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Case Report

Diffuse large B-cell lymphoma of the uterus suspected of having transformed from a marginal zone B-cell lymphoma harboring trisomy 18: a case report and review of the literature

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Abstract: The patient was a 72-year-old female with the chief complaint of abdominal fullness. A giant primary myoma of the uterine cervix was suspected, and total hysterectomy was performed. Based on a postoperative histopathological examination of the tumor a diagnosis of diffuse large B-cell lymphoma (DLBCL) was made in the uterus and a mass in the greater omentum was diagnosed as a marginal zone B-cell lymphoma (MZBCL). No flow-cytometry studies or chromosome or gene examinations were performed on a fresh specimen. The results of an examination of a paraffin block histopathology specimen by fluorescence in-situ hybridization (FISH) showed no *mucosa associated lymphoid tissue lymphoma translocation gene 1 (MALT1)* (18q21.1), *B-cell lymphoma 2 (BCL2)* (18q21.3), or *BCL6* (3q27) split signals in either the uterus or the greater omentum, however, trisomy 18 was detected in approximately 50%-70% of the tumor cells in both the uterus and the greater omentum. Trisomy 18 was present in around 15-33% of the DLBCL cells and MZBCL cells. These findings suggested a strong possibility that the tumor cells in the uterus and greater omentum were the same clone and that transformation from MZBCL to DLBCL had occurred. Since DLBCLs that result from a transformation usually have a worse outcome than de novo DLBCLs, even when a DLBCL seems to have originated in the uterus the surrounding tissue should always be examined, and caution should be exercised in regard to transformation from a low-grade B-cell lymphoma to a DLBCL.

Keywords: Marginal zone lymphoma, diffuse large B-cell lymphoma, transformation, trisomy 18, uterus, greater omentum

Introduction

Although infiltration of the uterus during the course of a malignant lymphoma is not rare, few malignant lymphomas originate in the uterus, and in a study of a large number of cases it was reported that they accounted for 0.5% of extranodal lymphomas, that they were most common in the cervical region and according to histological type were diffuse large B-cell lymphomas (DLBCLs) [1]. Even when the uterus is the main lesion site clinically, in the advanced stage it is difficult to determine whether the uterus is the primary site or a site of infiltration.

We report the case of a patient who developed a giant uterine mass in whom a marginal zone B-cell lymphoma (MZBCL) of the surrounding tissue, including the greater omentum, appeared to have transformed into DLBCL and infiltrated the uterus. Even when some fresh specimen is not available, *B-cell lymphoma 2 (BCL2)* (18q21.3), *BCL6* (3q27), *immunoglobulin heavy-chain (IgH)* (14q32), and *mucosa associated lymphoid tissue lymphoma translocation gene 1 (MALT1)* (18q21.1) split signals can be measured by paraffin-embedded tissue section-fluorescence in situ hybridization (PS-FISH) on tissue sections prepared from par-

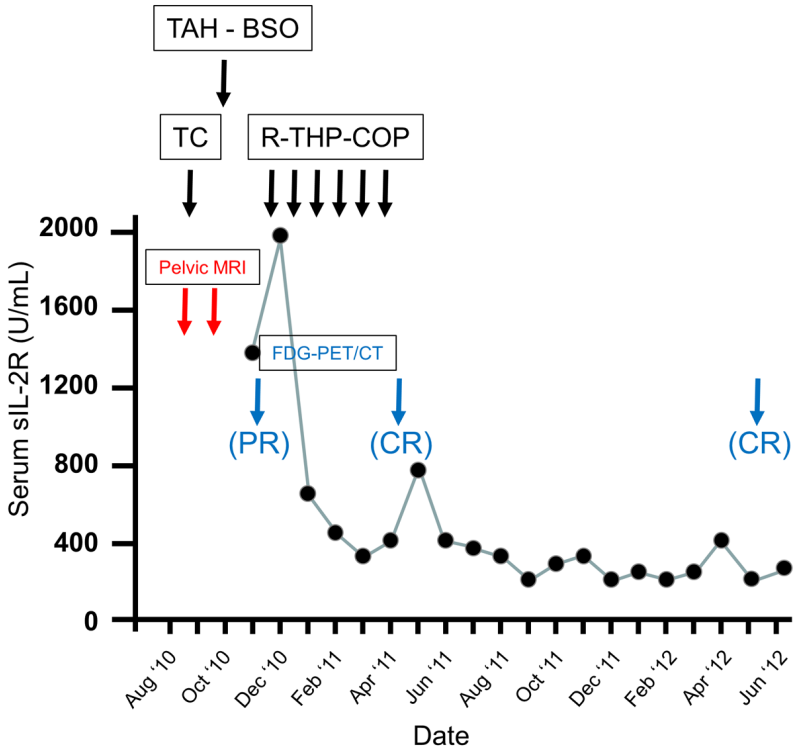

Figure 1. Clinical course. In August 2010, an magnetic resonance imaging (MRI) examination performed after TC therapy (paclitaxel and carboplatin) showed regression of the uterine tumor, and total abdominal hysterectomy with bilateral salpingo-oophorectomy (TAH-BSO) was performed in September, but ^{18}F-fluoro-2-deoxy-D-glucose-positron emission tomography/computed tomography (FDG-PET/CT) demonstrated residual lesions, and the serum soluble interleukin-2 receptor (sIL-2R) level (1380 U/mL) was elevated. After rituximab plus THP-COP (THP-adriamycin, cyclophosphamide, vincristine, and prednisone) chemotherapy, the serum sIL-2R level rapidly decreased to within the normal range, and a complete remission was confirmed by FDG-PET/CT in April 2011 and in May 2012.

affin block specimens, and the results are useful. Trisomy 18 is detected in approximately 16% of MZBCLs the cells, and trisomy 3 in approximately 37%, and with MALT1-API2 fusion protein being present in approximately 19%, these findings are sometimes more useful [2]. Thus, when a fresh specimen is not available, it is worth trying FISH on tissue sections when making the differential diagnosis, especially of low-grade B-cell lymphomas.

Case report

The patient was a 72-year-old woman with the chief complaint of abdominal fullness. The patient's past history was unremarkable. Her allergy history revealed having developed urticaria as a reaction to a computed tomography (CT) contrast medium. The history of the present illness showed that the patient first noticed abdominal distention in June 2010. In July she was examined by a local physician because she had developed a fever of 37°C, night sweats, had lost approximately 13% of her body weight over a 3-month period (down from 40 kg to 35 kg), and had developed bilateral lower leg swelling. Because a blood examination revealed high lactate dehydrogenase (LDH) and alkaline phosphatase (ALP) values, and the results of an magnetic resonance imaging (MRI) examination showed a giant mass measuring approximately 21 cm x 10 cm extending from the upper abdomen to the pelvis and aortic lymph node enlargement, in August 2010 the patient was referred to the gynecology department of our hospital and examined.

At the time of the initial examination the patient's body temperature was 37.5°C. No superficial lymph nodes were palpable. A giant, elastic-hard, smooth-surfaced mass that was about the size of a small childs head and centered in the umbilical area was palpated. There was no hepatosplenomegaly. The results of the laboratory studies showed elevated serum LDH (1029 IU/L) and serum ALP (434 IU/L) values. None of the other blood or biochemistry test values were abnormal. Tumor marker measurements showed a mildly elevated cancer antigen (CA) 125 level (49.4 U/mL), but the carcinoembryonic antigen (CEA) and CA19-9 levels were within normal limits. Serum soluble interleukin-2 receptor (sIL-2R) was not measured at the time of the initial examination.

At first, a uterine sarcoma was suspected, and in August 2010 the patient was admitted to the gynecology department of our hospital for a

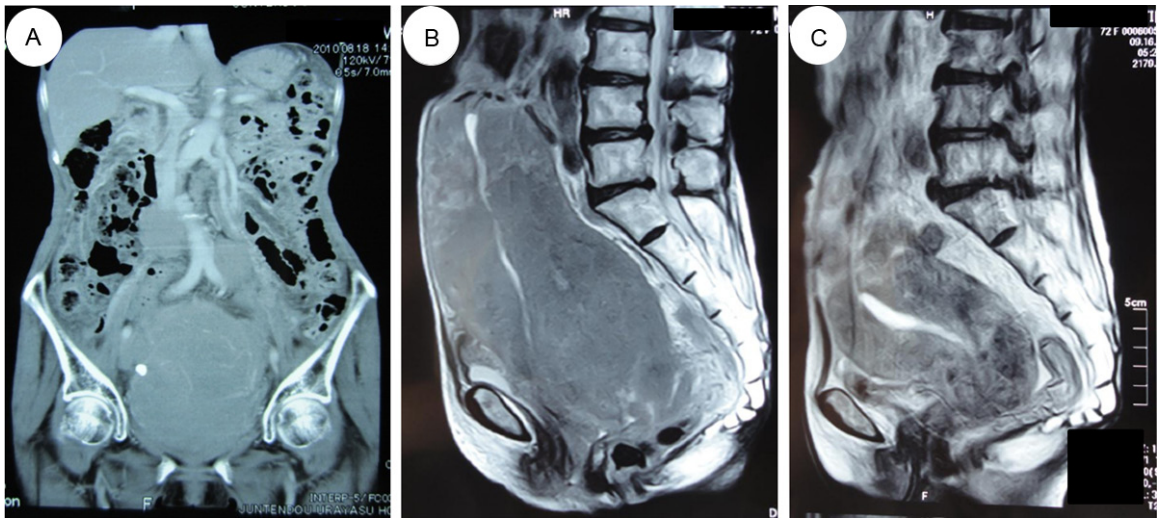

Figure 2. Pelvic computed tomography (CT) and Pelvic magnetic resonance imaging (MRI). A: The CT image before treatment showed intrapelvic lymph node enlargement. B: The MRI scan before treatment showed an enhanced uterine tumor with indistinct boundaries that measured 20 cm in diameter. C: MRI scan showing marked regression of the tumor after TC therapy (paclitaxel and carboplatin).

thorough examination and treatment. The patient's clinical course is summarized in **Figure 1**. The results of scraping cytology of the vaginal portion of the uterus and the endometrium were class II to class III, but because endometrial biopsy specimen was insufficient, it was impossible to make a definite diagnosis.

A thoracoabdominal CT examination showed an approximately 140 mm x 140 mm tumor lesion in the pelvis that was internally heterogeneously faintly enhanced and accompanied by calcification, and enlargement of the para-abdominal aortic lymph nodes, lymph nodes around the inferior vena cava, right obturator lymph nodes, right external iliac lymph nodes, and left common iliac lymph nodes (**Figure 2A**). Although there were scattered post-inflammatory changes in the lung fields and pleura, no enlargement of the mediastinal or pulmonary hilar lymph nodes or pleural fluid accumulation was observed. The results of a pelvic MRI examination (T2-weighted images) showed overall enlargement of the uterus, and the uterine fundus had reached the level of the upper margin of the 4th lumbar vertebra. There were numerous myoma-like masses in the myometrium, many of them having indistinct boundaries, and heterogeneous enhancement was observed with gadolinium contrast medium (**Figure 2B**).

Based on the MRI findings, a uterine sarcoma was suspected at first, but because the gigantic size of the tumor, it was concluded that total excision of the uterine myoma would be difficult, and TC therapy (paclitaxel and carboplatin) was started on hospital day 8. Although acute renal failure developed due to a temporary tumor lysis syndrome, it improved in response to hemodialysis therapy. An MRI examination after the chemotherapy showed that the uterine fundus had shrunk to the point that it only reached 1/2 the height of the sacrum (long diameter of the uterus: 20 cm x 12 cm), and the chemotherapy was very effective (**Figure 2C**). On hospital day 40 a simple abdominal hysterectomy and bilateral adnexectomy was performed. Although disseminated intravascular coagulation (DIC) developed intraoperatively, the symptoms were relieved by transfusion of fresh frozen plasma, etc., and the patient was discharged on hospital day 66.

In late October 2010 the patient was referred to our department and examined. Her serum sIL-2R level was elevated (1380 U/mL). A bone marrow examination did not show any evidence of marrow infiltration. ^{18}F-fluoro-2-deoxy-D-glucose-positron emission tomography/computed tomography (FDG-PET/CT) revealed residual lesions in the form of numerous nodules accompanied by increased FDG accumulation in the anterior mediastinum (maximum Standardized Uptake Value (SUVmax): 4.5), in the diaphragm (SUVmax: 9.8), and in the mes-

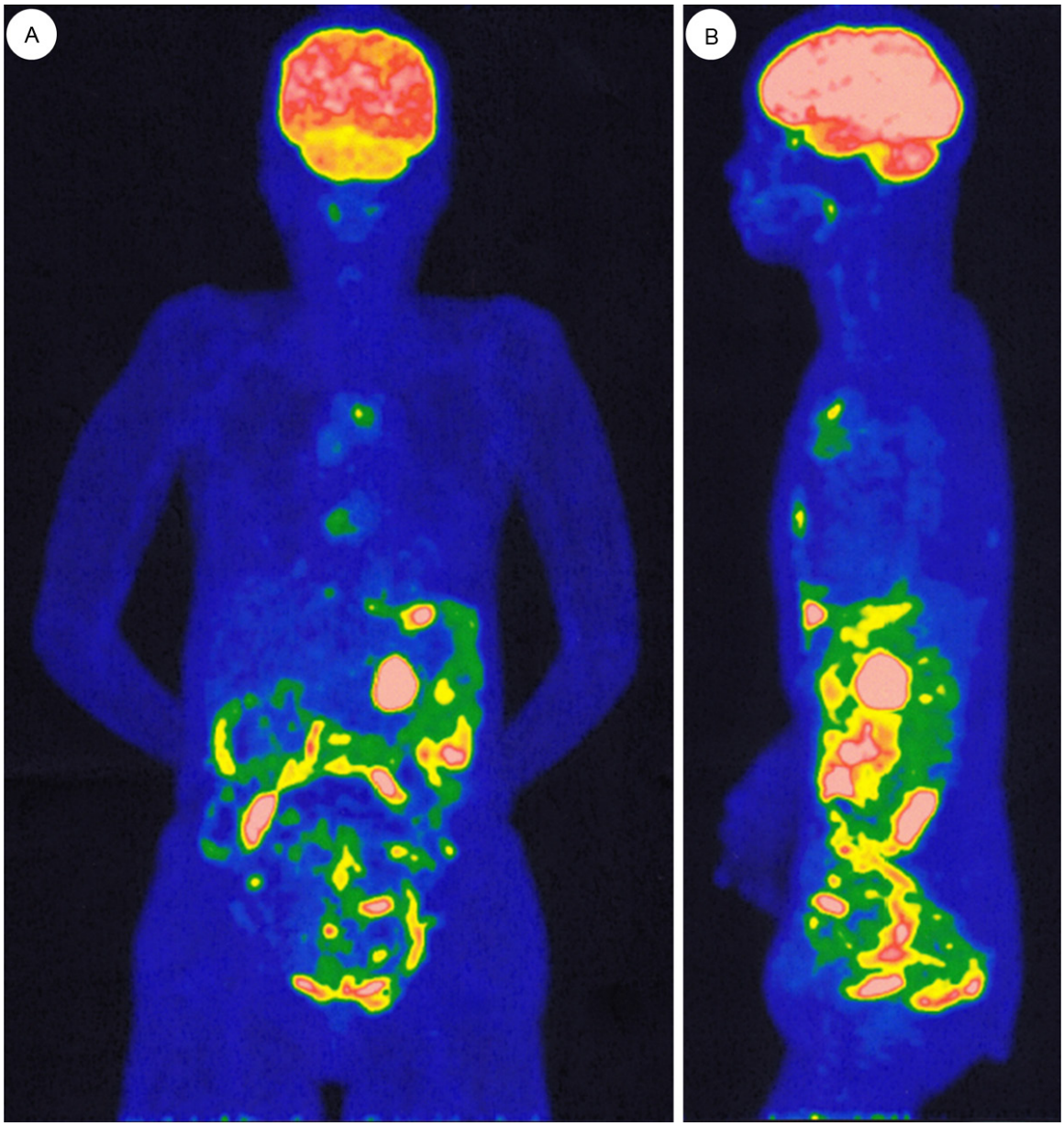

Figure 3. ^{18}F-fluoro-2-deoxy-D-glucose-positron emission tomography/computed tomography image (FDG-PET/CT) before Rituximab and THP-COP (THP-adriamycin, cyclophosphamide, vincristine, and prednisone) regimen. A: Frontal view. B: Lateral view. Numerous nodules accompanied by increased ^{18}F-fluoro-2-deoxy-D-glucose (FDG) accumulation were observed in the anterior mediastinum (maximum Standardized Uptake Value (SUVmax) = 4.5), diaphragm (SUVmax = 9.8), and mesentery and peritoneum (SUVmax = 12.4). Thickening accompanied by FDG accumulation were observed in the diaphragm and peritoneum. Bilateral pleural effusion and ascites were observed. A mass lesion (SUVmax = 21.5) measuring 3 cm in diameter accompanied by increased FDG was seen in the vicinity of the left adrenal gland.

entery and peritoneum (SUVmax: 12.4) (**Figure 3**). Thickening accompanied by FDG accumulation was observed in the diaphragm and peritoneum, and bilateral pleural effusion and ascites were observed. A mass lesion measuring 3 cm in diameter accompanied by increased FDG accumulation (SUVmax: 21.5) was seen in the vicinity of the left adrenal gland.

Histopathological examination of the uterine tumor showed diffuse growth by large cells containing multiple nucleoli, and immunostaining

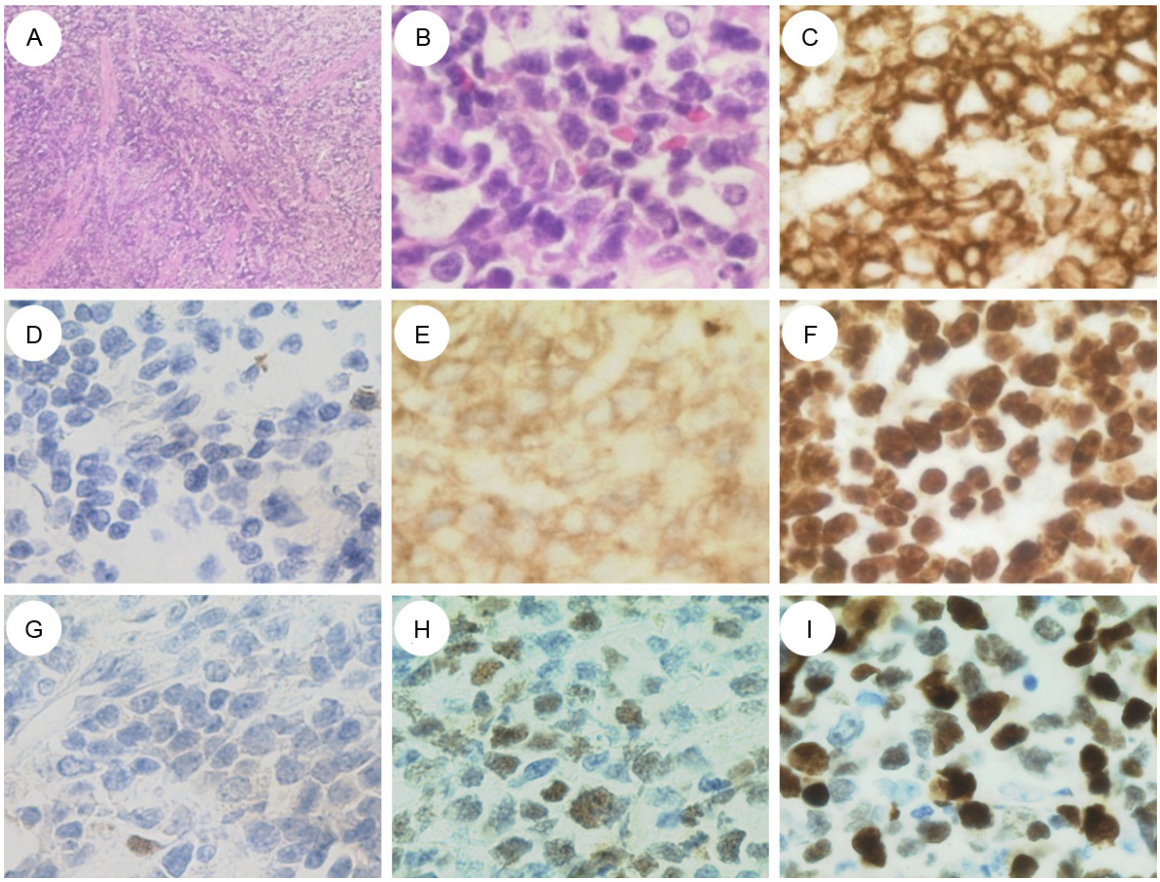

Figure 4. Immuno-histological findings (Uterus). A: Hematoxylin-eosin (HE) stain, 40x, B: HE stain, 600x, showing diffuse large-cell infiltration. C-I: 600x. C: The invading large cells were CD20-positive. D: CD5-negative. E: CD10 was weakly positive. F: The Ki-67 (MIB-1) labeling index was high, over 90%. G: Cyclin D1-negative. H: BCL6-negative. I: MUM1 was negative.

revealed that the tumor cells were CD20-positive and weakly CD10-positive, and that a high proportion of them were Ki-67 (MIB-1)-positive (**Figure 4**). The tumor cells in the greater omentum ranged from small to intermediate in size, were CD10-negative, and had a low Ki-67-positive rate, and it was a low-grade B-cell lymphoma. CD5, cyclin D1, and CD23 were all negative, thereby ruling out a mantle cell lymphoma or small lymphocytic lymphoma, and the tumor appeared to be an MZBCL (**Figure 5**).

No split signals were observed when PS-FISH using *BCL2* (18q21.3), *MALT1* (18q21.1), and *BCL6* (3q27) as probes was performed on the uterine tumor and greater omentum, but three *BCL2* signals were observed in 51% of the uterine cells and in 61% of the greater omentum cells, and three *MALT1* signals were observed in 72% of the uterine cells and in 54% of the greater omentum cells (**Figure 6**). This appeared to mean that trisomy 18 was present in both the uterus and the greater omentum, and our diagnosis was that transformation into a DLBCL had occurred in an MZBCL that was present in the uterus and had caused infiltration of the uterus. Because trisomy 18 was not present in the bone marrow and there was no bone marrow infiltration, and there were no chromosome abnormalities in the somatic cells, the trisomy 18 chromosome abnormality appeared to have been present in the malignant lymphoma alone. Moreover, no split-signals or three signals of *BCL6* (3q27) were detected in either the uterus or the greater omentum (**Figure 6**).

Because of DLBCL with high-risk International Prognostic Index (IPI) score, the patient was treated with 6 courses of rituximab in combination with THP-COP (THP-adriamycin, cyclophosphamide, vincristine, and prednisone). Treat-

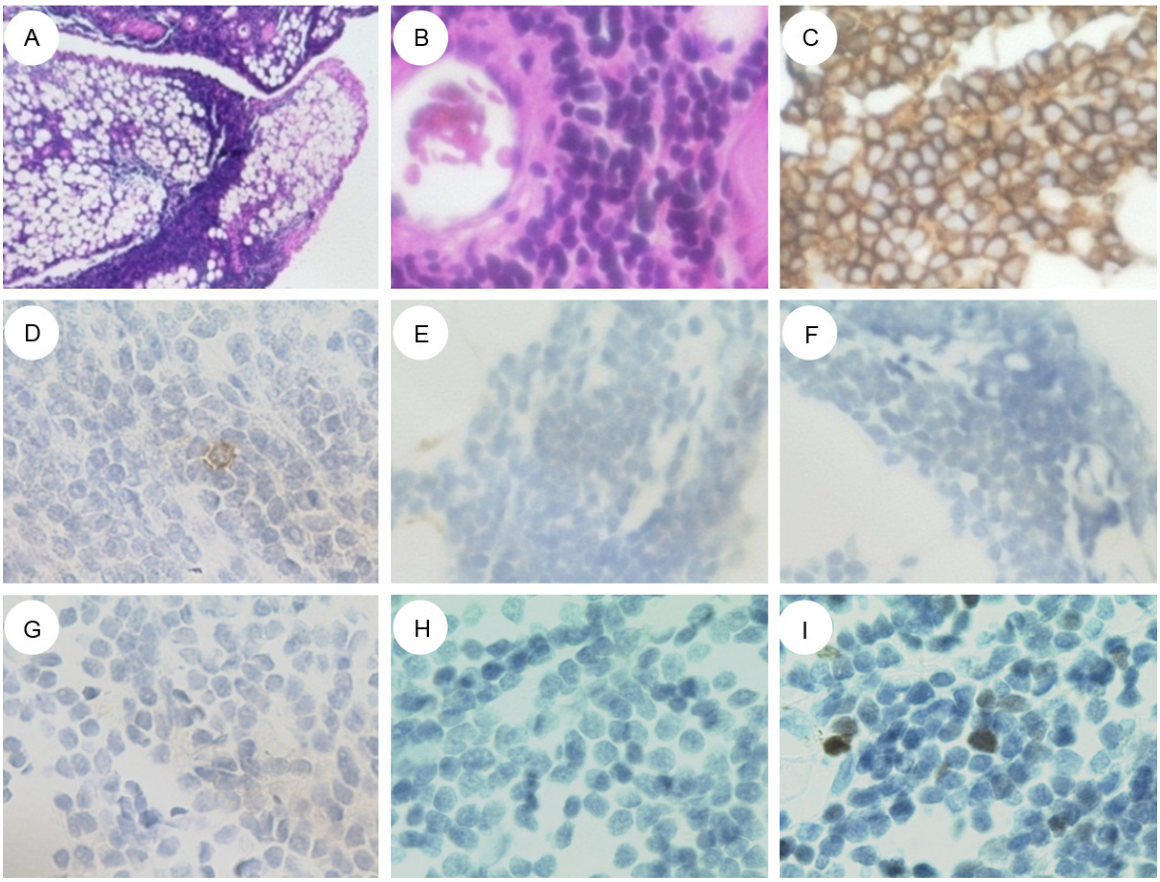

Figure 5. Immuno-histological findings (greater omentum). A: Hematoxylin-eosin (HE) stain, 40x, B: HE stain, 600x, infiltration by small round cells was seen. C-I: 600x. C: The invading small cells were CD20-positive. D: CD5 was negative. E: CD10 was negative. F: The Ki-67 (MIB-1) labeling index was low, less than 5%, and the lesion was low-grade. G: Cyclin D1 was negative. H: BCL6-negative. I: MUM1 was negative.

ment was followed by a complete remission, and FDG-PET/CT in May 2013 showed no evidence of relapse (data not shown). The complete remission has been maintained even after 36 months has passed since the onset of symptoms.

Discussion

Cervical canal mucus cytology is often negative in primary lymphomas of the uterus, and even in lymphoma of the cervix it is positive in only 10-40% of the cases [3]. Cytology was negative in our patient as well.

In a study by Harris NL et al. the histopathology of the primary lymphomas of the uterus was DLBCL in 67% and follicular lymphoma (FL) in 28% [4]. Infiltration of the uterine cervix was observed in 6% of female autopsy cases of non-Hodgkin's lymphoma (NHL), and infiltration of the uterine body in 10%, and there were high percentages of Burkitt lymphoma and DLBCL [5]. MZBCL of the uterus is rare, with only 12 cases ever having been reported (**Table 1**) [6-17], and transformation was suggested in only one of them. Many MZBCLs of the uterus have a low-risk IPI score. Many of the patients survive, and the prognosis is good. Since the Ki-67 index was high in the four stage IV cases in which the bone marrow had been infiltrated and in our own case, which was stage IV and in which there was extensive infiltration of the full thickness of the uterus, when our case is included, there seem to have been 2 cases of transformation among the total of 13 cases of MZBCL reported. A combination of surgical resection of the uterus and chemotherapy was useful in both of these cases. In many of the 13 cases of MZBCL of the uterus in **Table 2** the surface markers were CD5-negative, CD10-

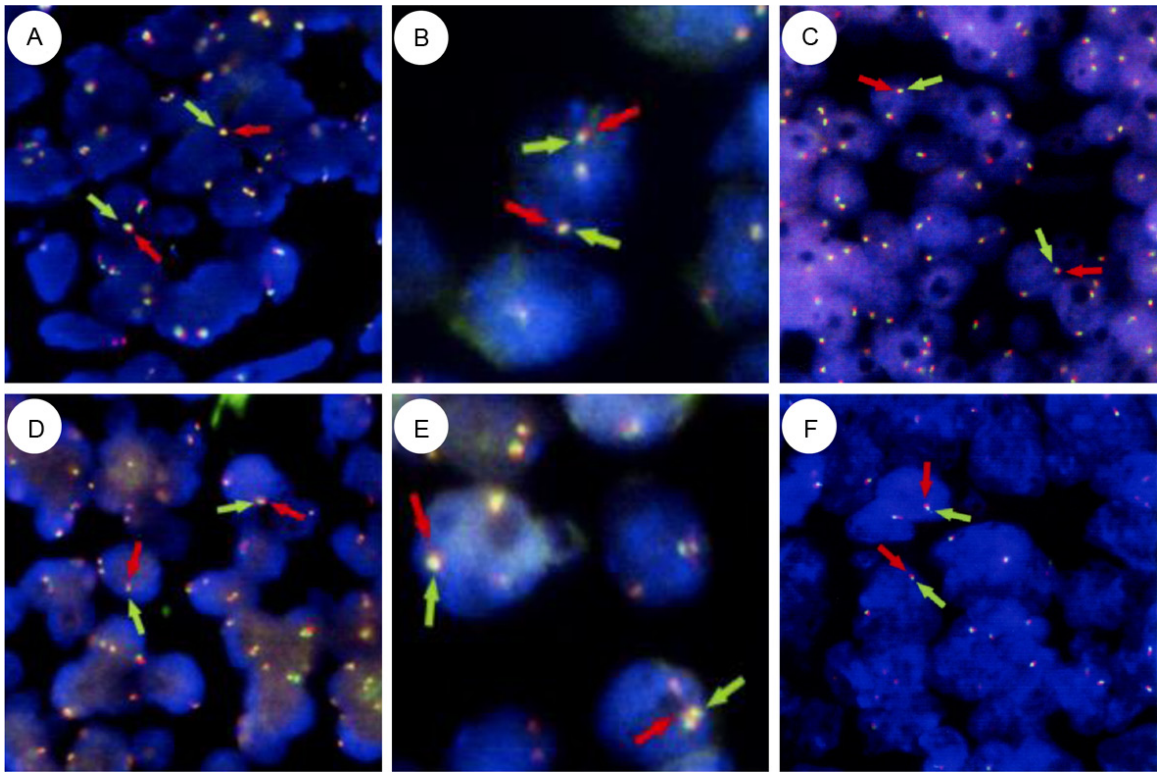

Figure 6. Trisomy 18 detected by PS-FISH. A: Uterus (BCL2). The proportion of cells that had findings (split signals) suggesting a break of the region within the *BCL2 gene* (18q21.3) was 0.0%, but tumor cells that seemed to have trisomy 18 that showed a red signal (telomere side):green signal (centromere side) ratio of 3:3 or more accounted for 51.0% of the total. B: Uterus (MALT-1). The proportion of cells that had findings (split signals) suggesting a break of the region within the *MALT-1 gene* (18q21.1) was 0.0%, but, similarly, tumor cells that seemed to have trisomy 18 that showed a red signal (telomere side):green signal (centromere side) ratio of 3:3 or more accounted for 72.0% of the total. C: Uterus (BCL6). The proportion of cells that had findings (split signals) suggesting a break of the region within the *BCL6 gene* (3q27) was 0.0%. D: Omentum (BCL2). The proportion of cells that had findings (split signals) suggesting a break within the region of the *BCL2 gene* was 0.0%, but tumor cells that seemed to have trisomy 18 that showed a red signal (telomere side):green signal (centromere side) ratio of 3:3 or more accounted for 61.0% of the total. E: Omentum (MALT-1). The proportion of cells that had findings (split signals) suggesting a break within the region of the MALT-1 gene (18q21.1) was 0.0%, but, similarly, tumor cells that seemed to have trisomy 18 that showed a red signal (telomere side):green signal (centromere side) ratio of 3:3 or more accounted for 54.0% of the total. F: Omentum (BCL6). The proportion of cells that had findings (split signals) suggesting a break within the region of the *BCL6 gene* (3q27) was 0.0%.

negative, CD20-positive, BCL6-negative, and cyclin D1-negative, and they were consistent with being an MZBCL.

In the past FISH was performed on cells that were used for chromosome analysis, but it has also become possible to use it on paraffin sections, and confirmation of translocations, reassessments of retrospective cases, and studies focused on lesion sites can now be conducted on paraffin block specimens in cases in which a chromosome analysis has not been performed [18]. Because the results of the PS-FISH analysis in our own case revealed the presence of three signals for the 18q21 segment in both the uterine DLBCL and the greater omentum MZBCL, the presence of the same trisomy 18 chromosome abnormalities suggested the possibility of being the same clone. It was impossible to confirm a translocation having the same breakpoint in *BCL2* or *BCL6*, and thus there appeared to be little possibility of being an FL. It has been reported that trisomy 18 is present in about 17% of DLBCLs, and they are usually hyperdiploid or have multiple chromosome rearrangements [19]. Some of them may have transformed from a low-grade B-cell lymphoma. It is said that MZBCLs have a low CD10-positive rate and that trisomy 18 is present in approximately 20% of them [20]. Trisomy 18 is

Table 1. Summary of clinical and staging data for Marginal zone B-cell lymphoma of the uterus

Cases	Age (years)	Transformation	Stage	IPI score	Treatment	Follow-up	Reference
1	40	-	IIEA	0	VH + P-RT	NED, 3 m	[6]
2	72	-	IIEA	1	TAH + P-RT (41 Gy) + PLN-RT (36 Gy)	NED, 11 m	[7]
3	53	-	IEA	0	Cervical conization	NA	[8]
4	46	+	IVEA	2	Polypectomy + 5 courses of proM-ACE/CytaBOM; later TAH because of local relapse	NED, 28 m	[9]
5	65	-	IEA	NA	TAH-BSO	NA	[10]
6	43	-	IIEA	0	TAH-BSO + LNS	NED, 28 m	[11]
7	52	-	IVEB	2	TAH-BSO	NED, 20 m	[12]
8	61	-	IEA	NA	TAH-BSO	NED, 8 m	[13]
9	56	-	NA	NA	TAH-BSO + P-RT + Rituximab	NED, 28 m	[14]
10	81	-	IEA	NA	Observation	NED, 24 m	[15]
11	55	-	IEA	NA	TAH-BSO	NED, 24 m	[23]
12	80	-	IIEA	NA	Observation	NED, 7 m	[17]
13	72	+	IVB	HI	TAH-BSO + LNS + R-THP-COP	NED, 36 m	present case

Abbreviations: AWD, alive with disease; BSO, bilateral salpingo-oophorectomy; H, high risk; HI, high-intermediate risk; IPI, International Prognostic Index; L, low risk; LI, low-intermediate risk; LNS, lymph node sampling; m, months; NA, not available; NED, no evidence of disease; PLN-RT, paraaaortic lymph node radiation therapy; proMACE/CytaBOM, cyclophosphamide, epirubicine, etoposide, prednisone, cytarabine, vincristine, bleomycin, and methotrexate; P-RT, pelvic radiation therapy; R, Rituximab; TAH, total abdominal hysterectomy; THP-COP, THP-adriamycin, cyclophosphamide, vincristine, and prednisone; VH, vaginal hysterectomy.

sometimes also found in FLs, but there is no *BCL2* or *BCL6* breakpoint, and in our own case there appeared to be a stronger possibility of being an MZBCL than an FL. The same as in our own case, even when a lesion seems to be a primary DLBCL of the uterus, examination by PS-FISH should be performed if there is a low-grade B-cell lymphoma in a lymph node around it.

In the past a variety of methods that combined surgical resection, radiation therapy, and chemotherapy have been used to treat primary malignant lymphomas of the uterus [4, 21]. The prognosis is relatively good, and the 5-year survival rate has been reported to be 73% [4, 22]. The prognosis of DLBCLs that have been resulted from a transformation, however, is generally poor, with a mean survival time after transformation of 1-2 years, and the cause of death in most cases is attributable to the DLBCL. An R-CHOP-like regimen should be performed in treatment-naive cases, and autologous stem-cell transplantation (ASCT), or depending on the case, reduced intensity transplantation (RIT), should be considered in high-risk or high-intermediate-risk IPI cases in patients age 65 years old or under. Because of the higher treatment-related mortality (TRM) and relapse rates in patients who receive a myeloablative allogeneic transplant, the results have usually been better in patients who have received autologous transplants than in patients who have received an allogeneic transplant.

Our case was a high-risk DLBCL case according to the IPI, and R-THP-COP therapy was performed. Because our patient was elderly (72 years old), ASCT entails high TRM, and maintenance therapy with rituximab, etc., for potentially residual MZBCL may be appropriate.

To recapitulate, it is difficult to make a definitive differential diagnosis of diseases that cause diffuse enlargement of the uterus based on cytology or a biopsy. Moreover, even when the disease is a malignant lymphoma, it is difficult to determine whether it is a primary lymphoma or is the result of infiltration. Because of the possibility of being the result of a transformation, advanced cases of DLBCL with a high Ki-67 index that have diffusely infiltrated the uterus or infiltrated the bone marrow, in particular, should be tested to determine whether transformation has occurred by exploring for the presence of a low-grade lymphoma in the

Table 2. Comparison of immunophenotypes in MZBCL of uterus

Cases	Site	TF	CD5	CD10	CD20	BCL2	BCL6	MUM1	Ki-67 (%)	Cyclin D1	Reference
1	Uterus	-	NA	NA	NA	NA	NA	NA	NA	NA	[6]
2	Uterus	-	NA	NA	+	NA	NA	NA	NA	-	[7]
2	Paraaortic LN	-	+	-	+	NA	NA	NA	NA	-	[7]
3	Uterus	-	NA	NA	NA	NA	NA	NA	NA	NA	[8]
4	Uterus	+	-	-	+	-	NA	NA	>70	-	[9]
4	Bone marrow	-	-	-	+	-	NA	NA	-	-	[9]
5	Uterus	-	-	-	+	+	-	NA	NA	-	[10]
6	Uterus	-	NA	NA	NA	NA	NA	NA	NA	NA	[11]
7	Uterus	-	NA	NA	+	NA	NA	NA	NA	NA	[12]
8	Uterus	-	-	-	+	NA	-	NA	ME	NA	[13]
9	Uterus	-	-	-	+	NA	NA	NA	NA	NA	[14]
10	Uterus	-	NA	-	+	NA	±	NA	<5	NA	[15]
11	Uterus	-	±	-	+	+	NA	NA	<5	-	[23]
12	Uterus	-	-	-	+	NA	-	NA	<1	NA	[17]
13	Uterus	+	-	+	+	+	±	±	>90	-	present case
13	Omentum	-	-	-	+	+	±	±	<5	-	present case

Abbreviations: LN, lymph node; ME, moderate expression; MZBCL, marginal zone B-cell lymphoma; NA, not available; TF, transformation.

greater omentum or surrounding lymph nodes, etc., by FDG-PET/CT and examining for the presence of transformation by performing a biopsy and chromosome analysis, PS-FISH, or gene exploration. When transformation is found to have occurred, if a complete remission is achieved with a CHOP-like regimen, it appeared necessary to consider maintenance therapy with rituximab, etc., or else ASCT.

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Disclosure of conflict of interest

None.

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