Lymphoproliferation in Autoimmunity and Sjögren's Syndrome

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Sjögren's syndrome is a chronic inflammatory process involving primarily the exocrine glands. Its association with lymphoma is well documented. A low-grade marginal-zone lymphoma related to mucosa-associated lymphoid tissue is the most common lymphoid neoplasia in Sjögren's syndrome. Among all autoimmune diseases, Sjögren's syndrome is the best tool to clarify the multiple components of autoimmunity and lymphomatogenesis. Herewith, the authors review the literature and discuss the molecular, clinical, histopathologic, and therapeutic aspects of these tumors in Sjögren's syndrome.

Introduction

Over the past years, numerous studies in humans linked several different autoimmune diseases with malignant lymphoproliferation. Adequate studies establish strong associations between malignant non-Hodgkin's lymphoma (NHL) and Sjögren's syndrome (SS) [1], autoimmune thyroiditis [2], and autoimmune hemolytic anemia [3]. The associations reported among NHL and systemic lupus erythematosus (SLE) [5] and rheumatoid arthritis are weaker [5]. There has been speculation that abnormal persistence of lymphocytes because of defective apoptosis, antigen-driven sustained proliferation of lymphocytes by exoantigen or autoantigen, and oncogenic events are usually required before clones become malignant. In order to understand these processes, SS forms the ideal model by dissecting the multiple components of autoimmunity and lymphomatogenesis. The spectrum of lymphoproliferation in SS extends from an increased frequency of circulating monoclonal immunoglobulins (Ig) or free light-chains, presence of mixed monoclonal cryoglobulinemia, and increased levels of circulating CD5-positive B cells to an increased frequency of NHL [6,7]. Malignant lymphoma was first reported in patients with SS in 1963 [8]. The risk of NHL is estimated 44 times greater than that in the normal population [1]. Subsequently, several reports supported the association of lymphoma with SS and recognized NHL as the major complication in the progression of the disease [9,10].

Pathogenesis

From polyclonal B cell activation to monoclonal expansion of B lymphocytes

A notable histologic feature in SS is the lymphoepithelial sialadenitis (LESA) characterized by a lymphoid infiltrate surrounding and infiltrating salivary ducts, with disorganization and proliferation of the ductal epithelial cells forming lymphoepithelial lesions [11]. The phenotype of the immunocompetent cells present in lymphoepithelial lesions, mainly composed of primed $CD4^+$ T lymphocytes, suggests functional structures in which activated B cells (monocytoid or marginal-zone type) produce autoantibodies [12]. These lesions first appear as small clusters and later enlarge to organize lymphoid follicle–like structures with germinal centers. It has been shown that glandular epithelial cells in SS may play an active role as presenting cells in the induction and perpetuation of the inflammatory process [13]. The detection of lymphoid chemokine messenger RNA in ductal epithelial cells, and the expression of CD40 protein by salivary gland epithelial cells, points further to their pathogenetic role in SS, and suggests their possible involvement in lymphoid tissue formation and accumulation of B cells in the inflamed salivary glands [14,15].

Over the past years, it has been demonstrated that monoclonality is the essential process of the transition from the autoimmune state to NHL. The benign LESA is composed of CD4 T cell lymphocytes [16] that secrete interleukin-2 and interferon-gamma [17,18] and of B cell lymphocytes, often oligoclonal, with a risk of progression to B cell lymphoma. Immunophenotyping and immunogenotyping techniques, used in tissue samples from patients with SS, show oligoclonal or monoclonal B cell expansion in several of these cases [19]. This process is mainly generated by the affected exocrine glands, but also arises from visceral organs and lymph nodes. However, monoclonal expansion is not necessarily malignant lymphoma. The issue of whether monoclonal B cell populations in early LESA represent lymphoma or more benign types of expansions has been very controversial. The presence of distinct clones in different biopsies of salivary glands from the same patient indicates that some LESA-

associated clones have not yet evolved into malignant lymphoma [20]. This could be interpreted as a possible suppression of the monoclonal B cell expansion by endogenous immune mechanisms. However, patients with SS exhibiting monoclonal Ig gene rearrangement are at high risk of lymphoma development, and the risk of malignant transformation is higher among patients showing identical B cell clones detected in different tissues at different times [21]. Although there is still a possibility that these clones represent early smoldering lymphoma, clones in benign LESA or early low-grade lymphoma behave clinically with an indolent course and remain localized without evidence of further spread. The most important feature in the differential diagnosis from lymphoma is the number and distribution of monocytoid B cells. If monocytoid B cells are found outside of the lymphoepithelial lesions or as broad anastomosing strands, a diagnosis of lymphoma should be suspected [11].

Mechanisms of lymphoproliferation

The transition of reactive LESA from monoclonality to monoclonal lymphoma is generally considered to represent a multistep process, yet it is poorly understood. There has been speculation that chronic stimulation by exoantigen or autoantigen is playing an essential role in the development of these tumors by driving the proliferation of specific B cells and by increasing the frequency of their transformation [22]. Among patients with SS, the idiotype repertoire of rheumatoid factor (RF) is highly restricted (*ie,* the 17.109 idiotype), because of the frequent utilization of the germ line–encoded Vk III gene segment [23]. Furthermore, B cells expressing cell surface RF (*ie,* encoded by Hum kv325) are frequently detected in the salivary glands [24,25]. This suggests that the clonal expansion in the salivary glands may arise from B cells bearing Ig of a specific crossreactive idiotype.

Antigen selection of specific B cells for transformation was suggested by a previous study of salivary gland mucosa-associated lymphoid tissue (MALT) lymphomas, in which three of five cases used the 51p1 VH and HumKv 325 VL genes that encodes the G6 and 17.109 idiotypes, respectively [26]. Furthermore, in the analysis of the Ig VH gene of 11 distinct LESA-associated clones from sequential biopsies of different patients, eight were derived from a V1-69 VH gene segment, whereas the other three were derived from a V3-7 VH gene segment. The marked VH gene restriction, along with similar amino acid sequence motifs in the complementarity-determining region (CDR3), suggests that LESA-associated clones even from different patients may bind the same or similar antigens and become selected by clonal expansion. In addition, the high rate of ongoing VH gene hypermutations in several cases and the low incidence of replacement mutations in the framework regions further suggest that antigen receptor is playing an important role in the development and expansion of LESA-associated clones [20].

The binding specificity of the surface Ig from these B cells is unknown. It has been speculated that the antigenic peptides may derive from endogenous autoantigens or from exogenous agents, including Epstein-Barr virus–encoded antigens. Previous studies have shown that these lymphomas are not associated with viruses, such as hepatitis C virus, Epstein-Barr virus, human herpes virus-8, or human T lymphotropic virus-1 [27]. There is a strong possibility that LESA-associated clones produce Ig owing RF activity. This could be suggested by heavy- and light-chain CDR3 showing highest similarity to antibodies with RF activity and IgVH genes of LESA-associated clones sharing the same restriction repertoire with RF. Finally, the choice of specific segments, in particular the D21/9 segment, in the assembling of the IgH chains seems to characterize a B cell disorder with the property to produce antibodies with RF activity [20].

It is unlikely that antigenic stimulation by itself could generate a clonal population behaving as malignant. Additional oncogenic events are usually required before clones become malignant, capable of widespread dissemination, and growth, such as inactivation of tumor suppression genes or activation of proto-oncogenes. Compared with other types of NHL where microsatelite instability seems absent, the replication error phenotype appears a common genetic feature of MALT lymphomas detected in approximately 50% of cases, and perhaps relates to the accumulation of genetic aberrations, such as p53 mutations [28]. P53 acting as a cell cycle checkpoint protein induces cell cycle arrest in the late G1 phase or apoptosis after DNA damage. Inactivation of p53 gene can be caused by mutation, deletion, or gene rearrangement. P53 inactivation, with abolishment of its tumor suppressor activity, is the most common genetic event in human malignancies. Thus, partial loss of p53 tumor suppressor activity may play an important role in the development of low-grade MALT lymphomas, whereas complete loss (mutation of one allele and loss of the other) is associated with high-grade transformation [29]. In several cases, mutations of p53 gene are accompanied by overexpression of the p53 protein and in half of the cases by detection of serum anti-p53 antibodies [30]. Anti-p53 antibodies were detected at a high titer in the sera of two of 14 tested patients with SS with low-grade lymphoma [27]. Sequence analysis of the P53 gene in five patients with SS with low histologic grade lymphomas revealed two novel mutations of exon 5. These mutations are single-base substitutions and appear functional, because exon 5 is included in the coding region of the p53 gene. Mutant p53 proteins without tumor suppressor activity may lead to checkpoint failure at the level of the cell cycle, followed by uncontrolled cell proliferation. This finding indicates a probable role of this tumor suppressor gene as a possible mechanism for lymphoma development in SS [31••].

Chromosomal translocations into Ig heavy-chain gene segments have been described in several types of B

cell lymphomas, such as translocation (14;18) in follicular lymphomas. This translocation juxtaposes the Bcl-2 gene with Ig heavy-chain locus leading to deregulation of apoptosis and increased B cell survival, further contributing to an increased chance of lymphomatogenesis. These translocations probably reflect a mistaken V(D)J recombination, a molecular process that is believed restricted to B cell precursors in the bone marrow, but occasionally taking place in the germinal centers. It is possible that the microenvironment of ectopic germinal centers in the salivary glands of patients SS, where B cell lymphocytes undergo intense proliferation, may hamper variable-region gene recombination. Failures in the control of V(D)J recombination process appear to play an important role in neoplastic transformation [32•]. Pisa *et al.* [9] found the translocation (14;18) in five of seven SS-associated lymphomas. Prelymphoma biopsies from seven patients with SS who subsequently developed lymphoma lacked detectable translocation (14;18) even though they exhibited oligoclonal rearrangements of their Ig genes. Finally, an increased prevalence of trisomy 18 has been detected in six of 13 salivary gland low-grade MALTs and in all of four high-grade lymphomas in patients with SS [33]. Although translocations involving oncogenes or mutations of anti-oncogenes are detected in SS-associated lymphomas, recurrent molecular abnormalities, which could explain the pathogenesis of these lymphomas, have not been established.

The autoimmune lymphoproliferative syndrome, a disorder usually associated with germinal Fas mutations, illustrates the role of defective apoptosis in the genesis of lymphoma and autoimmunity. Fas cell surface receptor initiates programmed cell death or apoptosis of activated lymphocytes. The risk of NHL in individuals with inherited Fas mutations is 14 times greater than expected. The most obvious possibility is that a general expansion of the lymphoid pool provides a target cell population for other genetic or environmental transforming events. Furthermore, it has been shown that somatic disruption of Fas may play a role in pathogenesis of some NHL, such as MALT-type. A significant proportion of patients with identified Fas mutations showed extranodal disease at presentation and high incidence of well-documented autoimmune diseases (SLE, SS, or Hashimoto's thyroiditis) [34]. The high incidence of autoimmune phenomena observed among patients with NHL with Fas-mutated tumors suggests that somatic mutations of Fas form a mechanism connecting autoimmunity and lymphoma. In view of these results, a recent study using a polymerase chain reaction single-strand conformation polymorphism method investigated the presence of Fas and Fas ligand germline mutations in patients with SS without lymphoma and in patients with essential mixed cryoglobulinemia [35]. Despite the negative results of this study, the possibility that somatic mutations in the Fas/Fas ligand system may be specifically involved in SS-associated lymphoproliferation has not been excluded.

Hepatitis C virus–associated non-Hodgkin's lymphoma

Hepatitis C virus (HCV) is considered as the major etiologic factor of type II mixed cryoglobulinemia, a B cell clonal proliferative condition, which potentially evolves into frank NHL of the lymphoplasmacytoid or immunocytoma subtype. It could be suggested that the malignant transformation of the HCV-driven proliferating B cells is a consequence of the accumulation of stochastic genetic alterations or the direct effect of the virus [36•]. In this regard, a large proportion of HCV-associated immunocytomas derives from B cell clones chronically stimulated by a common antigen. This is supported on the basis of the peculiar properties that these NHL express intraclonal diversity—an R/S mutation ratio in the framework segments of Ig lower than expected by chance and a highly restricted use of gene segments in assembling IgH chain [37]. Furthermore, the significant homologies between gene segments used by these NHL and gene segments used by antibodies with RF activity suggest that these NHL derive from B cell clones that produce RF [38••]. Because these genes have been associated with the crossreactive idiotypes expressed by serum monoclonal components in type II cryoglobulinemia, it is likely that HCV-associated NHL are originated by neoplastic transformation of the same clones expanded in type II cryoglobulinemia. It has been postulated that HCV, as an exogenous antigen in complex with IgG, triggers distinct RF B cell clones [36•].

The preferential usage of a V1-69 VH in combination with the HumKv 325 VL gene, the use of specific segments, in particular the D21/9 segment in assembling the IgH chain of Ig, and the restricted length of CDR3 region support that HCV-associated neoplasms are closely related to salivary gland MALT lymphomas in terms of pathogenesis [38••,39••]. Furthermore, SS lymphomas and HCV-associated immunocytomas bear surface Ig with RF reactivity. A recent study suggests that salivary gland lymphoma in patients with SS frequently develops from RF-producing B cells [40••]. In this study, a model for increased frequency of lymphomas involving specific RF B cells in patients with SS may include the following: chronic stimulation by IgG, or more likely by IgG complexed to auto- or exoantigens. It has been proposed that the large amounts of IgG produced in the salivary glands of patients with SS could result in uncontrolled antigenic stimulation of RF B cell clones in the ectopic germinal centers where the vigorous expansion makes them susceptible to mutational events. As RF activity of lymphoma cells show a broad spectrum of crossreactivity, it would be important for researchers to determine the real antigen that these lymphomas recognize. Because HCV has not been associated with salivary gland MALT lymphoma, the VH gene similarities between SS and HCV lymphoproliferation actually represent their common selection by an antigen generated or upregulated during a process of a localized chronic immune stimulation.

Predictive Factors of Lymphoma Development

Knowing that patients with SS are at higher risk of developing lymphoma, several investigators have attempted to establish predictive factors for this progression. Although some clinical parameters may herald the imminent onset of lymphoma, few reliable markers are available to predict this progression.

In 1971, Anderson and Talal [41] showed that a decrease in the level of serum Ig and disappearance of RF occurred at the time of progression to lymphoma. Kassan *et al.* [1] showed that patients with lymphadenopathy, splenomegaly, parotid gland enlargement, and previous low-dose irradiation or chemotherapy had an increased risk of lymphoma development. In a study performed in the authors' department [42], the presence of mixed monoclonal cryoglobulins was the most significant risk factor in predicting the risk of lymphoma development. The crossreactive idiotypes 17109 and G6 were also correlated with the lymphoma development. The evidence of monoclonal paraproteinemia and urinary free light-chains may identify patients who have a particular risk of later lymphoma development [43]. In another study, lymphoproliferative disorders were associated with the presence of palpable purpura, low C4, and mixed monoclonal cryoglobulinemia. Patients without any of these factors were at negligible risk of developing lymphoma during the follow-up [44••]. Others have suggested that leg ulcers, which also may be manifestations of vasculitis, are predictive of lymphoma development [10]. In conclusion, these data suggest that patients with these risk factors constitute a separate subgroup that should be monitored and managed closer compared with other patients with SS.

The characterization of earlier stages of lymphoproliferation in SS is still ill defined. B cell clonal expansion is an early event in the course of SS, and these stages may range from clearly benign lymphoid infiltrates to early malignant. The risk of developing extrasalivary lymphoma is closely related to the presence of broad strands of monocytoid B cells between the myoepithelial sialadenitis (MESA) and the presence of monotypic Ig expression by lymphoid cells or plasma cells as detected by immunoperoxidase [45]. Compared with the detection of monoclonality by Ig light-chain expression, detection of B cell clones in salivary glands lesions by Southern blot or polymerase chain reaction analysis of Ig gene rearrangement has not proved a reliable predictor of clinical behavior in MESA [20,45]. Thus, it seems that molecular genetic analysis has little or no practical role in the clinical diagnosis of salivary glands lymphoma in a setting of MESA. The different types of B cell clonal expansion (oligoclonal or monoclonal, smaller or larger, fluctuating or established, localized or disseminated) may imply a different risk of lymphoma progression [21]. Taken all these factors into account, the focus on prelymphomatous stages is crucial to a better understanding of the whole lymphomatogenesis in SS.

Clinical Aspects

Pathology

The prevalence of NHL in patients with SS is 4.3%, and usually develops later in the illness. The median time from SS diagnosis to lymphoma diagnosis is 7.5 years [1,46••]. Various histologic subtypes of NHL for patients with SS have been described in the literature, including follicle center lymphoma, lymphoplasmacytoid, diffuse, large B cell lymphoma, and especially MALT lymphomas [1,27,46••]. However, the majority of lymphomas in SS are marginal-zone B cell lymphomas (MZL) [27,46••,47••]. This term encompasses MALT lymphoma and monocytoid B cell lymphomas, the latter being the nodal counterpart of the former. These tumors have sufficient morphologic, immunophenotypic, and clinical similarity to suggest that they are morphologic manifestations of the same neoplastic process [47••]. Cases that were classified in the past as immunocytomas probably belong to the MZL entity. Neoplastic MZLs are expected to retain the homing pattern of their normal precursors, which explain the diverse distribution of lymphoma of this type [27,48]. It has been postulated that this dissemination may be because of the specific expression of special homing receptor on the surface of the B cells of MALT that regulates the traffic of lymphocytes to the mucosal tissues by binding to mucosal addressin cell adhesion molecule-1 [49]. These lymphomas arise frequently in mucosal extranodal sites, as well as in extranodal nonmucosal sites; most of these sites have the presence of epithelium in common, usually columnar, suggesting that the property of these cells is homing to epithelia rather than to mucosa [48]. In the authors' study [46••], the MZL in patients with SS are primary low-grade and localized (stage I and II) with extranodal manifestations. The salivary glands are the most common site, but other extranodal sites are also involved, such as stomach, nasopharynx, skin, liver, kidney, and lung. Presenting symptoms are caused by major gland enlargement mainly bilateral parotid gland enlargement. Furthermore, the clinical picture in these patients is not characterized by the presence of B symptoms, whereas the performance status is particularly good. Lymph node involvement is common, but is rarely seen exclusively. In disseminated disease, usually more than one extranodal site is involved, whereas bone marrow infiltration is very rare. This suggests that salivary glands in patients with SS are the most common initial site of B cell neoplastic transformation, but other MALT sites, lymph nodes, and bone marrow are also identified as initial sites of transformation. Nonexocrine manifestations occurring more often than the general SS populations are skin vasculitis and peripheral nerve involvement. Among the hematologic and serologic parameters, anemia, lymphopenia, monoclonal Ig, and cryoglobulin are particularly frequent in these patients.

Lymphoma may remain localized for many years and may undergo spontaneous remissions in the absence of therapy [46••]. The observed regression is frequently

regional, and not all involved sites necessarily regress. Approximately 10% of patients with NHL have multiple histologic types of lymphoma (discordant lymphoma) identified in the biopsy tissues taken during the staging evaluation. The authors identified discordant lymphomas in patients with SS, and the prognosis is that of the high-grade component [46••].

Lymphomas in patients with SS tend to evolve toward a less-differentiated cell type in some cases. Most high-grade lymphomas in salivary glands are diffuse large B cell lymphomas. Rare cases of peripheral T cell, lymphoblastic, and Burkitt-like lymphomas have been reported. During transformation, the clinical picture is characterized by further nodal and extranodal dissemination [46••]. It is not known how many of the diffuse large B cell lymphomas arise from preexisting MALT lymphomas and how many are of nodal type or represent transformation of follicular lymphomas. Immunohistochemical, karyotypic, and genotypic studies have provided convincing proof that the supervening large-cell lymphomas arise from the same clone as the low-grade lymphomas. Thus, the majority of the high-grade lymphomas in patients with SS may represent blastic-variance of marginal-zone B cell or follicular center cell lymphomas.

Therapy-prognosis

Because the histologic grade is a very important prognostic factor for overall survival, the treatment of SS-associated NHL depends on the histologic grade of lymphoma [46••]. Low-grade MZL can be localized at diagnosis and curable with local therapy (involved-field radiotherapy or surgical removal). In the authors' study, the median overall survival of patients with low-grade lymphomas is 6.3 years, and this does not differ between treated and untreated patients [46••]. Thus, in patients with localized low-grade lymphoma affecting exocrine glands, a wait-and-watch policy should be undertaken, and if the lymphoma is disseminated, patients may be treated with single-agent chemotherapy. In another recent study, the authors identified the achievement of complete response in 75% of the patients with SS-associated B cell lymphoproliferation during 4 years of follow-up and improvement in some SS features after 2-chloro-2-deoxyadenosine therapy (oral symptoms, parotidomegaly, salivary flows, hyposthenuria, disappearance of cryo/urine monoclonal bands) [50]. In contrast, combined chemotherapy is recommended for patients with low-grade lymphoma transforming to high-grade lymphoma, and also for those with high-grade lymphoma. Over the past 15 years, a number of aggressive induction regimens have been evaluated in a pilot single institution study that included patients with high-grade lymphomas. However, when these regimens were subsequently compared with cyclophosphamide, doxorubicin, vincristine, and prednisone (CHOP) in large randomized trials, patients treated with CHOP had comparable complete remission rates and overall survival. Consequently, the majority of patients with aggressive NHL in SS receive an anthracycline-containing regimen, such as CHOP. Unfortunately, the median survival is estimated to be only 1.8 years in these patients. The presence of B symptoms and a large tumor diameter (greater than 7 cm) are additional independent death risk factors [46••]. Researchers are unable to identify clinical features at the time of the initial presentation that possibly predispose histologic progression, and there is no data that address the efficacy of treatment in presenting high-grade transformation.

Conclusions

Patients with SS with high-risk factors, such as palpable purpura, low C4, and mixed monoclonal cryoglobulinemia, constitute a separate subgroup that should be monitored more closely than other patients with SS. A strong predisposition seems to exist in these patients for the development of lymphoproliferation especially for low-grade salivary gland MZL. It has been proposed that the large amounts of IgG produced in the salivary glands of patients with SS could result in uncontrolled antigenic stimulation of RF B cell clones in the ectopic germinal centers, where the vigorous expansion makes them susceptible to mutational events. It would be important to determine the real antigen that these lymphomas recognize for a better understanding of the whole lymphomatogenesis in SS.

References and Recommended Reading

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

- • Of importance
- •• Of major importance

1. Kassan SS, Thomas TL, Moutsopoulos HM, *et al.*: **Increased risk of lymphoma in sicca syndrome.** *Ann Inter Med* 1978, **89:**888–892.
2. Ansell SM, Grant CS, Habermann TM: **Primary thyroid lymphoma.** *Semin Oncol* 1999, **26:**316–323.
3. Sokol RJ, Hewitt S, Stamps BK: **Autoimmune hemolysis: an 18-year study of 865 cases referred to a regional transfusion center.** *Br Med J* 1981, **282:**2023–2027.
4. Abu-Shakra M, Gladman DD, Urowitz MB: **Malignancy in systemic lupus erythematosus.** *Arthritis Rheum* 1996, **39:**1050–1054.
5. Mellemkjaer L, Linet MS, Gridley G, *et al.*: **Rheumatoid arthritis and cancer risk.** *Eur J Cancer* 1996, **32A:**1753–1757.
6. Valesini G, Priori R, Bavoillot D, *et al.*: **Differential risk of non-Hodgkin's lymphoma in Italian patients with primary Sjögren's syndrome.** *J Rheumatol* 1997, **24:**2376–2380.
7. Youinou P, Mackenzie L, le Masson G, *et al.*: **CD5-expressing B lymphocytes in the blood and salivary glands of patients with primary Sjogren's syndrome.** *J Autoimmun* 1988, **1:**185-194.
8. Bunim JJ, Talal N: **Development of malignant lymphoma in the course of Sjögren's syndrome.** *Trans Assoc Am Physicians* 1963, **76:**45–56.
9. Pisa EK, Pisa P, Kang HI, Fox RI : **High frequency of t(14;18) translocation in salivary gland lymphomas from Sjögren's syndrome patients.** *J Exp Med* 1991, **174:**1245–1250.
10. Sutcliffe N, Inanc M, Speight P, Isenberg D: **Predictors of lymphoma development of primary Sjögren's syndrome.** *Semin Arthritis Rheum* 1998, **28:**80–87.

11. DiGiuseppe JA, Corio RL, Westa WH: **Lymphoid infiltrates of the salivary glands: pathology, biology and clinical significance.** *Curr Opin Oncol* 1996, **8:**232–237.
12. Stott DI, Hiepe F, Hummel M, *et al.*: **Antigen-driven clonal proliferation of B cells within target tissue of an autoimmune disease: the salivary glands of patients with Sjögren's syndrome.** *J Clin Invest* 1998, **102:**938–946.
13. Fox RI, Bumol T, Fantozzi R, *et al.*: **Expression of histocompatibility antigen HLA-DR by salivary gland epithelial cells in Sjögren's syndrome.** *Arthritis Rheum* 1986, **29:**1105–1111.
14. Xanthou G, Policronis M, Tzioufas AG, *et al.*: **"Lymphoid" chemokine messenger RNA expression by epithelial cells in the chronic inflammatory lesion of the salivary glands of Sjögren's syndrome patients: possible participation in lymphoid structure formation.** *Arthritis Rheum* 2001, **44:**408–418.
15. Dimitriou ID, Kapsogeorgou EK, Moutsopoulos HM, Manoussakis MN: **CD40 on salivary gland epithelial cells: high constitutive expression by cultured cells from Sjogren's syndrome patients indicating their intrinsic activation.** *Clin Exp Immunol* 2002, **127:**386–392.
16. Adamson TC, Fox RI, Frisman DM, Howell FV: **Immunohistologic analysis of lymphoid infiltrates in primary Sjogren's syndrome using monoclonal antibodies.** *J Immunol* 1983, **130:**203–208.
17. Fox RI, Kang HI, Ando D, *et al.*: **Cytokine mRNA expression in salivary gland biopsies of Sjögren's syndrome.** *J Immunol* 1994, **152:**5532–5539.
18. Boumba D, Skopouli FN, Moutsopoulos HM: **Cytokine mRNA expression in the labial salivary gland tissues from patients with primary Sjogren's syndrome.** *Br J Rheumatol* 1995, **34:**326–333.
19. Freimark B, Fantozzi R, Bone R, *et al.*: **Detection of clonally expanded salivary gland lymphocytes in Sjögren's syndrome.** *Arthritis Rheum* 1989, **32:**859–869.
20. Bahler DW, Swerdlow SH: **Clonal salivary gland infiltrates associated with myoepithelial sialadenitis (Sjögren's syndrome) begin as nonmalignant antigen-selected expansions.** *Blood* 1998, **91:**1864–1872.
21. De Vita S, Boiocchi M, Sorrentino D, *et al.*: **Characterization of prelymphomatous stages of B cell lymphoproliferation in Sjögren's syndrome.** *Arthritis Rheum* 1997, **40:**318–331.
22. Anaya JM, McGuff HS, Banks PM, Talal N: **Clinicopathological factors relating malignant lymphoma with Sjogren's syndrome.** *Semin Arthritis Rheum* 1996, **25:**337–346.
23. Fong S, Chen PP, Gilbertson TA, *et al.*: **Expression of three cross-reactive idiotypes on rheumatoid factor autoantibodies from patients with autoimmune diseases and seropositive adults.** *J Immunol* 1986, **137:**122–128.
24. Fox RI, Chen P, Carson DA, Fong S: **Expression of a cross-reactive idiotypes on rheumatoid factor in patients with Sjögren's syndrome.** *J Immunol* 1986, **136:**477–483.
25. Katsikis PD, Youinou PY, Galanopoulou V, *et al.*: **Monoclonal process in primary Sjögren's syndrome and cross-reactive idiotype associated with rheumatoid factor.** *Clin Exp Immunol* 1990, **82:**509–514.
26. Bahler DW, Miklos JA, Swerdlow SH: **Ongoing Ig gene hypermutation in salivary gland mucossa-associated lymphoid tissue-type lymphomas.** *Blood* 1997, **89:**3335–3344.
27. Royer B, Cazals-Hatem D, Sibilia J, *et al.*: **Lymphomas in patients with Sjögren's syndrome are marginal zone B-cell neoplasms, arise in diverse extranodal and nodal sites and are not associated with viruses.** *Blood* 1997, **90:**766–775.
28. Peng H, Chen G, Du M, *et al.*: **Replication error phenotype and p53 gene mutation in lymphomas of mucosa-associated lymphoid tissue.** *Am J Pathol* 1996, **148:**643–648.
29. Du M, Peng H, Singh N, Isaacson PG, Pan L: **The accumulation of p53 abnormalities is associated with progression of mucosa-associated lymphoid tissue lymphoma.** *Blood* 1995, **86:**4587–4593.
30. Soussi T: **The humoral response to the tumor-suppressor gene-product p53 in human cancer: implications for diagnosis and therapy.** *Immunol Today* 1996, **17:**354–356.
31.•• Tapinos NI, Polihronis M, Moutsopoulos HM : **Lymphoma development in Sjögren's syndrome: novel p53 mutations.** *Arthritis Rheum* 1999, **42:**1466–1472.

Described two novel mutations in exon 5 of the p53 gene in four patients with SS-associated low-grade NHLs. The authors postulate that inactivation of p53 tumor-suppressor activity is a potential mechanism of SS lymphomatogenesis.

32.• Kuppers R, Klein U, Hansmann ML, Rajewsky K: **Cellular origin of human B-cell lymphomas.** *N Engl J Med* 1999, **341:**1520–1529.

This is an excellent review about the origin of B cell NHLs. The authors review the literature and focus on variable IgH genes recombinational events proposing mechanisms for B cell clonal expansion after antigen-driven stimulation.

33. Ihrler S, Baretton GB, Menauer F, *et al.*: **Sjögren's syndrome and MALT lymphomas of salivary glands: a DNA-cytometric and interphase-cytogenetic study.** *Mod Pathol* 2000, **13:**4–12.
34. Gronbaek K, Straten PT, Palfkiaer E, *et al.*: **Somatic Fas mutations in non-Hodgkin's lymphoma: association with extranodal disease and autoimmunity.** *Blood* 1998, **92:**3018–3024.
35. Bertolo F, De Vita S, Dolcetti R, *et al.*: **Lack of Fas and Fas-L mutations in patients with lymphoproliferative disorders associated with Sjögren's syndrome and type II mixed cryoglobulinemia.** *Clin Exp Rheumatol* 1999, **17:**339–342.
36.• Dammacco F, Sansonno D, Piccoli C, *et al.*: **The lymphoid system in hepatitis C virus infection: autoimmunity, mixed cryoglobulinemia, and overt B-cell malignancy.** *Semin Liver Dis* 2000, **20:**143–157.

An interesting review focusing on the pathogenetic mechanisms involved in B cell expansion and progression to frank lymphoid malignancy in patients with HCV.

37. Ivanovski M, Silvestri F, Pozzato G, *et al.*: **Somatic hypermutation, clonal diversity, and preferential expression of the VH 51p1/VL kv325 immunoglobulin gene combination in hepatitis C virus-associated immunocytomas.** *Blood* 1998, **91:**2433–442.
38.•• De Re V, De Vita S, Marzotto A, *et al.*: **Sequence analysis of the immunoglobulin antigen receptor of hepatitis C virus-associated non-Hodgkin lymphomas suggests that the malignant cells are derived from the rheumatoid factor-producing cells that occur mainly in type II cryoglobulinemia.** *Blood* 2000, **96:**3578–3584.

Describes that a large proportion of HCV-associated NHLs derives from RF-producing B cell clones chronically stimulated by a viral antigen, such as E2 protein.

39.•• Miklos JA, Swerdlow SH, Bahler DW: **Salivary gland mucosa-associated lymphoid tissue lymphoma immunoglobulin V(H) genes show frequent use of V1-69 with distinctive CDR3 features.** *Blood* 2000, **95:**3878-3884.

The article indicates that salivary gland MALT lymphomas represent a highly selected B cell population that may recognize the same epitopes.

40.•• Martin T, Weber JC, Levallois H, *et al.*: **Salivary gland lymphomas in patients with Sjögren's syndrome may frequently develop from rheumatoid factors B cells.** *Arthritis Rheum* 2000, **43:**908–916.

An excellent molecular study attempting to prove the hypothesis that SS-associated B cell clones have RF activity. This article illustrates the link among primary SS, autoantibody-producing cells, and lymphoma.

41. Anderson LG, Talal N: **The spectrum of benign to malignant lymphoproliferation in Sjögren's syndrome.** *Clin Exp Immunol* 1972, **10:**199–221.
42. Tzioufas AG, Boumba DS, Skopouli FN, Moutsopoulos HM: **Mixed monoclonal cryoglobulinemia and monoclonal rheumatoid factor cross-reactive idiotypes as predictive factors for the development of lymphoma in primary Sjögren's syndrome.** *Arthritis Rheum* 1996, **39:**767–772.
43. Walters MT, Stevenson FK, Herbert A, *et al.*: **Urinary monoclonal free light chains in the primary Sjögren's syndrome: an aid to the diagnosis of malignant lymphoma.** *Ann Rheum Dis* 1986, **45:**210–219.

44.•• Skopouli FN, Dafni U, Ioannidis JP, Moutsopoulos HM: **Clinical evolution, and morbidity and mortality of primary Sjögren's syndrome.** *Semin Arthritis Rheum* 2000, **29:**296–304.
This is an interesting retrospective study of the clinical course and outcome of 261 patients with primary SS. The article provides predictors for lymphoma development in a large cohort of patients and clearly delineates the patients with SS in low- and high-risk groups.

45. Harris NL: **Lymphoid proliferations of the salivary glands.** *Am J Clin Pathol* 1999, **111(suppl):**S94–S103.

46.•• Voulgarelis M, Dafni UG, Isenberg DA, Moutsopoulos HM: **Malignant lymphoma in primary Sjögren's syndrome: a multicenter, retrospective, clinical study by the European Concerted Action on Sjögren's Syndrome.** *Arthritis Rheum* 1999, **42:**1765–1772.
A large European multicenter retrospective study that provides clearly the clinical and pathologic characteristics, the treatment outcome and prognosis of SS-associated NHLs.

47.•• Mariette X: **Lymphomas in patients with Sjögren's syndrome: review of the literature and physiopathologic hypothesis.** *Leuk Lymphoma* 1999, **33:**93–99.
An overall review of SS-associated lymphomas focusing on the clinical parameters, but also highlighting important pathologic and pathogenetic aspects.

48. Pelstring RJ, Essell JH, Kurtin PJ, *et al.*: **Diversity of organ site involvement among malignant lymphomas of mucosa-associated tissues.** *Am J Clin Pathol* 1991, **96:**738–745.

49. Dogan A, Du M, Koulis A, *et al.*: **Expression of lymphocyte homing receptors and vascular addressing in low-grade gastric B-cell lymphomas of mucosa-associated lymphoid tissue.** *Am J Pathol* 1997, **151:**1361–1369.

50. Voulgarelis M, Petroutsos G, Moutsopoulos HM, Skopouli FN: **2-chloro-2-deoxyadenosine in the treatment of Sjögren's syndrome-associated B cell lymphoproliferation.** *Arthritis Rheum* 2002, **46:**2248–2249.