Minimally Invasive Procedure for Accurate Diagnosis of Mucosa-associated Lymphoid Tissue Lymphoma of the Head and Neck

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Sonography-guided cutting needle biopsy for the diagnosis of malignant lymphoma has recently come into wide use. However, surgery is sometimes unavoidable for the diagnosis of malignant lymphoma, particularly for low-grade malignant lymphoma such as extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue, because cutting needle biopsy offers limited diagnostic accuracy for low-grade malignant lymphoma. Of course, unnecessary invasive procedures like open biopsy should be avoided wherever possible, given the cosmetic problems and burden on the patient. We tried to diagnose malignant lymphoma using the combination of cutting needle biopsy, flow cytometry and polymerase chain reaction to identify monoclonal rearrangement of immunoglobulin heavy chain genes. We have used this method in two cases in whom malignant lymphoma was suspected in the head and neck region, allowing diagnosis of mucosa-associated lymphoid tissue lymphoma in both cases. One case involved a 23-year-old woman with mucosa-associated lymphoid tissue lymphoma in the parotid glands, and the other involved a 77-year-old man with mucosa-associated lymphoid tissue lymphoma in the thyroid. The combination of cutting needle biopsy, flow cytometry and immunoglobulin heavy chain gene rearrangement testing might offer a useful alternative to open biopsy for the diagnosis of mucosa-associated lymphoid tissue lymphoma. We recommend this procedure, particularly for young women or patients with poor performance status in whom malignant lymphoma is suspected.

Key words: extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue – sonography – cutting needle biopsy – flow cytometry – immunoglobulin heavy chain gene rearrangement test

INTRODUCTION

Malignant lymphoma (ML) requires adequate tissue sampling to establish the diagnosis. Image-guided cutting needle biopsy (CNB) for the diagnosis of ML has recently been implemented for a wide variety of organs, with few complications and excellent results. CNB is generally performed using a spring-loaded automatic biopsy gun with side-notch needles (12–18 gauge) and is less invasive than open biopsy. The diagnostic effectiveness of CNB in patients with suspected ML in the head and neck region has also
been studied and a diagnosis of ML with sufficient information such that a therapeutic decision could be made was reportedly obtained in over 90% of ML patients using CNB alone (1,2). On the other hand, Hodgkin’s lymphoma or low-grade ML such as extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue (MALT lymphoma) is sometimes difficult to diagnose using CNB alone and some subtypes of ML remain a diagnostic challenge even after open biopsy (2–5). Such patients presenting to our department with asymptomatic swelling or tumorous lesions in the head and neck region are often otherwise healthy, and in many cases, we have to exclude the possibility of low-grade ML. However, open biopsies are time-consuming and still quite invasive for older patients or patients with poor performance status, and permanent unesthetic scars in conspicuous regions may be problematic for young female patients.

Several studies have reported the usefulness of flow cytometry (FCM) in conjunction with fine-needle aspiration (FNA) in the assessment of lymphoid proliferation, and a diagnosis of ML could be accurately achieved in >80% of ML patients using this method (6,7). However, the diagnostic accuracy of this method varies among lymphoma subtypes and the subclassification of MALT lymphoma is particularly difficult because specific cytological features are often lacking (6). In addition, FCM is nearly universally acknowledged to be of little use in the diagnosis of Hodgkin’s lymphoma (7).

To compensate for the weaknesses of these diagnostic tools in minimally invasive procedures, we tried coupling CNB with FCM for diagnosis in patients for whom low-grade ML is suspected but open biopsy should be avoided. Moreover, polymerase chain reaction testing to identify monoclonal rearrangement of immunoglobulin heavy chain (IgH) joining region genes was applied as an ancillary test to confirm the diagnosis of ML. Rearrangement of IgH genes has been reported as a highly sensitive marker for a wide spectrum of B- and T-cell neoplasms (8). We present herein two cases of MALT lymphoma that we diagnosed using the combination of CNB, FCM and the IgH gene rearrangement test.

CLINICAL PRESENTATION

CASE 1

A 23-year-old Japanese woman was referred to our department for further evaluation of masses in bilateral parotid glands. She had a 5-year history of recurring acute parotitis with fever and pain, which had been treated as mumps or recurrent parotitis at other hospitals. She had experienced about 5–6 episodes/year, achieving remission each time with oral antibiotics. However, the masses in bilateral parotid glands kept growing little by little. She was otherwise in her usual state of health and had no other past medical history. Physical examination revealed several firm swellings in bilateral infra-auricular regions (right > left), with a maximum diameter of 25 mm. About 4 years earlier, the patient had once presented to our department and had been diagnosed with chronic inflammation of the parotid glands based on clinical examination and FNA cytology (class II). Computed tomography (CT) at that time showed diffuse swelling of bilateral parotid glands with some masses and cystic lesions (Fig. 1A). The condition of the disease did not seem to have deteriorated compared with the initial CT (Fig. 1B). Sonographic evaluation revealed the lesions as markedly hypoechoic compared with the surrounding parenchyma, heterogeneous with interspersed linear echogenic strands, and hypervascular compared with the rest of the parenchyma on power Doppler sonography (Fig. 1C and D). ML was therefore suspected. Blood examination revealed positive results for Sjögren’s syndrome (SS)-A and SS-B autoantibodies and rheumatoid factors. No other significantly abnormal data [soluble interleukin-2 receptor (sIL-2r), 415 U/ml; lactate dehydrogenase (LDH), 157 IU/l; IgG4, 23.3 mg/dl; mumps, IgM(−)]. Although gum test and Schirmer’s test results were normal, we considered the possibility of low-grade ML with SS. Considering the importance of cosmetic outcomes, we decided to perform sonography-guided CNB using an 18-gauge cutting needle, and FCM and polymerase chain reaction to diagnose monoclonal IgH gene rearrangement with sonography-guided FNA using 23-gauge needles. For FCM and the IgH gene rearrangement test, separate needle passes were performed and rinsed in 10 ml of physiological saline as a result. CNB showed diffuse infiltration of atypical lymphoid cells with lymphoepithelial lesions (LELs) (Fig. 2). Immunohistochemical study revealed dominancy of CD20-positive cells with negative findings for CD3. Ki-67 index did not seem high. In situ hybridization showed no restriction for K- or λ-immunoglobulin light chains. Although MALT lymphoma was suspected, accurate diagnosis could not be made using CNB alone. FCM showed clonal B-cells with CD19 expression in association with K-light-chain restriction (Fig. 3). In addition, monoclonal IgH gene rearrangement was detected. The diagnosis of MALT lymphoma of the parotid glands was confirmed. Subsequent ¹⁸F-deoxyglucose positron emission tomography (¹⁸FDG-PET) revealed no additional abnormal foci other than the parotid glands (maximum standardized uptake value, 6.11). The patient has been treated using rituximab, and obvious reductions of the masses have been observed along with fewer episodes of acute parotitis.

CASE 2

A 77-year-old Japanese man was referred to our department for further evaluation of a thyroid tumor that had been detected by full-body CT as a follow-up screening after surgery for renal carcinoma (Fig. 4A). About 5 months earlier, immediately before the renal surgery, MALT lymphoma of the rectum had been detected by colon fiberscopy as
the preoperative screening. The rectal lesion had been treated using radiotherapy (RT) (total, 36 Gy) soon after surgery. Performance of 18FDG-PET after RT had not detected any abnormal lesions, including in the rectum and thyroid (Fig. 4B). Blood examination revealed no abnormal data (LDH, 175 IU/l; sIL-2r, 415 U/ml; free-thyroxine, 1.33 ng/dl; thyroid-stimulating hormone, 0.99 μU/ml; thyroglobulin antibodies, 6.1 IU/ml), but a little high value for thyroid peroxidase antibodies (75.8 IU/ml). The sonographic appearance was similar to that in Case 1, and MALT lymphoma was suspected (Fig. 4C and D). Considering the poor performance status, we performed the same diagnostic procedures as in Case 1. CNB showed diffuse infiltration of atypical lymphoid cells. The immunohistochemical study revealed that this tumor was strongly positive for CD20 with negative results for CD3. The Ki-67 index did not seem to be high. Pancytokeratin-immunostaining (AE1/AE3) revealed obvious LELs. However, immunoglobulin light-chain restriction was not detected by in situ hybridization. Although MALT lymphoma was suspected, accurate diagnosis could not be made using CNB alone. FCM showed clonal B-cells with CD19 expression in association with κ-light chain restriction (Fig. 5). In addition, monoclonal IgH gene rearrangement yielded positive results. The
diagnosis of MALT lymphoma of thyroid was confirmed. Three months later, after reconfirming complete response of rectal MALT lymphoma to RT on colon fiberscopy, subtotal thyroidectomy was performed. The post-surgical course was good.

DISCUSSION

We first suspected the present two cases as MALT lymphoma on the basis of the clinical history and imaging studies. Sonography may represent the most useful tool to differentiate benign lesions from ML, although the subtype of ML cannot be determined using this examination (4). Our cases showed characteristic sonographic features, in that the lesions were well-demarcated, markedly hypoechoic, hypervascular and contained interspread linear echogenic strands. Sonographic features of our cases were very similar to those of some other reports and these findings seem to be one of the characteristics for MALT lymphoma of the head and neck region (9–11). We think that sonographic evaluation should be the initial radiological procedure for patients in whom ML is suspected. CT and MRI often reveal MALT lymphoma in the salivary gland as localized or diffuse lesions accompanied by multiple cysts or calcifications. These are not specific findings for MALT lymphoma, but have also been described in lymphoproliferative disorders like SS or acquired immunodeficiency syndrome (12). However, at the very least, these CT and MRI findings suggest the possibility of MALT lymphoma. On the other hand, CT and MRI findings for thyroid lymphoma are entirely non-specific, appearing as ill-defined hypodense nodules, with low contrast enhancement (13). Actually, thyroid lymphoma is often identified from pathological findings in a gland resected for an apparently benign disease, and correct preoperative diagnosis is extremely difficult (14). The present study might have overlooked the thyroid lesion as a benign nodule, if sonographic evaluation had not been performed. Although non-Hodgkin lymphoma reportedly showed an overall 18F-FDG-avidity of 91%, avidity was lower in less-aggressive ML, and MALT lymphoma demonstrated an 18F-FDG avidity of only 54% (15). Conversely, 18F-FDG uptake in benign lesions of the salivary gland or thyroid is not uncommon (16,17). While 18FDG-PET has been considered the first-line modality for staging, restaging or monitoring therapeutic response in ML, it appears unreliable for detecting MALT lymphoma.

Histologically, LELs are one of the characteristic features of MALT lymphoma, which most frequently affect the salivary gland, thyroid and lungs (18), but LELs are also commonly observed in inflammatory diseases like SS, and
distinguishing between benign LEIs and MALT lymphoma represents a difficult diagnostic problem (19). The pattern of architectural findings is very important for histological diagnosis and distinguishing reactive lesions from low-grade ML with CNB alone is difficult (20). Dominantly of CD20-positive cells and low Ki-67-labeling index should be correlated with, but not an absolute criterion of, MALT lymphoma. Overt ML may not show clonality in paraffin-section immunohistochemistry and it is not possible to show restriction of $\kappa$- or $\lambda$-immunoglobulin light-chain in all cases of MALT lymphoma even in FCM, due to the frequent presence of interspersed reactive B cells (13). Fortunately, in both of the present cases, significant light-chain predominance was observed by FCM. Since expression of light chains was most intense in plasma cells and plasmacytoid lymphocytes (21), detection of cytoplasmic $\kappa$- or $\lambda$-expression in CD20-positive cells of MALT lymphoma in the paraffin-embedded sections might be difficult. On the other hand, FCM detects immunoglobulin at the cell surface and so could show restriction of $\kappa$- or $\lambda$-immunoglobulin light chains in the present study. Analysis of monoclonal IgH gene rearrangements does not permit histological subtyping of B-cell tumors, but this technique is helpful in distinguishing B-cell neoplasms among lymphoproliferative disorders that are difficult to evaluate histologically or that lack distinctive antigenic markers (22). In brief, we strongly suspected these two cases as representing MALT lymphoma from imaging results and pathological findings from CNB, and then confirmed the diagnoses using FCM and analysis of IgH rearrangement. The weak points of this method might be as follows. First, samples need to be acquired three times from patients, involving more pain and probably a higher risk of hematoma. Secondly, the cost of this method will be slightly higher than that of open biopsy. Finally, this method could be performed only in limited institutions like a hospital attached to a university.

Although MALT lymphoma in the salivary gland is usually an indolent neoplasm that tends to remain localized for long periods of time even without treatment, the tumor may disseminate or undergo high-grade transformation (23). The salivary glands do not normally contain MALT, but can acquire it as a result of chronic inflammation, which usually has an autoimmune basis, such as SS. SS is associated with $\sim 44$-fold increase in risk of ML compared with the general population (24). Rituximab, a chimeric monoclonal antibody targeting CD20-positive B-cells, has recently been reported as potentially effective not only in the treatment of non-Hodgkin’s lymphoma, but also in the treatment of SS (25). In particular, SS patients with higher levels of residual exocrine gland function might benefit more from rituximab (26).

We thus chose rituximab therapy for Case 1 in order to treat MALT lymphoma and SS at the same time, while preserving exocrine gland function.

Primary MALT lymphomas of the thyroid almost invariably develop from Hashimoto’s thyroiditis (27). The Case 2 might also have had inapparent Hashimoto’s thyroiditis. The prognosis of patients with primary thyroid lymphoma, especially MALT lymphoma, appears to be favorable and most are treated with RT alone, but the optimal treatment modality for this disease remains yet to be defined (28). We chose surgical treatment for this 77-year-old man, considering the limitation of the tumor within the thyroid capsule, the relatively short period of surgical therapy in comparison with RT, and the deleterious effects of RT on swallowing function and mucositis inflammation.

The combination of CNB, FCM and IgH gene rearrangement test might be effective as a relatively non-invasive means of diagnosing MALT lymphoma in the head and neck region. Sonographic evaluation is essential for patients in whom the possibility of ML must be excluded. In a future study, we want to use this method for the diagnosis of more patients in whom not only MALT lymphoma, but also the other types of ML are suspected in the head and neck region.

Conflict of interest statement

None declared.

References

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