Genomic profiles of MALT lymphomas: variability across anatomical sites

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ABSTRACT

MALT lymphomas present common features, but important differences are associated with involvement of specific anatomical sites, many likely contributing to the biology. To test the existence of genetic alterations specific for primary anatomical sites of involvement, genomic profiles obtained with high-density arrays were analyzed in 130 MALT lymphomas across a spectrum of anatomic sites.

Trisomies 3 and 18 and del(6q23) occurred at a similar frequency. Instead, gains at 6p appeared significantly more common among MALT lymphomas involving the orbital adnexa. Gastric involvement showed a trend for a higher frequency of 8q gains.

In conclusion, MALT lymphomas appear to bear a common set of recurrent unbalanced genomic alterations independent of the anatomical site. This differs from what has been observed for common chromosome translocations. Only a few alterations such as gains at 6p and, possibly, gains at 8q show preferential involvement at specific anatomical sites.

Key words: arrayCGH, lymphoma, *MALT1, TNFAIP3,* orbital adnexa, marginal zone lymphoma, 3q, 18q.

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Introduction

Extranodal marginal zone B-cell lymphomas (MZL) of mucosa-associated lymphoid tissue (MALT lymphoma) represent the most common MZL type and have been described in a variety of different anatomical sites.[1,2] As a clinico-pathological entity, MALT lymphomas share common histological, clinical and genetic features, but important differences have been reported for the different involved anatomical sites.[1-4] This is suggested by the fact that the occurrence of four recurrent and mutually exclusive chromosome translocations [t(11;18)(q21;q21), t(1;14)(p22;q32), t(14;18)(q32;q21), and t(3;14)(p14.1;q32)] can be detected in these lymphomas and their occurrence varies according to the anatomical sites.[3] Also, important differences with regard to the associations with autoimmune disorders or infectious agents appear to differ from organ to organ as well. These site-specific biological differences might impact outcome and therapeutic approaches, besides playing a role in the lymphomagenesis.[2]

Besides chromosome translocations, unbalanced DNA changes including gains on chromosomes 3 and 18 (usually trisomies), deletions affecting the nuclear factor-κB (NF-κB) inhibitor *TNFAIP3/A20* (6q23) and gains of the short arm of chromosome 6 are recurrent lesions in MALT lymphomas, and at least the latter two occur at a higher frequency compared to splenic MZL.[5-9] To the best of our knowledge, only a few studies have compared the occurrence of unbalanced DNA changes among MALT lymphomas from different organs, often analyzing only one or two anatomical sites at a time.[6,10,11] To test the existence of copy number alterations specific for anatomical sites of involvement, genomic profiles of 130 MALT lymphomas were analyzed.

Design and Methods

All cases were analyzed using the Affymetrix Human Mapping 250k Nsp array (Affymetrix, Santa Clara, CA, USA) directly by the authors in 57 cases (Gene Expression Omnibus dataset GSE24881)(9)

The online version of this article has a Supplementary Appendix.

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or drawn from publicly available raw CEL files in an additional 73 cases [Gene Expression Omnibus dataset GSE12906[12]]. Data mining was performed as previously described[9] but, due to the different genetic populations of the two series of samples, the appropriate Hapmap normal samples were used for copy number (CN) and loss of heterozygosity normalization. Mapping data for probes were derived from the National Center for Biotechnology Information (NCBI) Human Genome Build 36, as provided by Affymetrix, which was used for all subsequent analyses. Minimal common regions (MCR) were defined according to Lenz *et al.*[13] MCRs were compared with the Database of Genomic Variants (*http://projects.tcag.ca/variation/*): regions showing more than 80% overlapping between probes; known copy number variations (CNV) were considered bona fide CNV and discarded from further analyses. MCR containing the genes coding the immunoglobulin heavy and light chain genes were also similarly discarded since CN changes in these regions likely represent the physiological rearrangements occurring in B cells. Associations in two-way tables were tested for statistical significance using either the $\chi 2$ test or Fisher's exact test (two-tailed), as appropriate. All tests were two-sided, and P value ≤ 0.05 was considered significant. Statistical analysis was conducted using the STATA 11.0 software package (Stata Corporation, College Station, TX, USA). The study was approved by the Bellinzona ethical committee.

Results and Discussion

The main anatomical sites of the MALT lymphomas included in this analysis comprised ocular adnexa (54 of 130, 41%), stomach (28 of 130, 21%), parotid or other salivary glands (12 of 130, 9%), thyroid (9 of 130, 6%), lung (8 of 130, 6%) and skin (5 of 130, 4%). Due to the need for diagnostic frozen tumor biopsies, the frequency distribution of cases did not fully reflect the actual pattern of anatomical site involvement of MALT lymphomas.[2] Figure 1 represents the lesions observed in the entire series of patients. As a whole, the different MALT lymphomas presented similar genomic aberrations (Figure 2 and *Online Supplementary Table S1*). Trisomy 3 and 18 and losses at 6q23(135,101,251-139,048,099) occurred at similar frequencies in MALT lymphomas primarily involving stomach, orbital adnexa, thyroid, salivary glands and lung. The first two alterations are shared by all MZL, although they are more common in MALT lymphomas than in splenic marginal zone B-cell lymphoma (MZL),[9] as well as by diffuse large B-cell lymphomas (DLBCL).[12,14] The *MALT1* gene, altered by chromosomal translocations in MALT lymphomas, maps to 18q but the usual detection of gains affecting the whole chromosomes 3 and 18 suggest the deregulation of multiple genes. Indeed, we recently reported that these lesions affect genes involved in the B-cell receptor signaling pathway, Wnt signaling, cell cycle and apoptosis regulation, chemokine and cytokine signaling pathway and ubiquitin proteasome pathway.[9] Losses at 6q23 lead to inactivation of the nuclear factor-κB (NF-κB) inhibitor *TNFAIP3*, a recently described pathogenetic event occurring in MALT lymphomas and in DLBCL, less com-

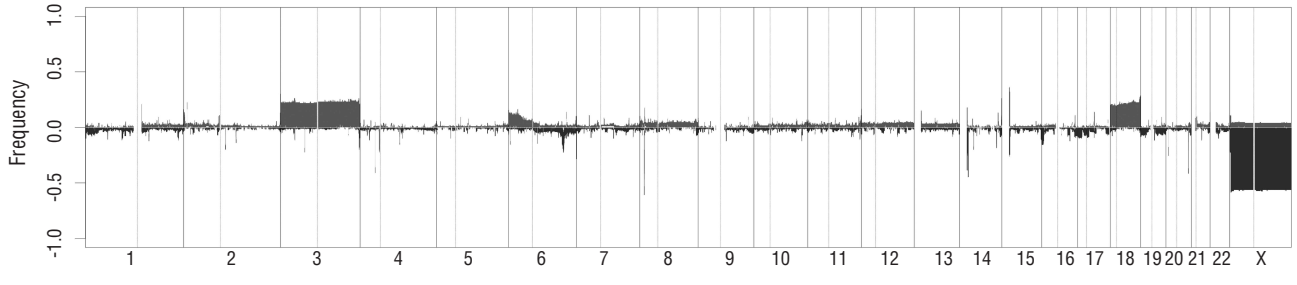

Figure 1. Frequency plots of DNA gains (up) and losses (down) observed in 130 MALT lymphomas. X axis: chromosome localization and physical mapping. Y axis: percentage of cases bearing the aberration.

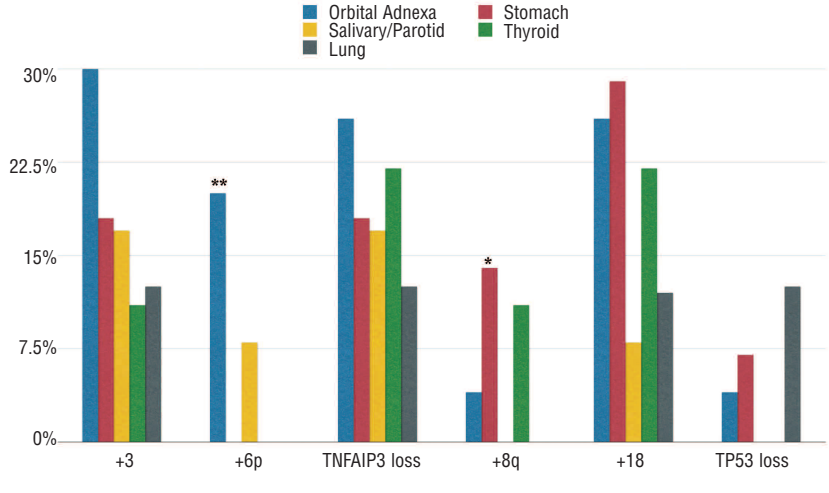

Figure 2. Distribution of the most common DNA gains and losses across the main anatomical sites investigated. Y axis: percentage of cases bearing the lesion; $^{**}P<0.001$; $^{*}P=0.05$.

monly observed in splenic MZL.[5,9,12,14] In contrast to chromosomal translocations, which display important site-specific differences,[3] trisomies 3 and 18 as well as TNFAIP3 inactivation appear relevant for the pathogenesis of all MALT lymphomas. Similarly, no statistical differences were demonstrated for loss of the *TP53* locus which is much less common in MALT lymphomas than in splenic MZL.[9]

In contrast, gains of genomic material at 6p appeared significantly more common among MALT lymphomas of the orbital adnexa than those occurring at other sites [the whole arm, 20% *vs.* 0%; *P*<0.001; 6p25.3p21.32(0-32,904,801) region, 20% *vs.* 4%, *P*=0.009]. Interestingly, MALT lymphomas of the orbital adnexa have a low frequency of the most common chromosomal translocations *BIRC3/MALT1* or *IGHV/MALT1* suggesting that transcripts mapped at 6p could affect similar pathways. Gastric involvement showed a trend for a higher frequency of gains at 8q11q24(50,310,000-126,643,685) (14% *vs.* 3%, *P*=0.05), comprising MYC.

Both gains of 6p and 6q23 losses (*TNFAIP3*) were more commonly associated with gains of 3q, although they were observed also as single unbalanced lesions: 6p gains occurred in 7 of 99 (7%) cases without 3q gains and in 8 of 31 (26%) with 3q gains (*P*=0.004). Losses at 6q23 occurred in 17 of 99 (17%) cases without 3q gains and in 11 of 31 (35%) with 3q gains (*P*=0.03). Also, as previously shown in a smaller series of MALT lymphomas[9] and different from other reports,[10] 6p gains and 6q23 losses tended not to occur in the same cases. These two lesions were concomitantly observed in only 4 cases, while 24 of 28 (86%) had 6q23 losses without gains of 6p and 11 of 15 (73%) had gains of 6p without 6q23 losses (*P*=0.607).

In conclusion, MALT lymphomas appear to bear a common set of unbalanced genomic copy number alterations independent of the anatomical site of presentation. This differs to want has been observed for chromosome translocations. Gains at 6p and, possibly, at 8q show a preferential occurrence in MALT lymphomas of the orbital adnexa and of the stomach, respectively, two diseases strongly associated with chronic infectious conditions. These abnormalities deserve to be further characterized in large studies addressing anatomical site-related alterations in MALT lymphomas and their relationship with potentially linked infectious agents.

Authorship and Disclosures

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