Re-evaluation of preoperative endometrial smears for the cytodiagnosis of uterine leiomyosarcoma

K. Sonoda1,2, M. Nogami4, K. Kodama2, Y. Oda3, K. Kato2
1Gynecologic Service, National Kyushu Cancer Center, Fukuoka
2Department of Obstetrics and Gynecology, 3Department of Anatomic Pathology, Graduate School of Medical Sciences, Kyushu University, Fukuoka
4Division of Diagnostic Pathology, Kyushu University Hospital, Fukuoka (Japan)

Summary
Introduction: Uterine leiomyosarcoma (ULS) is a highly aggressive and lethal tumor. The absence of specific symptoms and diagnostic imaging findings makes the diagnosis of ULS challenging. Endometrial sampling reportedly has a significantly lower predictive value in diagnosing ULS compared with epithelial uterine malignancies.

Methods: The authors retrospectively reviewed the preoperative endometrial cytology findings in seven of the 12 patients with ULS who were treated in this institution between 2008 and 2017. The other five patients did not have preoperative samples obtained for cytology.

Results: Only one of the seven patients was originally diagnosed with a malignant tumor, with preoperative cytology showing rounded tumor cells with enlarged, irregular, hyperchromatic, and sometimes multiple nuclei, with conspicuous nucleoli in a necrotic background. After re-evaluation of the preoperative specimens, atypical cells were detected in four of the six patients who were initially deemed to have negative findings. A monomorphic population of spindle cells with slightly hyperchromatic, elongated nuclei was detected in two patients. Another two patients had rounded tumor cells with a moderate amount of basophilic cytoplasm and round or oval nuclei with minimal atypia.

Conclusion: Novel diagnostic techniques are needed to accurately identify ULS in the preoperative period. Careful microscopic observation of the entire cytological specimen, together with detailed patient medical information, are essential to making a correct preoperative diagnosis of ULS.

Key words: Uterine neoplasms; Leiomyosarcoma; Cytodiagnosis.

Introduction
Uterine sarcomas are rare tumors that account for approximately 1% of female genital tract malignancies and 3% to 7% of uterine cancers [1]. Uterine leiomyosarcoma (ULS) remains the most common type of uterine sarcoma, affecting approximately 0.4 of 100,000 women every year [2]. The absence of specific symptoms and diagnostic imaging findings makes the diagnosis of ULS challenging, and most patients are diagnosed after the disease has already progressed to an advanced stage. A study of the Surveillance, Epidemiology, and End Results (SEER) database revealed that most tumors are Stage I (68%), while 3% and 7% of patients are diagnosed at Stages II and III, respectively, and 22% are diagnosed with Stage IV disease. The five-year survival estimate for Stages I, II, III, and IV is reportedly 76%, 60%, 45%, and 29%, respectively [3].

The combined use of dynamic MRI and serum measurement of lactate dehydrogenase (LDH; specifically, the LDH3 isozyme) may be useful for preoperative differentiation of ULS from degenerated leiomyoma [4]. Preoperative endometrial sampling correctly identifies only 25% to 50% of patients with ULS, which may be in part because of the myometrial origin of the tumor [5].

The authors performed a retrospective review of the preoperative endometrial cytology findings of patients with ULS who were treated at this institution in an attempt to elucidate an examination method that enables the early diagnosis of ULS.

Materials and Methods
The authors reviewed the records of patients with ULS who were treated at Kyushu University Hospital between January 2008 and June 2017. Patients underwent multidisciplinary therapy and were followed every three months for the first year after treatment, then every four months for the second year, biannually for the third through fifth years, and annually thereafter [6]. Follow-up included an intensive history, physical and pelvic examination, Papanicolaou smear, serum tumor-marker levels, and imaging studies. The authors excluded from further analysis patients who did not undergo pre-operative endometrial sampling.

Clinicopathological variables (patient age, tumor stage, histological subtype, tumor size, surgery type, administration of adjuvant therapy) and data on recurrence were collected and retrospectively analyzed after obtaining approval from this insti-
Table 1. — Clinicopathological variables (n = 12).

| Variable                          | Value       |
|----------------------------------|-------------|
| Patient age, years               | 63 (35-83)* |
| Observation period, months       | 25 (18-90)* |
| Tumor size, cm                   | 13 (9-30)*  |
| Tumor stage                      |             |
| IB                               | 7           |
| IIB                              | 2           |
| IIIIB                             | 1           |
| IVB                              | 2           |
| Histological subtype             |             |
| Ordinary                         | 9           |
| Myxoid                           | 2           |
| Epithelioid                      | 1           |
| Surgery type                     |             |
| TAH + BSO                        | 8           |
| TAH + BSO + tumor resection      | 4           |
| Postoperative adjuvant chemotherapy |         |
| Yes                              | 10          |
| No                               | 2           |
|Disposition                       |             |
| NED                              | 5           |
| AWD                              | 4           |
| DOD                              | 3           |

*Values are presented as median (range).

TAH: total abdominal hysterectomy; BSO: bilateral salpingo-oophorectomy; NED: no evidence of disease; AWD: alive with disease; DOD: dead of disease.

Endometrial cytological specimens were collected preoperatively using the endocyte scratch method. Alcohol-fixed- and air-dried smears were prepared for conventional Papanicolaou staining. The authors compared these results of the initial preoperative evaluation with those of the re-examination. Pathological tissue sections were stained with Hematoxylin and Eosin, and the cytological findings were compared with those of the Papanicolaou-stained specimens.

Results

A total of 12 patients were treated for ULS at this institution between January 2008 and June 2017. All patients underwent abdominal hysterectomy and bilateral salpingo-oophorectomy. Four patients also underwent resection of tumor that extended beyond the uterus. The clinicopathological variables of all 12 patients are summarized in Table 1. The median patient age was 63 (range, 35-83) years. The largest group of patients by tumor Stage was IB1, with seven patients; the largest group by histological subtype was ordinary type, with nine patients. Ten patients received postoperative adjuvant chemotherapy. The median postoperative follow-up period was 25 (range, 18-90) months. Three patients died of their disease, but five patients remained alive without disease, and four remained alive with disease at the time of this study.

Preoperative endometrial cytology was collected in seven patients (ordinary, n = 4; myxoid, n = 2; epithelioid, n = 1). A sample could not be obtained from the other five patients because of nonvisualization of the cervix or cervical stenosis. As shown in Table 2, only one of the seven patients was diagnosed preoperatively with a malignant neoplasm. In this patient, clusters of rounded tumor cells with enlarged, irregular, hyperchromatic, and sometimes multiple nuclei with conspicuous nucleoli were detected in a necrotic background (Figure 1). Macroscopically, the tumor protruded into the endometrial cavity of the resected uterus.

Upon re-evaluation of the preoperative specimens, atypical cells were detected in four of the six patients who were initially deemed negative for malignancy (Table 2). A monomorphic population of spindle cells with slightly hyperchromatic elongated nuclei was detected in two patients (Figure 2 a, c). Another two patients had rounded tumor cells with a moderate amount of basophilic cytoplasm and round or oval nuclei with minimal atypia (Figure 2 e, g). These atypical cells resembled the tumor cells seen in the pathologic tissue sections of the resected uteri. Macroscopically, the tumors occupied the intramuscular and submucosal areas of the resected uteri. For the other two patients who had negative findings on both preoperative and repeat evaluation, the tumors were located in the intramuscular and subserous area in the resected specimen.

Discussion

It is challenging to reach an accurate preoperative diagnosis of ULS. Because of the low sensitivity of cytologic and pathologic diagnosis, one may presume that sarcomatous lesions are covered with normal endometrium in most patients [7]. However, ULS reportedly has characteristic cytological findings that are useful for diagnosis [8]. Leiomyosarcoma cells exhibit elongated or multinucleated
Re-evaluation of preoperative endometrial smears for the cytodiagnosis of uterine leiomyosarcoma

Giant cells with scanty cytoplasm. The nuclei are elongated, cigar shaped, and slightly hyperchromatic, while the nuclear chromatin is coarse with conspicuous nucleoli [9]. Pappa et al. indicate that the cytological features of ULS are enlarged spindle, polygonal, or giant multinucleated cells, with centrally located nuclei and thin cytoplasm with ill-defined borders [10]. In addition, Ito et al. recommend careful microscopic observation of the entire specimen to avoid misdiagnosis if only a few sarcomatous cells are present [11].

Barbazza et al. performed a preliminary study to assess the feasibility and the diagnostic role of percutaneous fine-needle aspiration cytology (FNAC) in the preoperative evaluation of uterine smooth muscle tumors, including ULS [12]. Samples with ULS had a large number of single cells and fragments of loosely arranged tapering cells with nuclear enlargement, crowding, and ill-defined, and scanty cytoplasm. The authors claim that FNAC appears to be a feasible diagnostic procedure for ULS, with cellularity, cohesiveness, cellular atypia, necrosis, and hemorrhage apparently the most important diagnostic parameters in distinguishing between a benign neoplasm and ULS.

Transcervical needle biopsy and hysteroscopic excisional biopsy have been employed to increase the diagnostic sensitivity for ULS [13, 14]. Both methods are reportedly reliable methods for the early diagnosis and treatment of ULS. Significant advances in molecular biology and genetic research have allowed better identification of the molecular signatures of ULS and may help to establish optimal diagnostic strategies for this disease in the future [15].

Since ULS often forms a mass in the muscular layer, it is difficult to detect tumor cells on endometrial cytology. In this review, only a single patient was diagnosed with malignancy during the preoperative period; this patient had tumor cells protruding into the endometrial cavity that were caught on preoperative sampling. On re-evaluation, the authors detected atypical cells, including spindle-shaped cells, in patient samples that were initially diagnosed as negative for malignancy. Because the tumor was located in the submucosal area in these patients, the atypical cells were present in the cytological specimens. These atypical cells resembled the tumor cells detected in the pathologic tissue sections of the resected uteri, but ULS could not be diagnosed by the preoperative cytologic findings alone.

Novel diagnostic techniques are needed to accurately identify ULS in the preoperative period. Careful microscopic observation of the entire cytological specimen, combined with detailed patient medical information, is essential in making a correct diagnosis of ULS.

Table 2. — Cytologic re-examination of preoperative endometrial specimens.

| Patient | Pathologic subtype | Tumor size (cm) | Location              | Preoperative diagnosis         | Re-evaluation results          |
|---------|-------------------|-----------------|-----------------------|--------------------------------|--------------------------------|
| 1       | Ordinary          | 10              | Protruding into endometrial cavity | Malignant neoplasm            | Malignant neoplasm            |
| 2       | Ordinary          | 20              | Intramuscular to submucosal | Negative                      | Atypical spindle cells         |
| 3       | Myxoid            | 17              | Intramuscular to submucosal | Negative                      | Atypical spindle cells         |
| 4       | Ordinary          | 8               | Intramuscular to submucosal | Negative                      | Atypical rounded cells         |
| 5       | Myxoid            | 13              | Intramuscular to submucosal | Negative                      | Atypical rounded cells         |
| 6       | Epithelioid       | 19              | Subserous             | Negative                      | Negative                      |
| 7       | Ordinary          | 11              | Intramuscular to submucosal | Negative                      | Negative                      |

Figure 2. — Atypical cells’ detected on re-evaluation.
(a), (c), (e) and (g) Papanicolaou smear (magnification ×40): (a) patient 2, (c) patient 3, (e) patient 4, and (g) patient 5.
(b), (d), (f), and (h) Hematoxylin and Eosin staining (magnification ×40): (b) patient 2, (d) patient 3, (f) patient 4, and (h) patient 5.
Acknowledgements
This study was supported in part by a grant-in-aid for scientific research from the Japan Society for the Promotion of Science (number 17K11281).

References
[1] D’Angelo E., Prat J.: “Uterine sarcomas: a review”. Gynecol. Oncol., 2010, 116, 131.
[2] Brown L.: “Pathology of uterine malignancies”. Clin. Oncol. (R. Coll. Radiol.), 2008, 20, 433.
[3] Kapp D.S., Shin J.Y., Chan J.K.: “Prognostic factors and survival in 1396 patients with uterine leiomyosarcomas: emphasis on impact of lymphadenectomy and oophorectomy”. Cancer, 2008, 112, 820.
[4] Goto A., Takeuchi S., Sugimura K., Maruo T.: “Usefulness of Gd-DTPA contrast-enhanced dynamic MRI and serum determination of LDH and its isozymes in the differential diagnosis of leiomyosarcoma from degenerated leiomyoma of the uterus”. Int. J. Gynecol. Cancer, 2002, 12, 354.
[5] Bansal N., Herzog T.J., Burke W., Cohen C.J., Wright J.D.: “The utility of preoperative endometrial sampling for the detection of uterine sarcomas”. Gynecol. Oncol., 2008, 110, 43.
[6] Ueoka Y., Kobayashi H., Wake N.: “Carcinoma of the endometrium surveillance counterpoint: Japan”. In: Johnson F.E., Maehara Y., Brownman G.P., Margenthaler J.A., Audisio R.A., Thompson J.F., et al. (eds). Patient surveillance after cancer treatment. New York: Humana Press, 2013, 311.
[7] Oda K., Okada S., Nei T., Shirai T., Takahashi M., Sano Y., et al.: “Cytodiagnostic problems in uterine sarcoma. Analysis according to a novel classification of tumor growth types”. Acta. Cytol., 2004, 48, 181.
[8] An-Foraker S.H., Kawada C.Y.: “Cytodiagnosis of endometrial malignant mixed mesodermal tumor”. Acta. Cytol., 1985, 29, 137.
[9] Massoni E.A., Hajdu S.I.: “Cytology of primary and metastatic uterine sarcomas”. Acta. Cytol., 1984, 28, 93.
[10] Pappa L., Zagorianakou N., Kitsiou E., Sintou-Mantela E., Bafa M., Malamnou-Mitsi V.: “Breast metastasis from uterine leiomyosarcoma diagnosed by fine needle aspiration: a case report”. Acta. Cytol., 2008, 52, 485.
[11] Ito E., Saito T., Suzuki T., Fujii M., Kudo R.: “Cytology of vaginal and uterine sarcomas”. Acta. Cytol., 2004, 48, 601.
[12] Barbazza R., Chiarelli S., Quintarelli G.F., Manconi R.: “Role of fine-needle aspiration cytology in the preoperative evaluation of smooth muscle tumors”. Diagn. Cytopathol., 1997, 16, 326.
[13] Kawamura N., Ichinuma T., Ito F., Shibata S., Takahashi K., Tsujimura A., et al.: “Transcervical needle biopsy for the differential diagnosis between uterine sarcoma and leiomyoma”. Cancer, 2002, 94, 1713.
[14] Shveiky D., Revel A., Rojansky N., Benshushan A., Shushan A.: “Diagnosis of malignant mesenchymal uterine tumors by hysteroscopic excisional biopsy”. J. Minim. Invasive. Gynecol., 2005, 12, 29.
[15] Sonoda K.: “Uterine leiomyosarcoma: An aggressive and challenging malignant tumor”. Fukuoka. Acta. Med., 2017, 108, 24.

Corresponding Author:
K. SONODA, M.D., Ph.D.
Gynecologic Service
National Kyushu Cancer Center
Notame 3-1-1
Minami-ku, Fukuoka 811-1395 (Japan)
e-mail: sonoda.k@nk-cc.go.jp