Primary Low-Grade B-Cell Lymphoma of Mucosa-Associated Lymphoid Tissue Type Arising in the Urinary Bladder

Report of 4 Cases With Molecular Genetic Analysis

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Context.—Primary lymphoma of the urinary bladder is rare. Only 84 cases have been reported in the English literature to date, and none of these cases has had molecular confirmation of clonal immunoglobulin gene rearrangement.

Objectives.—To review all cases with primary urinary bladder lymphoma in our records, to classify them using the REAL classification, to confirm their immunophenotype and genotype, and to determine their outcome.

Design.—We identified 4 cases of primary urinary bladder lymphoma in our medical records from a 30-year period. Immunohistochemical detection of immunoglobulin light chains and molecular analysis of immunoglobulin heavy-chain genes using the polymerase chain reaction were performed on paraffin-embedded material.

Results.—All patients were older than 60 years. The male-female ratio was 1:3. All patients had a history of chronic cystitis. Histologic features of mucosa-associated lymphoid tissue lymphoma with centrocyte-like cells, plasmacytoid B cells, or both were observed in all cases. Monoclonality of B cells was demonstrated by immunohistochemistry, polymerase chain reaction, or both methods in every case. All patients presented with stage IAE disease, were treated with radiotherapy alone, and have been in continuous complete remission for 2 to 13 years.

Conclusions.—Primary bladder lymphomas are usually of low-grade mucosa-associated lymphoid tissue type. They are more common in females and are associated with a history of chronic cystitis. Lymphoepithelial lesions are seen only in association with areas of cystitis glandularis. B-cell clonality is readily demonstrable by immunohistochemistry and/or polymerase chain reaction analysis. Local radiotherapy appears to confer long-term control.

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Primary malignant lymphoma of bladder is a rare disease, accounting for only 0.2% of all cases of extranodal lymphoma in North America. The prognosis of primary bladder lymphoma has been favorable, with many patients alive and well several years after treatment. A large proportion of primary lymphomas of the bladder are lymphomas of mucosa-associated lymphoid tissue (MALT). Mucosa-associated lymphoid tissue lymphomas were first described by Isaacson and Wright in the gastrointestinal tract. Both low- and high-grade B-cell lymphomas arising in MALT have been recognized, the majority of these being low grade. Recently, this concept has been extended as cases have been described in other sites, including lung, trachea, thymus, skin, meninges, gall bladder, salivary gland, thyroid, conjunctiva, breast, sinonasal cavities, and urinary bladder.

We report 4 additional cases of MALT lymphomas of the urinary bladder seen at the Princess Margaret Hospital, Toronto, Ontario, during the last 30 years.

MATERIALS AND METHODS

We searched medical records at the Princess Margaret Hospital for cases of primary bladder lymphoma. Four cases were found from a period of 30 years (1969–1999). The clinical histories were reviewed and are summarized in Table 1. Blocks were recut and sections stained with hematoxylin-eosin. Immunohistochemical studies were performed on paraffin sections using the avidin-biotin peroxidase complex, and the panel of antibodies included those against CD45, CD45RO, CD20, CD43, k biotin peroxidase complex, and the panel of antibodies included those against CD45, CD45RO, CD20, CD43, k and l light chains (Dako Corporation, Carpinteria, Calif), CD5, CD10 (Novocastra Laboratories, Ltd, Newcastle Upon Tyne, United Kingdom). B-cell monoclonality was assessed using polymerase chain reaction (PCR), which was performed on DNA extracted from paraffin-embedded tissue, using methods described previously. For PCR analysis, 500 ng to 1 μg DNA was amplified using the appropriate primer, electrophoresed through a 2% agarose gel, and visualized using ethidium bromide. Appropriate positive controls, negative controls, and internal controls were run with each specimen. Polymerase chain reaction analysis was done using primers specific for the framework 3 (FR3A) and J consensus regions of the immunoglobulin heavy-chain gene (JH), essentially as described by Trainor et al. A second PCR using consensus primers specific for framework 256 regions and J consensus regions was...
also used to increase our detection rate. Internal control primers were multiplexed with each reaction to ensure the presence of amplifiable DNA, as described previously.28

REPORT OF CASES

Case 1
A 64-year-old woman presented in 1985 with episodic hematuria and urinary frequency, and was diagnosed to have recurrent urinary tract infection. Multiple urine cultures revealed staphylococci, streptococci, Escherichia coli, and diptheroid bacillus species. She had been treated with repeated courses of antibiotics, her symptoms persisted and she subsequently developed gross hematuria. At cystoscopy, a 4-cm pedunculated tumor was seen in the bladder. Antibiotics during a 3-year period. Cystoscopy revealed extensive patches of uneven mucosa involving most of her bladder, as well as a small area of ulceration. A diagnosis of lymphocytic lymphoma was reported on the basis of a cold cup biopsy. The tumor involved part of the left ureter. Intravenous pyelography revealed bilateral hydronephrosis. Chest radiography, abdominal computed tomography, and lymphangiogram were normal. Bone marrow biopsy and aspirate revealed no involvement, and the tumor was staged as IAE disease. She received radiation therapy to the bladder and pelvis (35 Gy in 20 fractions) and was followed with cystoscopy every 6 months. She was last seen in May 1999 and had no evidence of recurrence.

Case 2
In 1993, a 69-year-old woman presented to her family physician with urinary frequency and urgency, and was diagnosed to have urinary tract infection. Multiple urine cultures showed E coli infection. No lymphadenopathy was detected. Chest, cardiac, and abdominal examinations were unremarkable. She was placed on antibiotics. The patient’s symptoms did not improve, and at cystoscopy 2 mucosal nodules were observed. Computed tomographic scan of the abdomen showed right hydronephrosis with a dilated right ureter and some thickening of the bladder wall consistent with a bladder tumor. Retrograde pyelogram could not be performed because of obstruction of the distal ureter. A bladder biopsy obtained during cystoscopy was reported as low-grade MALT lymphoma. A bone marrow biopsy showed no involvement by lymphoma.

The tumor was staged as IAE, and the patient received radiation therapy to the bladder and pelvis (35 Gy in 20 fractions). Treatment was completed in September 1993. She has been followed regularly every 3 months since and was last seen in October 1998 with no evidence of recurrence.

Case 3
In October 1996, a 72-year-old woman was referred for consideration and management of recurrent urinary tract infection. Multiple urine cultures revealed E coli infection. Two and a half years previously, she developed nocturia 4 or 5 times nightly, and microscopic hematuria was detected. Despite multiple courses of antibiotics, her symptoms persisted and she subsequently developed gross hematuria. At cystoscopy, a biopsy revealed low-grade malignant lymphoma of MALT type. On examination, no lymphadenopathy was detected, and the chest and abdomen were unremarkable. The patient’s tumor was staged with computed tomographic scan of the abdomen and pelvis, bone scan, and chest radiography. Gallium scan suggested some focal areas of increased uptake that were possibly compatible with adenopathy; however, computed tomographic scan of the abdomen did not reveal adenopathy. The tumor was staged as IAE. The patient was treated with radiotherapy (35 Gy in 20 fractions) to the bladder and pelvis. She was monitored with cystoscopy every 6 months. She was last seen in March 1999 and had no evidence of recurrence.

Case 4
A 62-year-old man presented in July 1997 with hematuria and urgency. Urine culture revealed Staphylococcus aureus infection. At cystoscopy, a 4-cm pedunculated tumor was seen in the bladder.

Biopsy revealed low-grade, B-cell malignant lymphoma of MALT type. Bone marrow biopsy was negative. Computed tomographic scan of the thorax, abdomen, and pelvis were negative except for the presence of the mass on the left posterolateral wall of the bladder. The tumor was staged as IAE, and the patient was treated with radiotherapy to the bladder and pelvis (35 Gy in 20 fractions). He was last seen in August 1999 with no evidence of recurrence.

PATHOLOGIC FINDINGS

Review of the biopsies from these 4 cases showed the classic histologic pattern of a low-grade MALT lymphoma, including the first case, which had been initially diagnosed as lymphocytic lymphoma. Monomorphic infiltrates of centrocyte-like cells, monocyctoid cells, and/or small lymphocytes with plasmacytoid differentiation were noted (Figure 1). Focal lymphoepithelial lesions were seen in the biopsies of cases 2 and 4, and in both cases the involvement was restricted to areas of cystitis glandularis (Figure 2). Reactive germinal centers were also seen in cases 1 and 2. No evidence of transformation was seen in any of the specimens. No muscularis propria was seen in the biopsies, except in specimen 4, which showed infiltration by lymphoma. Plasmacytoid differentiation was seen in all of these cases, but was more marked in case 4 with Russell body formation.

Peripheral blood cell count at presentation was unremarkable in patients 1, 3, and 4. Patient 2 showed leukocytosis with an increased neutrophil count. No absolute lymphotoysis was detected in any of the 4 patients.

Immunophenotyping
Light-chain restriction was demonstrated in 3 cases (cases 2, 3, and 4). Results are summarized in Table 2. Flow cytometric data were available for case 4 and showed typical marginal zone B-cell immunophenotype (positive for CD45 and CD20, negative for CD5, CD23, and CD10) with κ light-chain restriction and an S phase of 1%, which is consistent with a low-grade lymphoma.

PCR for Immunoglobulin Heavy-Chain Gene
Polymerase chain reaction analyses (Figure 3) revealed clonal immunoglobulin heavy-chain (IgH) gene rearrangement in 3 cases (cases 1, 2, and 4); PCR was not informative in case 3. Immunohistochemistry, however, showed κ light-chain restriction as well as α heavy-chain restriction in this case.
Figure 1. Case 2. Mucosa-associated lymphoid tissue lymphoma involving the lamina propria of the urinary bladder (hematoxylin-eosin, original magnification x100).

Figure 2. Case 2. Focal lymphoepithelial lesions in area of cystitis glandularis (hematoxylin-eosin, original magnification x400).

Figure 3. Case 4. B-cell-specific polymerase chain reaction using primers directed at the framework 256 (FR256) regions of the immunoglobulin heavy-chain gene (IgH). The top arrow represents the internal control that is used to ensure the presence of amplifiable DNA in each sample. The bracket in the FR256 figure denotes the size range in which IgH gene products can be seen. Although the DNA is degraded and the signal is weak, patient B (case 4) clearly shows the presence of a clonally rearranged IgH gene using the FR256 primers. Clonal rearrangements of IgH genes were also noted in cases 1 and 2 (not shown in figure). Lanes A and C are from cases unrelated to this article.

Table 2. Immunohistochemical and Polymerase Chain Reaction (PCR) Findings*

<table>
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<th>Case No.</th>
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</table>

* Plus sign indicates positive reaction; I, inconclusive; minus sign, negative reaction; ND, not done; F+, focally positive; LLCR, λ-light chain restriction; and KLCR, κ-light chain restriction.

COMMENT

The incidence of secondary involvement of the urinary bladder in lymphoma is about 13%,29 whereas primary malignant lymphoma of the urinary bladder is an uncommon neoplasm.30-44 It accounts for 0.2% of all cases of extranodal lymphoma in North America.1 In a literature search, Simpson et al identified 66 cases of primary bladder lymphoma and concluded that such cases generally have a good prognosis. The same conclusion of favorable prognosis was reported by Ohsawa et al,46 who also noted that the age of patients at diagnosis ranged from 20 to 85 years (median age 64 years), with a striking female predominance (male-female ratio, 1:1.8:3). Where race was stated, most of the patients were described as white or Caucasian.45 The most common presenting symptoms were hematuria followed by dysuria or nocturia.46 The vast majority of the cases were non-Hodgkin lymphomas of B-cell type, and among them 14% were called follicular lymphoma. Siegelbaum et al noted that the majority of these tumors were low grade and had a good prognosis.

Simpson et al were among the first to raise the possibility of a lower urinary tract lymphoma being of MALT type. Although many case reports describe histologic features that are typically seen in MALT lymphoma, the first case report of MALT lymphoma of the urinary bladder was described by Kuhara et al.20 Since that case was described, there have been at least 17 cases of bladder MALT lymphoma reported in the literature.2,19,21-24 All except 2 of the 17 cases were of low grade.22 Generally the prognosis was very good. Pawade et al described 5 cases of MALT lymphoma that arose in the urinary bladder. Four were in women, and one in a man. One case remained untreated for 8 years. These authors found lymphoepithelial lesions in 1 case, which was the only case showing cystitis glan-
dularis. In a review of the Mayo Clinic records for the last 56 years, Kempton et al\textsuperscript{21} reported 6 cases of primary bladder lymphoma, all of which were considered of MALT type, stage IA, and all of which received radiotherapy with or without surgery. No patient had recurrent lymphoma or died of lymphoma.

Lymphoepithelial lesions have been described only in the areas of cystitis glandularis in the cases described by Pawade et al and Kempton et al. We found focal lymphoepithelial lesions in 2 cases, and in both they were also restricted to areas of cystitis glandularis. The rarity of lymphoepithelial lesions in MALT lymphoma of the urinary bladder and the confinement to the areas of cystitis glandularis and cystitis cystica suggest that the transitional epithelium is resistant to invasion by lymphoma cells. Thus, the presence of lymphoepithelial lesions is not mandatory for the diagnosis of MALT lymphoma of the bladder if there are other features typical of MALT lymphomas.

No molecular findings have been reported in previously reported studies of bladder lymphomas. In our cases, IgH gene rearrangement was present in 3 of 4 cases, thus PCR analysis should be useful in the differential diagnosis of chronic cystitis.

Most MALT lymphomas arise in extranodal sites in the setting of acquired MALT.\textsuperscript{46} In these sites, acquired MALT could be induced by Helicobacter pylori infection in the stomach\textsuperscript{46-51} or by autoimmune diseases, such as Sjögren syndrome in salivary glands\textsuperscript{7} or Hashimoto disease in the thyroid gland.\textsuperscript{14} Since there is no naturally occurring lymphoid tissue in the bladder, it is possible that preexisting chronic inflammation can induce acquired MALT. Simpson et al\textsuperscript{59} found that 22% of patients had a history of chronic cystitis, but in most cases convincing histologic evidence of long-standing inflammation was lacking. Ohsawa et al\textsuperscript{60} noted in his review that 20% of patients with primary lymphoma of the urinary bladder had a history of chronic cystitis. Owing to the relatively low percentage of patients with a history of chronic cystitis, some authors have agreed that primary bladder lymphomas are not of the MALT type. The incidence of chronic cystitis may be underestimated. All our patients had a history of chronic cystitis, and most of the recently described cases have the same history.\textsuperscript{22} Escherichia coli infection was found in 3 of our patients; however, we cannot conclude any positive correlation from this small number. The other less likely possible origin of MALT arises from the fact that the bladder is an embryonic derivative of the cloaca, and the bladder lymphoma might arise from inherent lymphoid tissue that is related to Peyer patches in the gut. Most studies, however, have failed to identify lymphoid tissue in healthy bladder.\textsuperscript{22} Luppi et al\textsuperscript{62} demonstrated the presence of hepatitis C virus RNA in 8 of 16 patients with MALT lymphoma of different sites and suggested hepatitis C virus as a potential infectious cofactor in the pathogenesis of MALT lymphoma; however, the relation of hepatitis C virus infection and bladder MALT lymphomas has not been established.

All our patients’ tumors were stage IA, and all were managed with radiotherapy alone. All the patients were in complete remission at the time this article was written, with follow-up periods of 13 years, 5 years, 3 years, and 2 years. We conclude that MALT lymphoma of bladder is more common in women, potentially as a result of increased incidence of chronic cystitis in women. Primary bladder lymphomas are usually low grade and remain localized with a favorable prognosis, although the literature contains 2 reported cases\textsuperscript{22} showing transformation to a diffuse large cell lymphoma. Molecular studies are very useful in confirming the diagnosis by demonstration of monoclonality, especially in small biopsies in which the morphologic examination is not diagnostic alone. Local radiotherapy appears to result in excellent control of the disease.

References


