Curr Oncol Rep (2017) 19: 74
DOI 10.1007/s11912-017-0629-y

Double-Hit Large B Cell Lymphoma

Yousef Khelfa[1] · **Yehuda Lebowicz**[1] · **Muhammad Omer Jamil**[1]

Published online: 26 September 2017

Abstract Diffuse large B cell lymphoma (DLBCL) is the most common type of non-Hodgkin lymphoma (NHL), accounting for approximately 25% of NHL cases. It is a heterogeneous group of diseases. *BCL2, BCL6*, and *MYC* are the most frequent mutated genes in DLBCL. Double-hit lymphoma (DHL) is an aggressive form of DLBCL with an unmet treatment need, in which *MYC* rearrangement is present with either *BCL2* or *BCL6* rearrangement. Patients typically present with a rapidly growing mass with B symptoms. DHL has been linked to very poor outcomes when treated with RCHOP chemotherapy. Dual-expressor lymphoma is a form of DLBCL with overexpression of MYC and BCL2/BCL6. There is a paucity of prospective trials evaluating the treatment of DHL. Retrospective series suggest that more aggressive treatment regimens such as DA-EPOCH and hyper CVAD may be more efficacious. However, there remains a lack of consensus regarding optimal treatment for DHL. Further clinical trials, including novel agents, are needed for improvement in outcomes.

Keywords Double-hit lymphoma · DLBCL · MYC · BCL2 · BCL6

Introduction

Diffuse large B cell lymphoma (DLBCL) is a clinically and genetically heterogeneous disease. *MYC, BCL2*, and *BCL6* are the three most commonly altered oncogenes in DLBCL. *BCL2* (chromosome 18q21) is altered in more than 30% of DLBCL and does not impact DLBCL outcomes when present as a sole abnormality [1]. *BCL6* (chromosome 3q27) alteration is seen in about a third of patients and has no prognostic value when present alone [2]. However, *MYC* (chromosome 8q24) rearrangement is seen in up to 15% of cases of DLBCL and portends a worse prognosis following treatment with standard combination chemotherapy [3]. Double-hit lymphoma (DHL) is an aggressive form of DLBCL in which *MYC* rearrangement is coexistent with *BCL2* or less commonly *BCL6* rearrangement. When all three alterations are present, it is known as triple-hit lymphoma (THL) [1]. IHC can be used to identify MYC, BCL2, and BCL6 overexpression, but these are not diagnostic of DHL. DLBCL with overexpression of MYC and BCL2/BCL6 is referred to as dual- or double-expresser lymphoma (DEL) [4]. *MYC* and *BCL2* rearrangements are more common than *MYC* and *BCL6* in DHL [5]. DHL is typically of germinal center B cell origin (GCB, GC COO), while DEL is usually non-GCB. Both forms are usually aggressive, and standard R-CHOP therapy may not be adequate. In its most recent classification, WHO reclassified DHL from DLBCL NOS, or B cell unclassifiable between DLBCL and Burkitt lymphoma to high-grade B cell lymphoma, with *MYC* and *BCL2* and/or *BCL6* rearrangements [6]. In this review, we discuss the biology, testing for, and treatment of DHL in light of available literature.

Biology of DHL

The introduction of gene expression profiling (GEP) has helped in classifying DLBCL under two main groups, the germinal center B cell-like type (GCB) and the post germinal center B cell-like type (non-GCB), often called activated B

This article is part of the Topical Collection on *Lymphomas*

✉ Muhammad Omer Jamil
Omerjamil83@hotmail.com

[1] Division of Hematology and Oncology, Department of Internal Medicine, Marshall University School of Medicine-Edwards Comprehensive Cancer Center, 1400 Hal Greer Boulevard, Huntington, WV 25701, USA

cell type (ABC) [7]. GCB subtype has a better five-year survival with current therapy (CHOP or R-CHOP) as compared to non-GCB lymphomas [8]. Since GEP is not yet a commercially available tool, clinicians rely more on different algorithms using available IHC stains to differentiate the two types, all of whom show similar degree of correlation with GEP [9].

BCL2 rearrangement [t(14;18) (q32;q21)] is more common in GCB lymphomas vs non-GCB subtype (30 vs. < 5%). The expression of BCL2 protein does not always correlate with *BCL2* rearrangement [t(14;18)]. In one study, BCL2 protein expression was observed in 44% of GCB and 62% of non-GCB DLBCL [10]. BCL2 overexpression was correlated with a poor outcome with R-CHOP therapy only in the GCB subtype. Same study showed that the non-GCB subtype of DLBCL rarely has the t(14;18), yet amplifications of 18q21 were seen in up to two thirds of cases, which could possibly provide mechanism for BCL2 overexpression in these tumors [10]. Similarly, MYC overexpression is found in much higher frequency in DLBCL as compared to *MYC* rearrangement [11], suggesting other mechanisms, such as copy number variations, mutations, and transcriptional upregulation (by B cell receptor and NF-κB signaling) may be responsible. *MYC* rearrangement in DLBCL is associated with increased risk of CNS relapse, poor progression-free survival (PFS), and overall survival (OS) with conventional therapy [12, 13]. These lymphomas also present with complex karyotype [14] in contrast with Burkitt lymphoma. This poor prognosis is largely seen when *MYC* rearrangement is present in conjunction with *BCL2* and/or *BCL6* rearrangement (DHL) [3, 15, 16]. In 90% of DHL, *MYC*, and *BCL2*, rearrangements are found [17, 18]. It has been shown that the combination of genetic abnormalities, *MYC*, and *BCL2* rearrangements is predictive of refractory disease to conventional therapy (e.g., R-CHOP) [19]. Similarly, triple-hit lymphoma (THL) with rearrangement in *MYC*, *BCL2*, and *BCL6* has also been defined; these are rare and are also associated with poor prognosis [20].

A study looking at mutational profiling showed DHL to fall intermediate between Burkitt lymphoma (BL) and DLBCL, hence the previous WHO category of B cell lymphoma NOS [21]. In this study, DLBCL pattern showed mutations in *BCL2, EZH2, CREBBP, EP300, MEF2B*, and *SGK1*, while BL pattern showed mutations in BL-associated genes including *ID3/TCF3*, *CCND3*, and *MYC*. DHL and DLBCL with *MYC* rearrangement only showed pattern which was combined between DLBCL and BL. *MYC/BCL2* rearranged DHL shows increased incidence of *TP53* mutation when compared with *MYC/BCL6* [22].

In DHL, *BCL2* translocation is thought to be the first hit followed by *MYC* which could be an early or a late event in germinal centers leading to DHL [23, 24]. DHL often has alterations in genes in addition to *MYC, BCL2*, and *BCL6* like *TP53* [17]. In some studies, high expression of BCL2 and MYC has been associated with worse risk of relapse and poor OS [25, 26], while smaller studies show that DHL (about 15–20%) with low or no expression of MYC and BCL2/BCL6 may have relatively better prognosis despite presence of characteristic translocations [3, 27–29].

Clinical Presentation and Diagnosis of DHL

There is lack of consensus regarding which patients need to be tested for DHL status. Both NCCN and ESMO do not require, rather suggest evaluation in their most recent guidelines [30, 31]. A majority of DHL present as high-grade lymphomas with features suggesting poor prognosis, often with CNS disease and high stage [3, 32•]. In a cohort of Japanese patients, the presence of B symptoms, extra nodal sites, advanced stage, high serum lactate dehydrogenase level, and bone marrow involvement was more prevalent in patients with DHL [33]. Landsburg et al. published a series of 53 patients, in which the double gene rearrangements were detected in 32% of patients; however, no baseline factors, including age International Prognostic Index (IPI) score or histology, were statistically significantly associated with DHL status [34]. In another study of 152 patients with high-grade B cell non-Hodgkin lymphoma (NHL), 21 patients displayed *MYC* rearrangement, 9 of whom had a concurrent *BCL2* rearrangement with one patient with triple hit. The overall frequency of DHL was 6%. All patients had GCB/GC COO by Hans criteria and suggested referral of all patients with DLBCL for *MYC* rearrangement testing at the time of initial diagnosis [35]. Fluorescence in situ hybridization (FISH) testing in all patients with large B cell lymphoma have identified DHL with low-risk features, with somewhat better outcomes [36]. While these patients are usually treated with aggressive regimens as DHL, it is unclear if this is helpful in this subset of patients. One strategy would be to test all patients presenting with high-grade large B cell lymphomas = with IHC for MYC, BCL2, and BCL6, which is rather inexpensive, and if positive, further testing with FISH should be undertaken. As previously discussed, high expression of these proteins in addition to FISH testing portends worse prognosis compared to patients positive for FISH abnormality for DHL but no overexpression. The downside to this strategy is the inter-observer variability in reporting expression of these proteins. There is no consensus what is considered high expression. A good reference point would be 40% for MYC and 50% for BCL2, as this has been shown to have prognostic significance [28]. Another starting point could be to evaluate patients presenting with GCB DLBCL, as 95% of DHL are GCB subtype which can decrease the number of patients to be tested by half, keeping in mind that only a small fraction of GCB DLBCL will be DHL [17, 28].

DHL Treatment

Initial Treatment of DHL

Due to low incidence and resultant paucity of prospective trials in DHL, there is no consensus about the standard treatment for DHL. DHL treatment studies available are listed in Table 1.

One study evaluated the outcomes of patients with MYC+ and DHL and demonstrated that only age and achievement of complete response (CR) were correlated with better outcomes. After median follow up of 28.5 months, the median PFS for patients with documented double-hit NHL who achieved a CR had not yet been reached (95% CI, NR to NR) vs 3.9 months (95% CI, 1.8–8.0 months) for those with no CR ($p < .0001$). The median OS for those patients who did not achieve a physician-assessed CR was 7.0 months (95% CI, 2.0–12.5 months) compared with a median OS not reached for those who did achieve a CR (95% CI, NR to NR; $p < .00001$) [44].

Petrich et al. [37] published a multicenter retrospective analysis looking at DHL patients, comparing the outcomes of those that received standard R-CHOP vs more intensive regimens (R-Hyper CVAD, DA R-EPOCH, or R-CODOX-M/IVAC). Patients who had CR and underwent autologous stem cell transplant (ASCT) were compared with patients who had CR and did not get ASCT. After a median follow up of 23 months, the median PFS and OS for all patients were 10.9 and 21.9 months, respectively, with no difference in OS for those that received intensive regimens vs R-CHOP. However, median PFS was significantly better for intensive regimen patients over R-CHOP patients 26.6 vs 7.8 months ($p = 0.0463$ for DA R-EPOCH group, $p = 0.001$ for R-HyperCVAD group and $p = 0.036$ for R-CODOX/M IVAC group). Of note, there was no difference between the three intensive treatment regimens. In multivariate analysis, advanced stage, central nervous system involvement, leukocytosis, and LDH, > 3 times the upper limit of normal were associated with higher risk of death. When corrected for these variables, intensive induction was associated with improved OS. Based on these findings, a novel risk score for DHL was developed based on PS ≥ 2 and bone marrow involvement with 0–2 scoring, which divides patients into high-risk (2), intermediate-risk (1), and low-risk (0) groups. In another study, Oki et al. [39•] analyzed the outcome of 129 cases of DHL; DHL was defined as B cell lymphoma with translocations and/or extra signals involving *MYC* plus *BCL2* and/or *BCL6*. The 2-year event-free survival (EFS) rate in all patients was 33%; however, when analyzed by individual regimen, those who received R-CHOP, R-EPOCH, and R-HyperCVAD/MA had 2-year EFS of 25, 67, and 32%, respectively.

A multicenter phase II study using R-DA-EPOCH for MYC rearranged aggressive B cell lymphoma demonstrated, though with a short-term median follow-up time of 14 months, PFS, time to progression (TTP), and OS were 79, 86, and 77%, respectively [43]. A phase I study incorporating lenalidomide (LEN) into dose-adjusted EPOCH plus rituximab (DA-EPOCH-R) in patients with double-hit (DHL) or double-expressing (DEL) lymphomas was recently presented and concluded that LEN can safely be added to DA-EPOCH-R in DHL and DEL patients and preliminary safety/efficacy data appear promising. With a median follow-up of 10.7 months (range 1.3–18.6 months), for 15 patients enrolled in the study, OS was 93%. A phase II study in this patient population with LEN + DA-EPOCH-R is underway [45].

First-line consolidation with autologous stem cell transplant (ASCT) has been evaluated and the results are mixed. In one study with 311 patients, there was a trend seen toward improved survival with first-line consolidation with ASCT in patients who achieved CR with first-line induction chemotherapy [32•]. SWOG9704 also showed a trend in improved PFS with ASCT after first-line RCHOP, but study included only one DHL patient who had ASCT($n = 1$) [46]. In the study by Oki et al., ASCT after CR did not improve OS in patients achieving complete response with initial therapy ($n = 71$); 2-year EFS rates in patients who did ($n = 23$) or did not ($n = 48$) receive frontline stem cell transplantation were 68 and 53%, respectively ($p = 0{\cdot}155$). A prospective study recently published looked at patients with DHL receiving ASCT after achieving first CR [47•]. A total of 159 patients who achieved CR after RCHOP or intensive therapy were included. No difference in 3-year relapse-free survival (RFS) or OS was seen in patients who achieved ASCT vs no ASCT. Patients showed inferior 3-year RFS with RCHOP-based induction vs intensive therapy (56 vs. 88%, respectively).

Treatment of Relapsed DHL

Standard treatment for relapsed DHL has not been established yet. Most recently, in a phase II study, the efficacy of autologous T cells genetically modified to express a chimeric antigen receptor consisting of an external anti-CD19 single-chain murine antibody domain with CD3ζ and 4-1BB signaling domains (CTL019 cells) was tested in patients with relapsed/refractory GCB and non-GCB DLBCL, DHL, and transformed follicular lymphoma (tFL). Thirteen patients (7 pts. GCB, 5 pts. NGC, 1 undetermined) were enrolled. The median number of prior therapies were 5 (range 2–8) and number of pts. with prior transplant 7 (54%). Lymphodepleting chemotherapy regimens were given prior to giving CTL019 cells. At 3 months' post CTL019, overall response rate (ORR) was 52% (7/13 pts); ORR for GCB 71% (5/7 pts) and non-GCB was 40% (2/5 pts). Best response for all patients was CR in 46%, CR for GCB 57%, and non-GCB 40%. Three of 7 pts. with GC DLBCL had tFL, and all 3 achieved CR; two of seven patients with GCB DLBCL had DHL and both

Table 1 Currently published studies on DHL

	Year	No. of patients and lymphoma types	Type of study	Treatment	Median age	Results
Johnson et al. [3]	2009	54 DLEICL or BCLU in 93%	Retrospective analysis	CHOP ± R; (63%); HD chemo; other	52% were > 60 years of age	Median OS 1.4 years and 1 years in R-CHOP and CHOP
Dunleavy et al.	2013	66; 20% with high MYC/BCL2	Retrospective analysis from a prospective study	EPOCH-R	43 years old	10-year survival compared in 4 groups: MYC+/BCL2+ vs all others (MYC+/BCL2-, MYC-BCL2+, MYC−/BCL2-). Global $p = 0.5$ (PFS) and $p = 0.8$ (OS). R-EPOCH overcome inferiority of DHL
Petrich et al. [37]	2014	311 (100%)	Multicenter retrospective analysis	DA EPOCH-R 64 (21%) R-HYPERCVAD 65 (21%) R-CODOX-MIVAC 42 (14%) R-CHOP 100 (32%)	60 years old	mFollow-up 23 months mPFS 10.9 months, mOS 21.9 months SCT after CR/all regimen on OS benefit. Better mPFS 26.6 months all intensive regimens vs R-CHOP 7.8 months DA-EPOCH-R resulted in superior CR compared with R-CHOP and other intensive regimens ($p < 0.05$).
Snuderl et al. [18]	2010	20 (100%)	Single-institution retrospective analysis	R-ICE + MTX/ASCT (1); CHOP (1); RCHOP (3); RCHOP + MTX (6); RCHOP + MTX ASCT (1); R-E POCH + MTX (3); CODOX- + MTX/R-IVAC (3); P (1); NK(1)	64 years old	ORR 10/20** (50) mOS 0.38 years
Lin et al. [38]	2012	52; DLEICL or BCLU in > 90%	Retrospective analysis	R-CHOP or R-Hyper-CVAD	55 years old	Median OS of 13.6 months; more intense therapy ($p = 0.54$) or SCT ($p = 0.73$) was not associated with a better outcome
Oki et al. [39•]	2014	129 (72% MYC/BCL2)	Single-institution retrospective analysis	R-EPOCH R-HYPERCVAD/MA R-CHOP	62 years old	Overall 2-year EFS 33% Better OS R-EPOCH vs R-CHOP (p value of 0.057) SCT did not improve OS. CR R-EPOCH (68%), R-HYPERCVAD (70%), R-CHOP (20%)
Nitsu et al. [33]	2009	19 (100%)	Retrospective analysis from a prospective study	CyclOBEAP (6); CHOP + HD MTX (3); CHOP (4); RCHOP (3), CyclOBEAP □ + R (3)	61 years old	ORR 17/19 (89%) mOS 1.5 years
Tomita et al. [40]	2009	27 (100%)	Retrospective analysis	CHOP or CODOX-M/IVAC or HyperCVAD (+ R, $n = 14$; − R, $n = 3$)	51 years old	ORR 6/23 (26%) mOS 0.5 years
Le Gouill et al. [41]	2007	16 (100%)	Retrospective analysis	CEEP/CO PAD M + auto-SCT/BEAM (1); CHOP/IVAM (1); CO PADM/CYVE (3); COPADM (1); COPADM + auto-SCT/BEAM (1); COPADM + allo-SCT/Bu/Cy (1) CEEP/DHAP + auto-SCT/BEAM (1); RCHOP (4); CHOP (1); Steroids# (1); R-CEEP allo-SCT/TBI/Cy (1)	61 years old	ORR 12/16 (75%) mOS 0.42 years
Kanugo et al. [42]	2006	14 (100%)	Retrospective analysis	CT-NOS (11); R¶ (1); CT and BMT (1); CT, EIMT, and RT (1)	55 years old	< 1 year

Table 1 (continued)

	Year	No. of patients and lymphoma types	Type of study	Treatment	Median age	Results
Dunleavy et al. [43]	2014	52 (45%)	Prospective analysis of Myc-rearranged aggressive B cell lymphoma	R-DA-EPOCH	61 years old	14 months 0S 79% 14 months PFS 86%

DLBCL diffuse large B cell lymphoma; *BCLU* B cell lymphoma unclassified; *R-CHOP* rituximab, cyclophosphamide, doxorubicin, vincristine, prednisone; *OS* overall survival; *PFS* progression-free survival; *m* median; *DA R-EPOCH* dose-adjusted, rituximab, etoposide, prednisone, vincristine, doxorubicin, cyclophosphamide; *R-HyperCVAD* riruximab, cyclophosphamide, vincristine, doxorubicin, prednisone; *MA* methotrexate, cytarabine; *CODOX-MIVAC* vincristine, cyclophosphamide, doxorubicin, cytarabine, ifosfamide, mesna, etoposide, cytarabine; *COPAD* cyclophosphamide, doxorubicin, vincristine, prednisone; *CycloBEAP* cyclophosphamide, vincristine, bleomycin, etoposide, doxorubicin, prednisolone; *AutoSCT* autologous stem cell transplant; *BEAM BCNU* etoposide, cytarabine, melphalan; *DHAP* dexamethasone, cytarabine, and cisplatin

achieved CR. At the time of this data presentation, no patient achieving CR relapsed. Median PFS was 5.8 months for all patients, 3.0 months for non-GCB patients, and not reached for GCB patients (57.1% [95%CI: 17.2–83.7%] progression-free at median follow-up 21.9 months). At median follow-up 23.3 months for responding patients, 85.7% [95%CI: 33.7–97.9%] maintain response [48]. KTE-C19, which is autologous CD3ζ/CD28 chimeric antigen receptor (CAR) modified T cells, has shown promising results. ZUMA 1 study of KTE-C19 included two cohorts, one with relapsed/refractory DLBCL (N = 72) and the other with transformed follicular lymphoma and primary mediastinal B cell lymphoma. In the intention to treat analysis, an ORR of 82% was seen in DLBCL patients with 49% CR rate. Median duration of response was 8.2 months with majority of patients having durable responses if they achieved CR [49•].

ASCT for patients in second line has been evaluated as well in a retrospective series of 117 patients having DEL and DHL [50]. The study showed inferior outcomes for patients who underwent ASCT.

Summary

Considering the available data, it can be concluded that DHL has an inferior outcome when treated with standard R-CHOP therapy. Further, the data imply that this inferior outcome may be overcome by using more intense regimens such as DA R-EPOCH, R-Hyper CVAD, or R-CODOX/M IVAC. Based on the MD Anderson Experience, DA R-EPOCH may be best tolerated while maintaining the largest improvement in PFS. Until a prospective trial demonstrates improved survival, these suggestions will remain somewhat speculative. Several prospective trials are ongoing that will hopefully answer some of these questions. Several of these trials are looking at multiple targets involved in the pathogenesis of lymphoma at the molecular and genetic level.

Future Directions

Multiple pathways have been studied in the treatment of double-hit lymphoma. Preclinical data has shown that a second-generation proteasome inhibitor, MLN9708/ixazomib, degrades MYC and induces cell death at nanomolar concentrations in lymphoma models and resulted in significant tumor/growth inhibition ($p < 0.001$) and improvement in survival ($p < 0.001$) compared with controls in Jurkat and L540-derived SCID xenograft models [51]; however, its single-agent activity was modest in relapsed/refractory lymphoma in another study [52]. Of 22 response-evaluable patients, one achieved CR, three achieved PR, and four patients had stable disease (SD).

Table 2 Ongoing studies in treatment of DHL

ID number	Therapy/pathway	Trial phase	Disease
NCT02272686	Ibrutinib/Bruton's tyrosine kinase	Phase 2	DHL
NCT02213913	Lenalidomide/DA EPOCH-R	Phase 1/2	MYC-associated B cell lymphomas
NCT01856192	(R2CHOP) vs RCHOP	Phase 2	DLBCL
NCT01092182	DA EPOCH-R ± linalidomide	Phase 2	Burkitt lymphoma and c-MYC+ DLBCL
NCT02110563	DCR-MYC/MYC	Phase 1	Refractory NHL
NCT01949883	CPI-0610/BET (bromodomain and extra-terminal) proteins	Phase 1	Lymphoma
NCT01181271	Tandem auto-allo transplant for lymphoma	Phase 2	DLBCL
NCT02226965	PNT2258/DNAi, BCL-2	Phase 2	Relapsed or refractory DLBCL
NCT01897012	Alisertib and romidepsin/aurora A kinase	Phase 1	MYC+/DLBCL
NCT01490723	Zevalin-containing nonmyeloablative conditioning for SCT	Phase 2	Lymphoma
NCT01943851	GSK525762/BET (bromodomain and extra-terminal) proteins	Phase 1/2	Relapsed, refractory hematologic malignancies
NCT02674750	CUDC-907/dual HDAC and MYC inhibitor	Phase 2	Relapsed refractory DLBCL with MYC

BTk Bruton's tyrosine kinase; *R-CHOP* rituximab, cyclophosphamide, doxorubicin, vincristine, prednisone; *OS* overall survival; *PFS* progression-free survival; *m* median; *DA R-EPOCH* dose-adjusted, rituximab, etoposide, prednisone, vincristine, doxorubicin, cyclophosphamide; *HDAC* histone deacetylase

The mammalian target of rapamycin (mTOR) complex 1-dependent evasion of senescence is essential for cellular transformation and lymphoma maintenance by MYC in B-lymphocytes. In one study, everolimus selectively cleared premalignant B cells from the bone marrow and spleen, restored a normal pattern of B cell differentiation, and strongly protected against lymphoma development. Established Eμ-Myc lymphoma also regressed after everolimus therapy [53]. In a phase II study, temsirolimus (an mTOR inhibitor) demonstrated a single-agent activity in DLBCL with an overall and complete response rate of 28.1 and 12.5%, respectively, and median PFS of 2.6 months and median OS of 7.2 months. [54]. The platelet-sparing BCL2 inhibitor, ABT-199, has single-agent activity shown in a preliminary report in three of the eight patients (38%) with relapsed/refractory DLBCL treated in the higher-dose cohorts (at doses ≥ 600 mg) [55]. Another study showed ABT-199 enhancement of the antitumor activity of chemotherapy agents including doxorubicin, cytarabine, and bortezomib in DHL cell lines [56].

Early preclinical data demonstrated that an aurora A kinase inhibitor (Aki) in combination with a histone deacetylase inhibitor enhanced lymphoma cell death through repression of MYC and MYC-responsive microRNAs. In one study, either of the AKi alone at 100 to 500 nmol/L resulted in approximately 50% reduced cell growth and 10 to 40% apoptosis. Addition of vorinostat reactivated proapoptotic genes and enhanced lymphoma cell death. [57]; however, in a small clinical trial of this combination, the three patients with DHL developed progressive disease [58]. In another clinical trial of a selective aurora A kinase inhibitor in patients with relapsed or refractory DLBCL, only three out of 21 patients had clinical response [59].

Inhibition of the BET bromodomain diminishes the effect of MYC overexpression by preventing signal transduction, essential in regulating *MYC* transcriptional initiation and elongation [60]. JQ1, bromodomain BRD4 inhibitor, showed preclinical activity, including specifically in DHL cell lines [56].

Clinical trials of the Bruton's tyrosine kinase inhibitor ibrutinib [61], and phosphatidylinositol-3-kinase delta isoform inhibitor idelalisib, showed activity for inhibitors of signaling downstream of the B cell receptor in DHL [62]. In a phase III, multicenter, randomized, double-blind, placebo-controlled study, the efficacy and safety of idelalisib were assessed in combination with rituximab vs rituximab plus placebo. Two hundred and twenty patients were assigned to receive rituximab and either idelalisib (at a dose of 150 mg) or placebo twice daily. The median PFS was 5.5 months in the placebo group and was not reached in the idelalisib group (hazard ratio for progression or death in the idelalisib group, 0.15; $p < 0.001$). Patients receiving idelalisib vs those receiving placebo had improved rates of overall response (81 vs. 13%; odds ratio, 29.92; $p < 0.001$) and OS at 12 months (92 vs. 80%; hazard ratio for death, 0.28; $p = 0.02$) [63].

Other promising therapies for double-hit lymphoma include modified autologous T cells engineered to recognize other B cell surface targets such as CD22, CD20, CD30, and

CD79a. which are in various stages of development [62, 64]. Also, small-molecule inhibitors of BCL6 could have potential therapeutic activity as well [65].

In previous translational studies, CUDC-907 treatment has demonstrated dose-dependent decreases in MYC protein expression and a more potent inhibition of MYC expression than various HDAC and PI3K inhibitors, either alone or in combination [66]. In an ongoing study by Landsburg et al., patients with RR DLBCL, including those who have MYC-altered disease per central testing, are being enrolled to receive either CUDC-907 alone or in combination with rituximab (NCT02674750). Multiple ongoing studies are evaluating different pathways in order to improve the outcomes (Table 2).

Conclusion

DHL is an aggressive disease with no consensus on what would be the standard treatment for it. The most consistent result of these retrospective analyses indicates that DHL has an inferior outcome when treated with standard R-CHOP therapy. However, those studies indicate that this inferior outcome may be overcome by using more intense regimens such as R-DA-EPOCH, R-HYPERCVAD, or R CODOX/M IVAC.

Until a prospective trial demonstrates improved survival, these suggestions will remain somewhat speculative.

Compliance with Ethical Standards

Conflict of Interest Yousef Khelfa, Yehuda Lebowicz, and Muhammad Omer Jamil declare that they have no conflict of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

References

Papers of particular interest, published recently, have been highlighted as:
- Of importance

1. Dunleavy K. Double-hit lymphomas: current paradigms and novel treatment approaches. Hematology Am Soc Hematol Educ Program. 2014;2014(1):107–12.
2. Shustik J, Han G, Farinha P, et al. Correlations between BCL6 rearrangement and outcome in patients with diffuse large B-cell lymphoma treated with CHOP or R-CHOP. Haematologica. 2010;95(1):96–101.
3. Johnson NA, Savage KJ, Ludkovski O, et al. Lymphomas with concurrent BCL2 and MYC translocations: the critical factors associated with survival. Blood. 2009;114(11):2273–9.
4. Hu S, Xu-Monette ZY, Tzankov A, et al. MYC/BCL2 protein coexpression contributes to the inferior survival of activated B-cell subtype of diffuse large B-cell lymphoma and demonstrates high-risk gene expression signatures: a report from The International DLBCL Rituximab-CHOP Consortium Program. Blood. 2013;121(20):4021–31. quiz 4250
5. Wagner SD, Amen F, Trivedi PS, Horncastle D, Elderfield K, Naresh KN. Bcl-6 and c-Myc are rarely co-expressed in adult diffuse large B-cell lymphoma. Leuk Lymphoma. 2007;48(8):1510–3.
6. Swerdlow SH, Campo E, Pileri SA, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood. 2016;127(20):2375–90.
7. Alizadeh AA, Eisen MB, Davis RE, et al. Distinct types of diffuse large B-cell lymphoma identified by gene expression profiling. Nature. 2000;403(6769):503–11.
8. Rosenwald A, Wright G, Chan WC, 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.
9. Hans CP, Weisenburger DD, Greiner TC, 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.
10. Iqbal J, Meyer PN, Smith LM, et al. BCL2 predicts survival in germinal center B-cell-like diffuse large B-cell lymphoma treated with CHOP-like therapy and rituximab. Clin Cancer Res. 2011;17(24):7785–95.
11. Chisholm KM, Bangs CD, Bacchi CE, Molina-Kirsch H, Cherry A, Natkunam Y. Expression profiles of MYC protein and MYC gene rearrangement in lymphomas. Am J Surg Pathol. 2015;39(3):294–303.
12. Savage KJ, Johnson NA, Ben-Neriah S, et al. MYC gene rearrangements are associated with a poor prognosis in diffuse large B-cell lymphoma patients treated with R-CHOP chemotherapy. Blood. 2009;114(17):3533–7.
13. Valera A, Lopez-Guillermo A, Cardesa-Salzmann T, et al. MYC protein expression and genetic alterations have prognostic impact in patients with diffuse large B-cell lymphoma treated with immunochemotherapy. Haematologica. 2013;98(10):1554–62.
14. Škunca Ž, Domimis M, Plninc-Peraica A, Jakšić B. Clinical features in DLBCL and translocation BCL2/c-MYC "double hit" lymphoma. Acta Med Croatica. 2014;68(3):299–305.
15. Pedersen MO, Gang AO, Poulsen TS, et al. MYC translocation partner gene determines survival of patients with large B-cell lymphoma with MYC- or double-hit MYC/BCL2 translocations. Eur J Haematol. 2014;92(1):42–8.
16. Pillai RK, Sathanoori M, Van Oss SB, Swerdlow SH. Double-hit B-cell lymphomas with BCL6 and MYC translocations are aggressive, frequently extranodal lymphomas distinct from BCL2 double-hit B-cell lymphomas. Am J Surg Pathol. 2013;37(3):323–32.
17. Aukema SM, Siebert R, Schuuring E, et al. Double-hit B-cell lymphomas. Blood. 2011;117(8):2319–31.
18. Snuderl M, Kolman OK, Chen YB, et al. B-cell lymphomas with concurrent IGH-BCL2 and MYC rearrangements are aggressive neoplasms with clinical and pathologic features distinct from Burkitt lymphoma and diffuse large B-cell lymphoma. Am J Surg Pathol. 2010;34(3):327–40.
19. Friedberg JW. Double-hit diffuse large B-cell lymphoma. J Clin Oncol. 2012;30(28):3439–43.
20. Horn H, Ziepert M, Becher C, et al. MYC status in concert with BCL2 and BCL6 expression predicts outcome in diffuse large B-cell lymphoma. Blood. 2013;121(12):2253–63.
21. Momose S, Weissbach S, Pischimarov J, et al. The diagnostic gray zone between Burkitt lymphoma and diffuse large B-cell lymphoma is also a gray zone of the mutational spectrum. Leukemia England. 2015;29:1789–91.
22. Gebauer N, Bernard V, Gebauer W, Thorns C, Feller AC, Merz H. TP53 mutations are frequent events in double-hit B-cell lymphomas with MYC and BCL2 but not MYC and BCL6 translocations. Leuk Lymphoma. 2015;56(1):179–85.

23. De Jong D, Voetdijk BM, Beverstock GC, van Ommen GJ, Willemze R, Kluin PM. Activation of the c-myc oncogene in a precursor-B-cell blast crisis of follicular lymphoma, presenting as composite lymphoma. N Engl J Med. 1988;318(21):1373–8.
24. Gauwerky CE, Haluska FG, Tsujimoto Y, Nowell PC, Croce CM. Evolution of B-cell malignancy: pre-B-cell leukemia resulting from MYC activation in a B-cell neoplasm with a rearranged BCL2 gene. Proc Natl Acad Sci U S A. 1988;85(22):8548–52.
25. Kluk MJ, Chapuy B, Sinha P, et al. Immunohistochemical detection of MYC-driven diffuse large B-cell lymphomas. PLoS One. 2012;7(4):e33813.
26. Kim S, Nam SJ, Kwon D, et al. MYC and BCL2 overexpression is associated with a higher class of Memorial Sloan-Kettering Cancer Center prognostic model and poor clinical outcome in primary diffuse large B-cell lymphoma of the central nervous system. BMC Cancer. 2016;16:363.
27. Ye Q, Xu-Monette ZY, Tzankov A, et al. Prognostic impact of concurrent MYC and BCL6 rearrangements and expression in de novo diffuse large B-cell lymphoma. Oncotarget. 2016;7(3):2401–16.
28. Johnson NA, Slack GW, Savage KJ, et al. Concurrent expression of MYC and BCL2 in diffuse large B-cell lymphoma treated with rituximab plus cyclophosphamide, doxorubicin, vincristine, and prednisone. J Clin Oncol. 2012;30(28):3452–9.
29. Copie-Bergman C, Cuilliere-Dartigues P, Baia M, et al. MYC-IG rearrangements are negative predictors of survival in DLBCL patients treated with immunochemotherapy: a GELA/LYSA study. Blood. 2015;126(22):2466–74.
30. Zelenetz AD, Gordon LI, Wierda WG, et al. Diffuse large B-cell lymphoma version 1.2016. J Natl Compr Cancer Netw. 2016;14(2):196–231.
31. Tilly H, da Gomes Silva M, Vitolo U, et al. Diffuse large B-cell lymphoma (DLBCL): ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Ann Oncol. 2015;26(Suppl 5):v116–25.
32.• Petrich AM, Gandhi M, Jovanovic B, et al. Impact of induction regimen and stem cell transplantation on outcomes in double-hit lymphoma: a multicenter retrospective analysis. Blood. 2014;124(15):2354–61.
33. Niitsu N, Okamoto M, Miura I, Hirano M. Clinical features and prognosis of de novo diffuse large B-cell lymphoma with t(14;18) and 8q24/c-MYC translocations. Leukemia. 2009;23(4):777–83.
34. Landsburg DJ, Nasta SD, Svoboda J, Morrissette JJ, Schuster SJ. 'Double-Hit' cytogenetic status may not be predicted by baseline clinicopathological characteristics and is highly associated with overall survival in B cell lymphoma patients. Br J Haematol. 2014;166(3):369–74.
35. McHugh DJ GP, Quinn J, Thornton P, Bird B, Sukor S, Fortune A, Parera K, Bacon E, Vandenberghe E, Flavin R, Grant C. Prevalence, clinico-pathological features and outcomes of 'double-hit' high-grade B-cell non-Hodgkins lymphoma (NHL): a single institution experience. Ann Oncol 2016; 27(suppl_6).
36. Pedersen MO, Gang AO, Poulsen TS, et al. Double-hit BCL2/MYC translocations in a consecutive cohort of patients with large B-cell lymphoma—a single centre's experience. Eur J Haematol. 2012;89(1):63–71.
37. Petrich AM, Gandhi M, Jovanovic B, et al. Impact of induction regimen and stem cell transplantation on outcomes in double-hit lymphoma: a multicenter retrospective analysis. Blood. 2014;124(15):2354–61.
38. Lin P, et al. Prognostic value of MYC rearrangement in cases of B-cell lymphoma, unclassifiable, with features intermediate between diffuse large B-cell lymphoma and Burkitt lymphoma. Cancer. 2012;118(6):1566–73.
39.• Oki Y, Noorani M, Lin P, et al. Double hit lymphoma: the MD Anderson Cancer Center clinical experience. Br J Haematol. 2014;166(6):891–901. **Describes the utility of intensive treatment regimens as compared to RCHOP for DHL.**
40. Tomita N, et al. Clinicopathological features of lymphoma/leukemia patients carrying both BCL2 and MYC translocations. Haematologica. 2009;94(7):935–43.
41. Steven LG, et al. The clinical presentation and prognosis of diffuse large B-cell lymphoma with t (14; 18) and 8q24/c-MYC rearrangement. Haematologica. 2007;92(10):1335–42.
42. Kanungo A, et al. Lymphoid neoplasms associated with concurrent t (14; 18) and 8q24/c-MYC translocation generally have a poor prognosis. Mod Pathol. 2006;19(1):25.
43. Dunleavy K, Fanale M, LaCasce A, et al. Preliminary report of a multicenter prospective phase II study of DA-EPOCH-R in MYC-rearranged aggressive B-cell lymphoma. Blood. 2014;124:395.
44.• Cohen JB, Geyer SM, Lozanski G, et al. Complete response to induction therapy in patients with Myc-positive and double-hit non-Hodgkin lymphoma is associated with prolonged progression-free survival. Cancer. 2014;120(11):1677–85. **Mainly describes the importance of first CR.**
45. Nabhan C, Karrison T, Kline J, Cohen K, Bishop MR, Karmali R, et al. Prospective phase I multi-center trial incorporating lenalidomide (LEN) into dose-adjusted epoch plus rituximab (DA-EPOCH-R) in patients with double hit (DHL) or double expressing (DEL) lymphomas: final results. Blood. 2016;128:4191.
46. Puvvada SD, Stiff PJ, Leblanc M, et al. Outcomes of MYC-associated lymphomas after R-CHOP with and without consolidative autologous stem cell transplant: subset analysis of randomized trial intergroup SWOG S9704. Br J Haematol. 2016;174(5):686–91.
47.• Landsburg DJ, et al. Outcomes of patients with double-hit lymphoma who achieve first complete remission. J Clin Oncol 2017; JCO-2017.
48. Schuster SSJ, Dwivedy Nasta S, Chong EA, Winchell N, Landsburg DJ, Porter DL, June CH. Treatment with chimeric antigen receptor modified T cells directed against CD19 (CTL019) results in durable remissions in patients with relapsed or refractory diffuse large B cell lymphomas of germinal center and non-germinal center origin, "Double Hit" diffuse large b cell lymphomas, and transformed follicular to diffuse large B cell lymphomas. Web site. https://ash.confex.com/ash/2016/webprogram/Paper93934.html. Published 2016. Updated.
49.• Locke FL NS, Bartlett NL, et al. Primary results from ZUMA-1: a pivotal trial of axicabtagene ciloleucel (Axi-cel; KTE-C19) in patients with refractory aggressive non-Hodgkin lymphoma (NHL). 2017. **Describes utility of CAR-19 in treatment of DLBCL in second line, this will likely be practice changing.**
50. Herrera AF, Mei M, Low L, et al. Relapsed or refractory double-expressor and double-hit lymphomas have inferior progression-free survival after autologous stem-cell transplantation. J Clin Oncol. 2017;35(1):24–31.
51. Dashnamoorthy R, Kandela I, Bhalla S, Mazar A, Evens AM. Evaluation of potency and the associated biology of the investigational proteasome inhibitor, ixazomib: redox, autophagic, and MAPK-dependent cell death in T-cell lymphoma (TCL) and hodgkin lymphoma (HL) cell lines and human lymphoma xenograft models. Blood. 2013;122:4407.
52. Assouline SE, et al. Phase 1 dose-escalation study of IV ixazomib, an investigational proteasome inhibitor, in patients with relapsed/refractory lymphoma. Blood Cancer J. 2014;4(10):e251.8.
53. Wall M, et al. The mTORC1 inhibitor everolimus prevents and treats Eμ-Myc lymphoma by restoring oncogene-induced senescence. Cancer Discov. 2013;3(1):82–95.
54. Smith SM, et al. Temsirolimus has activity in non–mantle cell non-Hodgkin's lymphoma subtypes: the University of Chicago Phase II Consortium. J Clin Oncol. 2010;28(31):4740–6.

55. Davids MS, Seymour JF, Gerecitano JF, Kahl BS, Pagel JM, Wierda WG, et al. Phase I study of ABT-199 (GDC-0199) in patients with relapsed/refractory (R/R) non-Hodgkin lymphoma (NHL): responses observed in diffuse large B-cell (DLBCL) and follicular lymphoma (FL) at higher cohort doses. ASCO Meeting Abstr. 2014;32(15_suppl):8522.
56. Tam CS, Bassett R, Ledesma C, et al. Mature results of the M. D. Anderson Cancer Center risk-adapted transplantation strategy in mantle cell lymphoma. Blood. 2009;113(18):4144–52.
57. Kretzner L, et al. Combining histone deacetylase inhibitor vorinostat with aurora kinase inhibitors enhances lymphoma cell killing with repression of c-Myc, hTERT, and microRNA levels. Cancer Res. 2011;71(11):3912–20.
58. Fanale MA, Hagemeister FB, Fayad L, Oki Y, Fowler N, Romaguera J, et al. A phase I trial of alisertib plus romidepsin for relapsed/refractory aggressive B- and T-cell lymphomas. Blood. 2014;124:1744.
59. Friedberg JW, et al. Phase II study of alisertib, a selective Aurora A kinase inhibitor, in relapsed and refractory aggressive B-and T-cell non-Hodgkin lymphomas. J Clin Oncol. 2013;32(1):44–50.
60. Delmore JE, Issa GC, et al. BET bromodomain inhibition as a therapeutic strategy to target c-Myc. Cell. 2011;146(6):904–17.
61. Novero A, Ravella PM, Chen Y, Dous G, Liu D. Ibrutinib for B cell malignancies. Exp Hematol Oncol. 32014:4.
62. Lannutti BJ, et al. CAL-101, a p110δ selective phosphatidylinositol-3-kinase inhibitor for the treatment of B-cell malignancies, inhibits PI3K signaling and cellular viability. Blood. 2011;117(2):591–4.
63. Furman RR, et al. Idelalisib and rituximab in relapsed chronic lymphocytic leukemia. N Engl J Med. 2014;370(11):997–1007.
64. Kochenderfer JN, et al. Donor-derived CD19-targeted T cells cause regression of malignancy persisting after allogeneic hematopoietic stem cell transplantation. Blood. 2013;122(25):4129–39.
65. Cerchietti LC, et al. A small-molecule inhibitor of BCL6 kills DLBCL cells in vitro and in vivo. Cancer Cell. 2010;17(4):400–11.
66. Sun K, Atoyan R, Borek MA, et al. Dual HDAC and PI3K inhibitor CUDC-907 downregulates MYC and suppresses growth of MYC-dependent cancers. Mol Cancer Ther. 2017;16(2):285–99.