion of their cancer therapy [98]. The most frequent mental health problems in survivors are post-traumatic stress disorder (PTSD), as well as symptoms of anxiety and depression [99–102]. Interestingly, some survivors report positive psychological outcomes as a consequence of surviving cancer, a phenomenon termed post-traumatic growth [103]. Adverse physical, neurocognitive, and psychosocial outcomes can reduce HRQoL [46, 90, 94, 104–106] with adolescent age at time of cancer diagnosis associated with poorer HRQoL [96]. Among NHL survivors, more symptoms of depression, reduced HRQoL, and lower social attainment have been reported in those with evidence of neurocognitive impairment, when compared with normative data. Survivors also report elevated rates of chronic pain, poorer social functioning, and inferior general and mental health, with strong associations between neurocognitive challenges and emotional distress [46]. In one study, lymphoma survivors were noted to have higher rates of psychological distress than siblings (OR 1.8 for anxiety and OR 1.8 for somatization) [94]. Survivors with medical complications related to therapy are at higher risk of poor HRQoL outcomes [107], as are those with chronic pain [108] or poor sleep quality [109]. Appropriate long-term care of NHL survivors must include periodic assessment for psychological challenges to ensure that the prior cancer or current late effects are not negatively impacting the survivor. Providers must consider issues unique to this cohort such as body appearance, fertility/sexual functioning, peer and romantic relationships, educational and occupational attainment, and fear of recurrence [87, 110, 111]. Peer support groups may be helpful for selected survivors and families [112]. Lack of access to mental health and allied health services such as physical/occupational health therapies have been reported to be strongly associated with inferior outcomes and poorer functioning [95]. Finally, a holistic approach to psychosocial needs and HRQoL in survivor care that encompasses attention to siblings and families is warranted, not just during the acute diagnostic period but, rather, throughout the survivorship journey [112, 113].

Care of Survivors

Risk-Based Care and Surveillance

In recognition of the growing population of survivors of both childhood and adult cancers, the US Institute of Medicine (IOM) has published recommendations for the care of survivors [114, 115]. Among these recommendations are the following:

1. Develop evidence-based clinical practice guidelines for the care of survivors of childhood cancer

As noted above, the North American Children's Oncology Group (COG) has published guidelines for the lifelong care of survivors of childhood, adolescent, and young adult cancers (available at www.survivorshipguidelines.org). These guidelines provide healthcare providers with recommendations for follow-up based on each survivor's specific treatment exposures, and not on the prior cancer. Thus, in order to determine the recommendations for an individual survivor, their healthcare provider needs to know the therapies that they received for their NHL. Several other international groups have also published care guidelines. In an effort to harmonize these multiple and sometime conflicting recommendations, the International Guideline Harmonization Group for Late Effects of Childhood Cancer (www.ighg.org) was formed. This multinational, multidisciplinary group has adopted a standardized methodology for systematically reviewing the literature to formulate consensus recommendations for survivor care. To date, the IGHG has published recommendations for surveillance for secondary breast [116]

and thyroid cancers [117], cardiomyopathy [118], premature ovarian insufficiency [119], and male gonadotoxicity [120].

2. Define a minimum set of standards for systems of comprehensive, multidisciplinary follow-up care that link specialty and primary care providers

During their childhood years, most cancer survivors receive care in the pediatric center that provided their NHL therapy. However, once they "age out" of the pediatric cancer center, the location and provider of their long-term care is highly variable. Only a minority of centers are able to offer specialized survivor care to adult survivors of childhood cancer. These specialized survivor clinics frequently have multidisciplinary teams that can include oncologists, nurses, psychologists, social workers, nutritionists, rehabilitation therapists, and vocational counselors. However, many survivors are transferred back to their primary care physician when they reach adulthood. Unfortunately, primary care physicians are often uncomfortable caring for childhood cancer survivors, have limited knowledge about follow-up guidelines [121, 122], and are unlikely to provide recommended surveillance [123]. Therefore, it is crucial that survivors are able to contact the oncology team that treated them or have access to a specialized survivorship program once they leave the pediatric cancer center. Where specialized programs do not exist, the survivor should be provided with a treatment summary and care plan (described below) that they can share with any new medical care provider.

3. Improve awareness of late effects and their implications for long-term health among childhood cancer survivors and their families

Children are often treated for NHL at a young age and so, frequently, are not aware of the complete details of their cancer diagnosis and treatment. It is critical that they receive this education prior to transition from pediatric care so that they are sufficiently informed to be able to advocate for their own healthcare, including ensuring that they receive appropriate surveillance for late effects. A treatment summary and care plan are key tools for patient empowerment. In fact, the American College of Surgeons Commission on Cancer has made provision of a care summary and follow-up plan to all cancer survivors, a condition of accreditation as a cancer program in the United States. Key elements in these documents include:

1. Information on the primary cancer (e.g., NHL type, stage, treatment protocol, key dates)
2. Chemotherapy received, including cumulative doses of specific classes of chemotherapy agents (e.g., anthracyclines, alkylating agents)
3. Radiation field and dose
4. Surgeries
5. Stem cell transplantation
6. Recommendations for follow-up (e.g., second cancer surveillance, cardiac surveillance)

If the treating center does not have a template for creating such a plan, templates are available from several groups including the American Society of Clinical Oncology and LIVESTRONG.

Conclusions

As the long-term survival of childhood NHL has improved, clinicians have focused on reducing exposures to toxic chemotherapies and radiation. However, the long-term impact of NHL therapies will never be completely eliminated, and there are many survivors of NHL who were treated intensively in prior eras. Thus, all survivors require lifelong healthcare that is adapted to their particular physical, psychological, and social risks, with the goal of maximizing both the quantity and quality of their lives.

MTX Methotrexate, *CNS* Central nervous system, *IT* Intrathecal therapy, TIT Triple intrathecal therapy

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MTX Methotrexate, *CNS* Central nervous system, *IT* Intrathecal therapy, *TIT* Triple intrathecal therapy, *IR* Intermediate, *VHR* Very high risk

References

1. Howlader N, et al. SEER cancer statistics review, 1975-2014. Bethesda: National Cancer Institute; 2017.
2. Phillips SM, et al. Survivors of childhood cancer in the United States: prevalence and burden of morbidity. Cancer Epidemiol Biomark Prev. 2015;24(4):653–63.
3. Nathan PC, et al. Financial hardship and the economic effect of childhood cancer survivorship. J Clin Oncol. 2018;36(21):2198–205.
4. Goldman S, et al. Rituximab and FAB/LMB 96 chemotherapy in children with Stage III/IV B-cell non-Hodgkin lymphoma: a Children's Oncology Group report. Leukemia. 2013;27(5):1174–7.
5. Meinhardt A, et al. Phase II window study on rituximab in newly diagnosed pediatric mature B-cell non-Hodgkin's lymphoma and Burkitt leukemia. J Clin Oncol. 2010;28(19):3115–21.
6. Minard-Colin V, et al. Non-Hodgkin lymphoma in children and adolescents: progress through effective collaboration, current knowledge, and challenges ahead. J Clin Oncol. 2015;33(27):2963–74.
7. Attarbaschi A, et al. Limited stage I disease is not necessarily indicative of an excellent prognosis in childhood anaplastic large cell lymphoma. Blood. 2011;117(21):5616–9.
8. Wrobel G, et al. Safety assessment of intensive induction therapy in childhood anaplastic large cell lymphoma: report of the ALCL99 randomised trial. Pediatr Blood Cancer. 2011;56(7):1071–7.

9. Brugières L, et al. Impact of the methotrexate administration dose on the need for intrathecal treatment in children and adolescents with anaplastic large-cell lymphoma: results of a randomized trial of the EICNHL group. J Clin Oncol. 2009;27(6):897–903.
10. Williams D, et al. Central nervous system involvement in anaplastic large cell lymphoma in childhood: results from a multicentre European and Japanese study. Pediatr Blood Cancer. 2013;60(10):E118–21.
11. Swerdlow SH, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127(20):2375–90.
12. Burkhardt B, et al. Impact of cranial radiotherapy on central nervous system prophylaxis in children and adolescents with central nervous system-negative stage III or IV lymphoblastic lymphoma. J Clin Oncol. 2006;24(3):491–9.
13. Sandlund JT, et al. Effective treatment of advanced-stage childhood lymphoblastic lymphoma without prophylactic cranial irradiation: results of St Jude NHL13 study. Leukemia. 2009;23(6):1127–30.
14. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2017. CA Cancer J Clin. 2017;67(1):7–30.
15. Asselin BL, et al. Effectiveness of high-dose methotrexate in T-cell lymphoblastic leukemia and advanced-stage lymphoblastic lymphoma: a randomized study by the Children's Oncology Group (POG 9404). Blood. 2011;118(4):874–83.
16. Termuhlen AM, et al. Disseminated lymphoblastic lymphoma in children and adolescents: results of the COG A5971 trial: a report from the Children's Oncology Group. Br J Haematol. 2013;162(6):792–801.
17. Griffin TC, et al. A study of rituximab and ifosfamide, carboplatin, and etoposide chemotherapy in children with recurrent/refractory B-cell (CD20+) non-Hodgkin lymphoma and mature B-cell acute lymphoblastic leukemia: a report from the Children's Oncology Group. Pediatr Blood Cancer. 2009;52(2):177–81.
18. Jetsrisuparb A, et al. Rituximab combined with CHOP for successful treatment of aggressive recurrent, pediatric B-cell large cell non-Hodgkin's lymphoma. J Pediatr Hematol Oncol. 2005;27(4):223–6.
19. Kewalramani T, et al. Rituximab and ICE as second-line therapy before autologous stem cell transplantation for relapsed or primary refractory diffuse large B-cell lymphoma. Blood. 2004;103(10):3684–8.
20. Schuster FR, et al. Immunotherapy with the trifunctional anti-CD20 x anti-CD3 antibody FBTA05 (Lymphomun) in paediatric high-risk patients with recurrent CD20-positive B cell malignancies. Br J Haematol. 2015;169(1):90–102.
21. Gross TG, et al. Hematopoietic stem cell transplantation for refractory or recurrent non-Hodgkin lymphoma in children and adolescents. Biol Blood Marrow Transplant. 2010;16(2):223–30.
22. Satwani P, et al. Sequential myeloablative autologous stem cell transplantation and reduced intensity allogeneic hematopoietic cell transplantation is safe and feasible in children, adolescents and young adults with poor-risk refractory or recurrent Hodgkin and non-Hodgkin lymphoma. Leukemia. 2015;29(2):448–55.
23. Berg SL, et al. Phase II study of nelarabine (compound 506U78) in children and young adults with refractory T-cell malignancies: a report from the Children's Oncology Group. J Clin Oncol. 2005;23(15):3376–82.
24. Prokoph N, et al. Treatment options for paediatric anaplastic large cell lymphoma (ALCL): current standard and beyond. Cancers (Basel). 2018;10(4):E99.
25. Broccoli A, et al. Italian real-life experience with brentuximab vedotin: results of a large observational study of 40 cases of relapsed/refractory systemic anaplastic large cell lymphoma. Haematologica. 2017;102(11):1931–5.
26. Gandolfi L, et al. Long-term responders after brentuximab vedotin: single-center experience on relapsed and refractory Hodgkin lymphoma and anaplastic large cell lymphoma patients. Oncologist. 2016;21(12):1436–41.
27. Pro B, et al. Five-year results of brentuximab vedotin in patients with relapsed or refractory systemic anaplastic large cell lymphoma. Blood. 2017;130(25):2709–17.
28. Sekimizu M, et al. Phase I clinical study of brentuximab vedotin (SGN-35) involving children with recurrent or refractory CD30-positive Hodgkin's lymphoma or systemic anaplastic large cell lymphoma: rationale, design and methods of BV-HLALCL study: study protocol. BMC Cancer. 2018;18(1):122.
29. Goebeler ME, et al. Bispecific T-cell engager (BiTE) antibody construct blinatumomab for the treatment of patients with relapsed/refractory non-Hodgkin lymphoma: final results from a phase I study. J Clin Oncol. 2016;34(10):1104–11.
30. Viardot A, et al. Phase 2 study of the bispecific T-cell engager (BiTE) antibody blinatumomab in relapsed/refractory diffuse large B-cell lymphoma. Blood. 2016;127(11):1410–6.
31. Alasaad T, Barr R. Successful treatment of multiply relapsed lymphoma with rituximab as a single agent. Pediatr Blood Cancer. 2010;55(2):356–8.
32. Anoop P, et al. Outcome of childhood relapsed or refractory mature B-cell non-Hodgkin lymphoma and acute lymphoblastic leukemia. Leuk Lymphoma. 2012;53(10):1882–8.
33. Budde LE, et al. A phase I study of pulse high-dose vorinostat (V) plus rituximab (R), ifosfamide, carboplatin, and etoposide (ICE) in patients with relapsed lymphoma. Br J Haematol. 2013;161(2):183–91.
34. Cortelazzo S, et al. Randomized trial comparing R-CHOP versus high-dose sequential chemotherapy in high-risk patients with diffuse large B-cell lymphomas. J Clin Oncol. 2016;34(33):4015–22.
35. McNeer JL, Bleyer A. Acute lymphoblastic leukemia and lymphoblastic lymphoma in adolescents and young adults. Pediatr Blood Cancer. 2018;65(6):e26989.
36. Fridrik MA, et al. Cardiotoxicity with rituximab, cyclophosphamide, non-pegylated liposomal doxorubicin, vincristine and prednisolone compared to rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisolone in frontline treatment of patients with diffuse large B-cell lymphoma: a randomised phase-III study from the Austrian Cancer Drug Therapy Working Group [Arbeitsgemeinschaft Medikamentose Tumortherapie AGMT] (NHL-14). Eur J Cancer. 2016;58:112–21.
37. Tao L, et al. Subsequent primary malignancies after diffuse large B-cell lymphoma in the modern treatment era. Br J Haematol. 2017;178(1):72–80.
38. Fleury I, et al. Rituximab and risk of second primary malignancies in patients with non-Hodgkin lymphoma: a systematic review and meta-analysis. Ann Oncol. 2016;27(3):390–7.
39. De Sanctis V, et al. Second cancer incidence in primary mediastinal B-cell lymphoma treated with methotrexate with leucovorin rescue, doxorubicin, cyclophosphamide, vincristine, prednisone, and bleomycin regimen with or without rituximab and mediastinal radiotherapy: results from a monoinstitutional cohort analysis of long-term survivors. Hematol Oncol. 2017;35(4):554–60.
40. Cho SF, et al. Risk of second primary cancer in patients with B-cell non-Hodgkin lymphoma receiving rituximab-containing chemotherapy: a nationwide population-based study. Anticancer Res. 2015;35(3):1809–14.
41. Pavanello F, Zucca E, Ghielmini M. Rituximab: 13 open questions after 20 years of clinical use. Cancer Treat Rev. 2017;53:38–46.
42. Cairo MS, et al. Results of a randomized international study of high-risk central nervous system B non-Hodgkin lymphoma and B acute lymphoblastic leukemia in children and adolescents. Blood. 2007;109(7):2736–43.
43. Seidemann K, et al. Short-pulse B-non-Hodgkin lymphoma-type chemotherapy is efficacious treatment for pediatric anaplastic

large cell lymphoma: a report of the Berlin-Frankfurt-Münster Group trial NHL-BFM 90. Blood. 2001;97(12):3699–706.
44. Bluhm EC, et al. Cause-specific mortality and second cancer incidence after non-Hodgkin lymphoma: a report from the Childhood Cancer Survivor Study. Blood. 2008;111(8):4014–21.
45. Ehrhardt MJ, et al. Late outcomes of adult survivors of childhood non-Hodgkin lymphoma: a report from the St. Jude Lifetime Cohort Study. Pediatr Blood Cancer. 2017;64(6):e26338.
46. Ehrhardt MJ, et al. Neurocognitive, psychosocial, and quality-of-life outcomes in adult survivors of childhood non-Hodgkin lymphoma. Cancer. 2018;124(2):417–25.
47. Haddy TB, et al. Late effects in long-term survivors of high-grade non-Hodgkin's lymphomas. J Clin Oncol. 1998;16(6):2070–9.
48. Inskip PD, Curtis RE. New malignancies following childhood cancer in the United States, 1973-2002. Int J Cancer. 2007;121(10):2233–40.
49. Leung W, et al. Second malignancy after treatment of childhood non-Hodgkin lymphoma. Cancer. 2001;92(7):1959–66.
50. Reulen RC, et al. Long-term cause-specific mortality among survivors of childhood cancer. JAMA. 2010;304(2):172–9.
51. Fidler MM, et al. Long term cause specific mortality among 34 489 five year survivors of childhood cancer in Great Britain: population based cohort study. BMJ. 2016;354:i4351.
52. Möller TR, et al. Decreasing late mortality among five-year survivors of cancer in childhood and adolescence: a population-based study in the Nordic countries. J Clin Oncol. 2001;19(13):3173–81.
53. Armstrong GT, et al. Late mortality among 5-year survivors of childhood cancer: a summary from the Childhood Cancer Survivor Study. J Clin Oncol. 2009;27(14):2328–38.
54. Bhakta N, et al. The cumulative burden of surviving childhood cancer: an initial report from the St Jude Lifetime Cohort Study (SJLIFE). Lancet. 2017;390(10112):2569–82.
55. Geenen MM, et al. Medical assessment of adverse health outcomes in long-term survivors of childhood cancer. JAMA. 2007;297(24):2705–15.
56. Chow EJ, et al. Individual prediction of heart failure among childhood cancer survivors. J Clin Oncol. 2015;33(5):394–402.
57. Chow EJ, et al. Prediction of ischemic heart disease and stroke in survivors of childhood cancer. J Clin Oncol. 2018;36(1):44–52.
58. Hoppe RT. Evolution of the techniques of radiation therapy in the management of lymphoma. Int J Clin Oncol. 2013;18(3):359–63.
59. Nysom K, et al. Degree of fatness after treatment of malignant lymphoma in childhood. Med Pediatr Oncol. 2003;40(4):239–43.
60. Moser EC, et al. Late non-neoplastic events in patients with aggressive non-Hodgkin's lymphoma in four randomized European Organisation for Research and Treatment of Cancer trials. Clin Lymphoma Myeloma. 2005;6(2):122–30.
61. Patel DA, et al. Clinical manifestations of noncoronary atherosclerotic vascular disease after moderate dose irradiation. Cancer. 2006;106(3):718–25.
62. Friedman DL, et al. Subsequent neoplasms in 5-year survivors of childhood cancer: the Childhood Cancer Survivor Study. J Natl Cancer Inst. 2010;102(14):1083–95.
63. Meadows AT, et al. Second malignant neoplasms following childhood Hodgkin's disease: treatment and splenectomy as risk factors. Med Pediatr Oncol. 1989;17(6):477–84.
64. Armstrong GT, et al. Aging and risk of severe, disabling, life-threatening, and fatal events in the childhood cancer survivor study. J Clin Oncol. 2014;32(12):1218–27.
65. Teepen JC, et al. Long-term risk of subsequent malignant neoplasms after treatment of childhood cancer in the DCOG LATER study cohort: role of chemotherapy. J Clin Oncol. 2017;35(20):2288–98.
66. Reulen RC, et al. Long-term risks of subsequent primary neoplasms among survivors of childhood cancer. JAMA. 2011;305(22):2311–9.
67. Mertens AC, et al. Late mortality experience in five-year survivors of childhood and adolescent cancer: the Childhood Cancer Survivor Study. J Clin Oncol. 2001;19(13):3163–72.
68. Pole JD, et al. Subsequent malignant neoplasms in a population-based cohort of pediatric cancer patients: a focus on the first 5 years. Cancer Epidemiol Biomark Prev. 2015;24(10):1585–92.
69. Maule M, et al. Risk of second malignant neoplasms after childhood leukemia and lymphoma: an international study. J Natl Cancer Inst. 2007;99(10):790–800.
70. Inskip PD, et al. Radiation-related new primary solid cancers in the childhood cancer survivor study: comparative radiation dose response and modification of treatment effects. Int J Radiat Oncol Biol Phys. 2016;94(4):800–7.
71. Tward J, et al. Incidence, risk factors, and pathogenesis of second malignancies in patients with non-Hodgkin lymphoma. Leuk Lymphoma. 2007;48(8):1482–95.
72. Cerhan JR, Slager SL. Familial predisposition and genetic risk factors for lymphoma. Blood. 2015;126(20):2265–73.
73. Jongmans MC, et al. Recognition of genetic predisposition in pediatric cancer patients: an easy-to-use selection tool. Eur J Med Genet. 2016;59(3):116–25.
74. Howe R, et al. Secondary myelodysplastic syndrome and acute myelogenous leukemia are significant complications following autologous stem cell transplantation for lymphoma. Bone Marrow Transplant. 2003;32(3):317–24.
75. Ruiz-Soto R, et al. Estimating late adverse events using competing risks after autologous stem-cell transplantation in aggressive non-Hodgkin lymphoma patients. Cancer. 2005;104(12):2735–42.
76. Steffens M, et al. Endocrine and metabolic disorders in young adult survivors of childhood acute lymphoblastic leukaemia (ALL) or non-Hodgkin lymphoma (NHL). Clin Endocrinol. 2008;69(5):819–27.
77. Green DM, et al. The cyclophosphamide equivalent dose as an approach for quantifying alkylating agent exposure: a report from the Childhood Cancer Survivor Study. Pediatr Blood Cancer. 2014;61(1):53–67.
78. Ben Arush MW, et al. Male gonadal function in survivors of childhood Hodgkin and non-Hodgkin lymphoma. Pediatr Hematol Oncol. 2000;17(3):239–45.
79. Bokemeyer C, et al. Long-term gonadal toxicity after therapy for Hodgkin's and non-Hodgkin's lymphoma. Ann Hematol. 1994;68:105.
80. Pryzant RM, et al. Long-term reduction in sperm count after chemotherapy with and without radiation therapy for non-Hodgkin's lymphomas. J Clin Oncol. 1993;11(2):239–47.
81. Romerius P, et al. High risk of azoospermia in men treated for childhood cancer. Int J Androl. 2011;34(1):69–76.
82. Kiserud CE, et al. Gonadal function in male patients after treatment for malignant lymphomas, with emphasis on chemotherapy. Br J Cancer. 2009;100(3):455–63.
83. Servitzoglou M, et al. Dose-effect relationship of alkylating agents on testicular function in male survivors of childhood lymphoma. Pediatr Hematol Oncol. 2015;32(8):613–23.
84. Patte C, et al. Results of the randomized international FAB/LMB96 trial for intermediate risk B-cell non-Hodgkin lymphoma in children and adolescents: it is possible to reduce treatment for the early responding patients. Blood. 2007;109(7):2773–80.
85. Burns KC, et al. Fertility preservation options in pediatric and adolescent patients with cancer. Cancer. 2018;124(9):1867–76.
86. Gertosio C, et al. Fertility preservation in pediatric oncology patients: new perspectives. J Adolesc Young Adult Oncol. 2018;7(3):263–9.
87. Nightingale CL, et al. Health-related quality of life of young adult survivors of childhood cancer: a review of qualitative studies. J Adolesc Young Adult Oncol. 2011;1(3):124–32.

88. Meadows AT, et al. Declines in IQ scores and cognitive dysfunctions in children with acute lymphocytic leukaemia treated with cranial irradiation. Lancet. 1981;2(8254):1015–8.
89. Butler RW, Mulhern RK. Neurocognitive interventions for children and adolescents surviving cancer. J Pediatr Psychol. 2005;30(1):65–78.
90. Copeland DR, et al. Neuropsychological effects of childhood cancer treatment. J Child Neurol. 1988;3(1):53–62.
91. Kazak AE, Meadows AT. Families of young adolescents who have survived cancer: social-emotional adjustment, adaptability, and social support. J Pediatr Psychol. 1989;14(2):175–91.
92. Lancashire ER, et al. Educational attainment among adult survivors of childhood cancer in Great Britain: a population-based cohort study. J Natl Cancer Inst. 2010;102(4):254–70.
93. Mitby PA, et al. Utilization of special education services and educational attainment among long-term survivors of childhood cancer: a report from the Childhood Cancer Survivor Study. Cancer. 2003;97(4):1115–26.
94. Zeltzer LK, et al. Psychological status in childhood cancer survivors: a report from the Childhood Cancer Survivor Study. J Clin Oncol. 2009;27(14):2396–404.
95. Smith AW, et al. Unmet support service needs and health-related quality of life among adolescents and young adults with cancer: the AYA HOPE study. Front Oncol. 2013;3:75.
96. Smith AW, et al. Health-related quality of life of adolescent and young adult patients with cancer in the United States: the adolescent and young adult health outcomes and patient experience study. J Clin Oncol. 2013;31(17):2136–45.
97. Peterson CC, Drotar D. Family impact of neurodevelopmental late effects in survivors of pediatric cancer: review of research, clinical evidence, and future directions. Clin Child Psychol Psychiatry. 2006;11(3):349–66.
98. Nathan PC, et al. Adverse mental health outcomes in a population-based cohort of survivors of childhood cancer. Cancer. 2018;124(9):2045–57.
99. Brinkman TM, et al. Prevalence and predictors of prescription psychoactive medication use in adult survivors of childhood cancer: a report from the Childhood Cancer Survivor Study. J Cancer Surviv. 2013;7:104–14.
100. Schultz KA, et al. Behavioral and social outcomes in adolescent survivors of childhood cancer: a report from the childhood cancer survivor study. J Clin Oncol. 2007;25(24):3649–56.
101. Zebrack BJ, et al. Psychological outcomes in long-term survivors of childhood leukemia, Hodgkin's disease and non-Hodgkin's lymphoma: a report from the childhood cancer survivor study. Pediatrics. 2002;110:42–52.
102. Stuber ML, et al. Prevalence and predictors of posttraumatic stress disorder in adult survivors of childhood cancer. Pediatrics. 2010;125:e1124–34.
103. Turner JK, Hutchinson A, Wilson C. Correlates of posttraumatic growth following childhood and adolescent cancer: a systematic review and meta-analysis. Psychooncology. 2018;27(4):1100–9.
104. Johannsdottir IMR, et al. Adverse health outcomes and associations with self-reported general health in childhood lymphoma survivors. J Adolesc Young Adult Oncol. 2017;6(3):470–6.
105. Johannsdottir IM, et al. Increased prescription rates of anxiolytics and hypnotics to survivors of cancer in childhood, adolescence, and young adulthood-a population-based study. Pediatr Blood Cancer. 2018;65(2).
106. Pierson C, Waite E, Pyykkonen B. A meta-analysis of the neuropsychological effects of chemotherapy in the treatment of childhood cancer. Pediatr Blood Cancer. 2016;63(11):1998–2003.
107. Arden-Close E, et al. Gender differences in self-reported late effects, quality of life and satisfaction with clinic in survivors of lymphoma. Psychooncology. 2011;20(11):1202–10.
108. Lu Q, et al. Pain in long-term adult survivors of childhood cancers and their siblings: a report from the Childhood Cancer Survivor Study. Pain. 2011;152(11):2616–24.
109. Hammersen F, et al. Sleep quality and health-related quality of life among long-term survivors of (non-) Hodgkin lymphoma in Germany. PLoS One. 2017;12(11):e0187673.
110. Smith K, et al. Alterations in self-perceptions among adolescent cancer survivors. Cancer Investig. 1991;9(5):581–8.
111. Hall A, et al. Supportive care needs of hematological cancer survivors: a critical review of the literature. Crit Rev Oncol Hematol. 2013;88(1):102–16.
112. Buchbinder D, Casillas J, Zeltzer L. Meeting the psychosocial needs of sibling survivors: a family systems approach. J Pediatr Oncol Nurs. 2011;28(3):123–36.
113. Smith SK, et al. Is there a relationship between posttraumatic stress and growth after a lymphoma diagnosis? Psychooncology. 2014;23(3):315–21.
114. Hewitt M, Weiner SL, Simone JC. Childhood cancer survivorship: improving care and quality of life. Washington, DC: The National Academies Press; 2003.
115. Hewitt M, Greenfield S, Stovall E, editors. From cancer patient to cancer survivor: lost in transition. Washington, DC: National Academies Press; 2005.
116. Mulder RL, et al. Recommendations for breast cancer surveillance for female survivors of childhood, adolescent, and young adult cancer given chest radiation: a report from the International Late Effects of Childhood Cancer Guideline Harmonization Group. Lancet Oncol. 2013;14(13):e621–9.
117. Clement SC, et al. Balancing the benefits and harms of thyroid cancer surveillance in survivors of childhood, adolescent and young adult cancer: recommendations from the International Late Effects of Childhood Cancer Guideline Harmonization Group in collaboration with the PanCareSurFup Consortium. Cancer Treat Rev. 2017;63:28–39.
118. Armenian SH, et al. Recommendations for cardiomyopathy surveillance for survivors of childhood cancer: a report from the International Late Effects of Childhood Cancer Guideline Harmonization Group. Lancet Oncol. 2015;16(3):e123–36.
119. van Dorp W, et al. Recommendations for premature ovarian insufficiency surveillance for female survivors of childhood, adolescent, and young adult cancer: a report from the International Late Effects of Childhood Cancer Guideline Harmonization Group in collaboration with the PanCareSurFup Consortium. J Clin Oncol. 2016;34(28):3440–50.
120. Skinner R, et al. Recommendations for gonadotoxicity surveillance in male childhood, adolescent, and young adult cancer survivors: a report from the International Late Effects of Childhood Cancer Guideline Harmonization Group in collaboration with the PanCareSurFup Consortium. Lancet Oncol. 2017;18(2):e75–90.
121. Nathan PC, et al. Family physician preferences and knowledge gaps regarding the care of adolescent and young adult survivors of childhood cancer. J Cancer Surviv. 2013;7(3):275–82.
122. Suh E, et al. General internists' preferences and knowledge about the care of adult survivors of childhood cancer: a cross-sectional survey. Ann Intern Med. 2014;160(1):11–7.
123. Nathan PC, et al. Medical care in long-term survivors of childhood cancer: a report from the childhood cancer survivor study. J Clin Oncol. 2008;26(27):4401–9.
124. Patte C, et al. The Société Française d'Oncologie Pédiatrique LMB89 protocol: highly effective multiagent chemotherapy tailored to the tumor burden and initial response in 561 unselected children with B-cell lymphomas and L3 leukemia. Blood. 2001;97(11):3370–9.
125. Reiter A, et al. Improved treatment results in childhood B-cell neoplasms with tailored intensification of therapy: a report of

the Berlin-Frankfurt-Münster Group trial NHL-BFM 90. Blood. 1999;94(10):3294–306.
126. Woessmann W, et al. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. Blood. 2005;105(3):948–58.
127. Link MP, et al. Treatment of children and young adults with early-stage non-Hodgkin's lymphoma. N Engl J Med. 1997;337(18):1259–66.
128. Le Deley MC, et al. Prognostic factors in childhood anaplastic large cell lymphoma: results of a large European intergroup study. Blood. 2008;111(3):1560–6.
129. Amos Burke GA, et al. Localized non-Hodgkin's lymphoma with B-cell histology: cure without cyclophosphamide? A report of the United Kingdom Children's Cancer Study Group on studies NHL 8501 and NHL 9001 (1985-1996). Br J Haematol. 2003;121(4):586–91.
130. Laver JH, et al. Advanced-stage large-cell lymphoma in children and adolescents: results of a randomized trial incorporating intermediate-dose methotrexate and high-dose cytarabine in the maintenance phase of the APO regimen: a Pediatric Oncology Group phase III trial. J Clin Oncol. 2005;23(3):541–7.
131. Mora J, et al. Lymphoblastic lymphoma of childhood and the LSA2-L2 protocol: the 30-year experience at Memorial-Sloan-Kettering Cancer Center. Cancer. 2003;98(6):1283–91.
132. Patte C, et al. Results of the LMT81 protocol, a modified LSA2L2 protocol with high dose methotrexate, on 84 children with non-B-cell (lymphoblastic) lymphoma. Med Pediatr Oncol. 1992;20(2):105–13.
133. Pullen J, et al. Significance of commonly used prognostic factors differs for children with T cell acute lymphocytic leukemia (ALL), as compared to those with B-precursor ALL. A Pediatric Oncology Group (POG) study. Leukemia. 1999;13(11):1696–707.
134. Abromowitch M, et al. Shortened intensified multi-agent chemotherapy and non-cross resistant maintenance therapy for advanced lymphoblastic lymphoma in children and adolescents: report from the Children's Oncology Group. Br J Haematol. 2008;143(2):261–7.

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