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A Case of Endometrioid Adenocarcinoma Resembling Sex Cord–Stromal Tumor of the Ovary with Concomitant Stromal Hyperthecosis

Mikiko Aoki[1], Sumie Nakamura[2], Kyoko Shirota[2], Hiroshi Iwasaki[1], Tatsuhiko Kawarabayashi[2], Shingo Miyamoto[2] and Kazuki Nabeshima[1]

[1] *Department of Pathology, Faculty of Medicine, Fukuoka University*
[2] *Department of Obstetrics and Gynecology, Faculty of Medicine, Fukuoka University*

Abstract：A 55－year－old Japanese woman with irregular vaginal bleeding underwent a right salpingo－oophrectomy for right ovarian tumor. Preoperative laboratory tests were normal except for elevated serum estradiol level (120 pg/ml), which decreased after the surgery. Intraoperative frozen sections suggested granulosa cell tumor based on presence of lobulated nests of tumor cells and small follicular structures, together with Call－Exner－like bodies. Postoperative examination of formalin－fixed paraffin－embedded sections showed two different－looking portions, solid and tubular portions, divided by thin fibrous septa. The former was sex cord－stromal tumor－like, and the latter endometrioid adenocarcinoma－like. However, tumor cells of both portions were positive for epithelial membrane antigen, cytokeratins, vimentin, estrogen receptor, and progesterone receptor, but negative for α－inhibin, calretinin, CD56 and cytokeratin 20, indicating endometrioid adenocarcinoma resembling sex cord－stromal tumor. This variant of endometrioid adenocarcinoma is uncommon, and was accompanied by stromal hyperthecosis in our case, which was responsible for the high serum estradiol level and irregular vaginal bleeding. Diagnosis in frozen sections is important, but accurate diagnosis is sometimes difficult and needs awareness of the tumor, together with multiple sampling and use of intraoperative cytologic preparations. A combination of this variant and stromal hyperthecosis with estrogenic manifestations should be considered for the differential diagnosis.

Key words：**Ovary, Endometrioid adenocarcinoma, Sex cord–stromal tumor, Granulosa cell tumor, Hyperthecosis**

Introduction

Endometrioid ovarian carcinoma is a specific histopathological entity that accounts for stromal tumors, which are composed of granulosa cells, theca cells, Sertoli cells, Leydig cells and fibroblasts of stromal origin, account for about 8% of all ovarian neoplasms and about 1－3 % of borderline malignant ovarian tumors.[1)–3)] Endometrioid adenocarcinoma resembling sex cord－stromal tumor is an uncommon variant of endometrioid carcinomas of the ovary.[4)–6)] This type of endometrioid adenocarcinoma sometimes exhibits a small and tubular or solid and microglandular pattern simulating sex cord－stromal tumors, such as Sertoli, Sertoli－Leydig cell tumor and granulosa cell tumor. To our knowledge, however, only several cases with the granulosa cell tumor－like variant of endometrioid adenocarcinoma have been

Correspondence to：Mikiko Aoki, Department of Pathology, Fukuoka University Faculty of Medicine, 7–45–1 Nanakuma, Jonan–ku, Fukuoka 814–0180, Japan.
Tel :092–801–1011 Fax :092–863–8383 E–mail：mikikoss@fukuoka–u.ac.jp

reported.[4)7)-10)]

Herein, we report a rare and diagnostically difficult case, especially with intraoperative frozen sections, of endometrioid adenocarcinoma resembling sex cord-stromal tumor associated with stromal hyperthecosis.

Clinical Summary

A 55-year old Japanese woman who had menopause at 50 years of age visited the hospital for vague complaints of itching paresthesia around the genitalia. She received hormone replacement therapy to control the post-menopause symptoms. About three months later, irregular vaginal bleeding appeared. Despite change in medication, bleeding continued, and therefore medication was stopped. She had received fertility treatment when she was 28 years old and undergone left oophorectomy for endometriosis 30 years earlier. Physical examination showed she was in good condition and laboratory tests were within the normal range except for elevated serum estradiol level (120 pg/ml). The right ovary was enlarged to 45 mm in diameter, as detected on magnetic resonance imaging (MRI) (Fig. 1). She underwent total hysterectomy, right salpingo-oophorectomy, and periaortic lymph node sampling. A 5.0 cm solid tumor was observed in the right ovary. The uterus and endometrium were grossly unremarkable. She remains alive without evidence of disease at two years after operation.

Pathological Findings

Macroscopically, the right ovarian tumor was well circumscribed. The cut surface was pale yellowish and divided by thin fibrous tissue. Neither hemorrhage nor necrosis was found (Fig. 2a). Microscopically, intraoperative frozen sections showed proliferating tumor cells containing oval or slightly elongated nuclei arranged in lobulated nests, accompanied by small follicular structures (Fig. 2b). Call-Exner-like bodies were also seen (Fig. 2c). In paraffin sections, the tumor consisted of two different-looking portions, divided by thin fibrous septa (Fig. 2d). One was a solid portion, where darkly-stained cells with oval nuclei and scant cytoplasm formed compact anastomosing cords, tightly packed nests and microglandular areas, accompanied by prominent fibrocellular stroma, mimicking adult-type granulosa cell tumor, but true Call Exner bodies were absent (Fig. 2e, f). Nuclear grooves were also occasion-

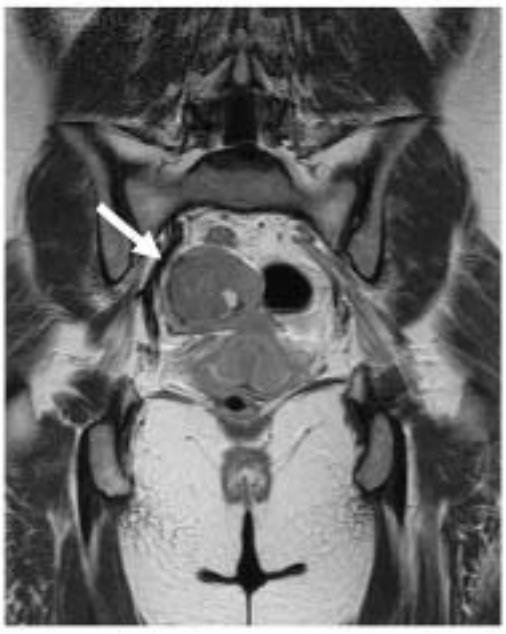

Figure 1. Magnetic resonance imaging of the right ovarian tumor (arrow) shows slightly low T1 signal intensity and a focal high signal intensity area, which indicates hemorrhage.

ally seen in this portion (Fig. 2f, arrows). The other was the tubular portion, where atypical cells (with large vesicular or hyperchromatic nuclei, prominent nucleoli and relatively abundant cytoplasm) proliferated, forming fused glandular structures, indicative of conventional endometrioid adenocarcinoma (Fig. 2g). This tubular portion was found only in a small part (Fig. 2a, dotted area), and differentiation between the solid and tubular portions was difficult on gross examination of the cut surface. The tumor cells of both portions contained Alcian blue-positive acid mucin in lumina (Fig. 3a, b). Small nests or scattered luteinized or clear stromal cells were observed in the tumor stroma (Fig. 2h). Endometriosis, which consisted of non-neoplastic endometrial glands, stromal cells and hemosiderin-laden macrophages, was also noted in some parts of the ovary (data not shown).

Immunohistochemistry

Immunohistochemistry was carried out using primary antibodies against epithelial membrane antigen (EMA) (E29, Dako, Carpentaria, CA), cytokeratin AE1/AE3 (Dako), vimentin (V9, Dako), estrogen receptor (1D5, Dako), progesterone receptor (PgR636, Dako), cytokeratin 7 (OV-TL12/

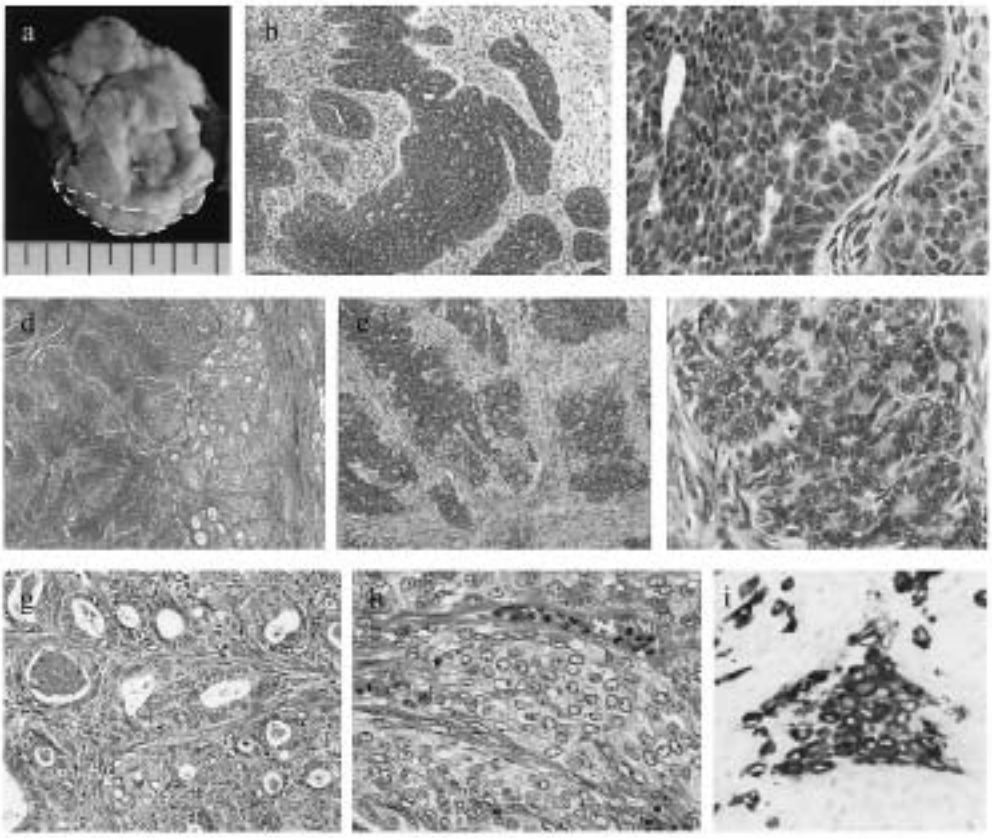

Figure 2. Pathological findings of the right ovarian tumor. a, Macroscopic appearance of the tumor (a cut section after formalin fixation). The tumor is solid and pale yellowish. Neither hemorrhage nor necrosis is observed. The area indicated by broken lines represents the endometrioid adenocarcinoma portion. b, Intraoperative frozen section shows lobulated nests accompanied by fibrothecomatous stroma. Note also the follicular or luminal structures. c, Call-Exner body-like structures. d, Permanent section shows two types textures, solid and tubular portions divided by thin fibrous septa (broken line). e, f, The solid portion shows sex cord-stromal tumor-like structures with occasional nuclear grooves (f, arrows). g, The tubular portion shows ordinary endometrioid adenocarcinoma. h, The luteinized cells have abundant cytoplasm and regular, round nuclei with prominent nucleoli. i, The luteinized cells are immunohistochemically positive for α-inhibin.

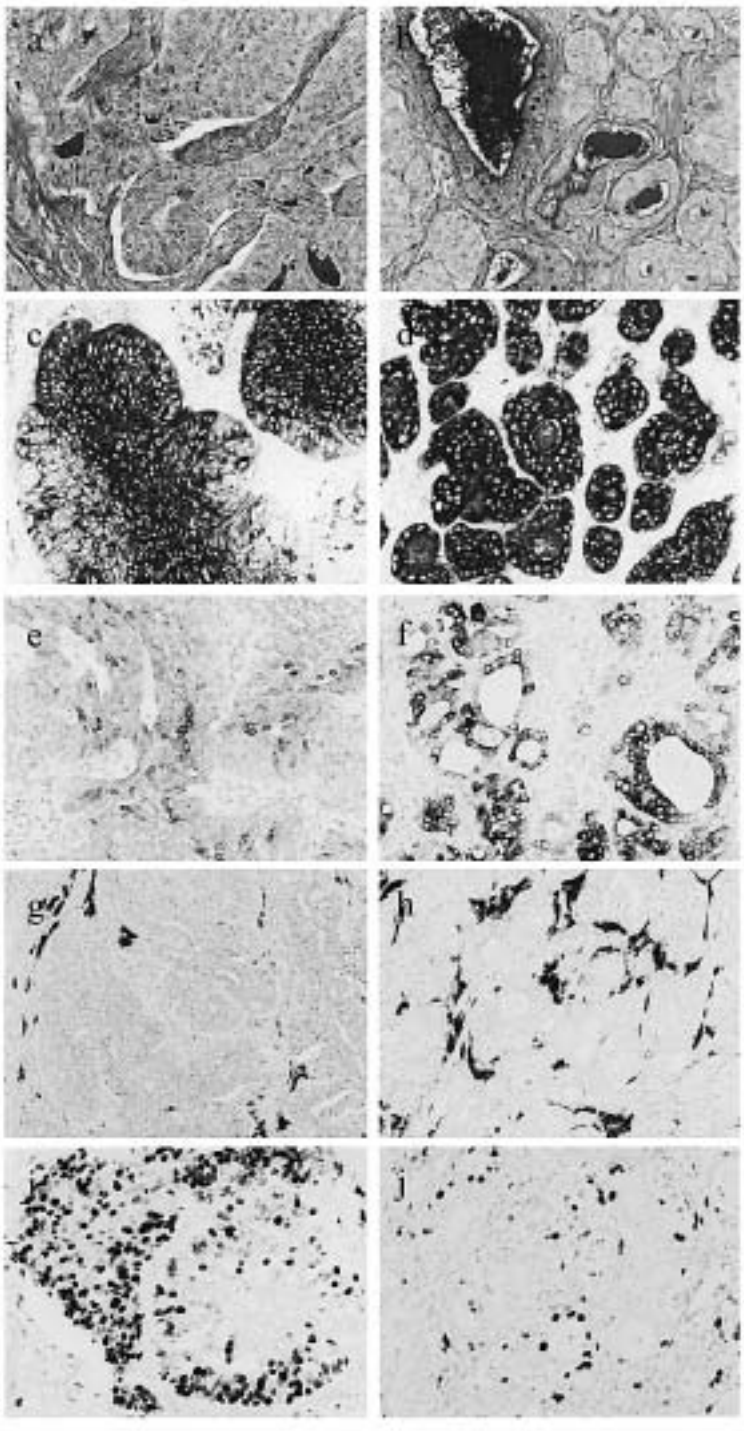

Figure 3. Immunohistochemistory and special staining. (a, b) Alcian－blue positive mucinous secretions in lumina. (c, d) EMA expression. (e, f) CK7 expression is focal in the solid portion, while it is diffuse in the tubular portion. (g, h) Expression of α－inhibin in the tumor stroma. Tumor cells in both the solid and tubular portions are negative. (i, j) MIB－1 expression. a, b, Alcian blue/PAS double stain ; c－j, immunohistochemistry ; a, c, e, g, i, the sex cord－stromal tumor－like solid portion ; b, d, f, h, j, the ordinary endometrioid adenocarcinoma－like tubular portion ; EMA, epithelial membrane antigen ; CK, cytokeratin.

30, Dako), α-inhibin (R1, Dako), calretinin (Z11-E3, Zymed, San Francisco, CA), S-100 protein (Zymed), α-smooth muscle actin (α-SMA) (1A4, Dako), chromogranin A (DAK-A3, Dako), CD56 (1B6, Novocastra, Newcastle, UK), cytokeratin 20 (ks20.8, Dako), and MIB-1 (Dako). The tumor cells of both solid and tubular portions were diffusely positive for EMA (Fig. 3c, d), cytokeratin AE1/AE3, vimentin, estrogen receptor, and progesterone receptor. Cytokeration 7-positive cells were more frequently seen in tubular portions and focally in solid portions (Fig. 3e, f). The tumor cells of both portions were negative for α-inhibin (Fig. 3g, h), calretinin, S-100 protein, α-SMA, chromogranin A, CD56 and cytokeratin 20. MIB-1 labeling index (LI) was about 50% in the solid portion (Fig. 3i) and 10% in the tubular portion (Fig. 3j). Luteinized stromal cells were positive for α-inhibin (Fig. 2i) and calretinin. Based on the hematoxylin-eosin and immunohistochemical findings, the tumor was diagnosed as endometrioid adenocarcinoma resembling sex cord-stromal tumor, associated with stromal hyperthecosis.

Discussion

Endometrioid adenocarcinoma resembling sex cord-stromal tumor is a rare variant of endometrioid adenocarcinoma that focally looks like sex cord-stromal tumor containing Sertoli, Leydig or granulosa cells. Differential diagnosis between endometrioid adenocarcinoma resembling sex cord-stromal tumor and real sex cord-stromal tumors is sometimes difficult. Features that favor a diagnosis of endometrioid adenocarcinoma are elderly patients, the usual absence of endocrine manifestations, and the occurrence elsewhere in the tumor of usual pattern of endometrioid adenocarcinoma, foci of squamous metaplasia, luminal mucin accumulation, adenofibromatous components, immunoreactivity for EMA, and keratin (including CK7), and negativity for α-inhibin, calretinin, and CD56.[7)8)11)-15)] Among these, focal occurrence of ordinary pattern of endometrioid adenocarcinoma was the most helpful clue to the accurate diagnosis in paraffin sections in our case. Furthermore, the same immunohistochemical phenotypes in both endometrioid adenocarcinoma portions (tubular portions) and sex cord-stromal tumor-like portions (solid portions) strongly supported the diagnosis. However, there was difficulty in frozen section diagnosis. Frozen section diagnosis is generally a reliable method for the surgical management of patients with ovarian masses and its accuracy is estimated between 91% and 97%.[16)17)] In the present case, however, diagnosis was not easy using frozen sections because 1) frozen sections included only solid portions of the tumor ;2) cellular characteristics, especially those of tumor cell nuclei, were difficult to observe in frozen sections ;3) the laboratory data showed elevated serum estradiol level. For the first reason, as shown in Fig. 2a, the purely tubular portion(endometrioid adenocarcinoma portions) of the tumor was present only in a small part, and both solid and tubular portions looked similar grossly on the cut surface. This resulted in absence of the tubular portion in frozen sections. Multiple sampling may favor accurate diagnosis of this variant. With regard to the second reason, it is known that nuclei of endometrioid adenocarcinoma cells are round and hyperchromatic, whereas those of granulosa cell tumor are characteristically round, oval or angular, pale, and often grooved.[3)] These features are generally difficult to observe in frozen sections, and the use of intraoperative touch preparations from the cut surface of the tumor may help in this differential diagnosis. With respect to the third, our case showed concomitant presence of endometrioid adenocarcinoma resembling sex cord-stromal tumor and stromal hyperthecosis with a consequent high estradiol level, which is a rare accompaniment. To our knowledge, estrogenic manifestations (as an actively hyperplastic endometrium) were reported in only one case in a study by Young et al.[4)] with 13 ovarian endometrioid carcinomas resembling sex cord-stromal tumors, although they described the presence of luteinized stromal cells in four cases. Focal, luteinized stromal cells were also found in 10 of 13 cases with sertoliform endometrioid carcinomas, but with signs of virilization and elevated serum testosterone level in one case only.[6)] The preoperative high estradiol level accompanied by irregular genital bleeding, as in our case, generally favors sex cord-stromal tumors. However, we should

hereafter be aware of a combination of a non-hormone-producing tumor and stromal hyperthecosis. Moreover, for differential diagnosis in frozen sections, Alcian-blue and/or PAS stains may be an additional help; detection of luminal mucinous secretions makes us suspicious of adenocarcinoma, although these stains need longer time to perform.

Recognition of the sex cord-stromal tumor-like portions is also important for grading of endometrioid adenocarcinoma. In our case, if the sex cord-stromal tumor-like portions are considered as just solid portions of endometrioid adenocarcinoma, the case can be diagnosed as grade 3 endometrioid adenocarcinoma according to the International Federation of Obstetrics and Gynecologists (FIGO) grading system.[18] However, it is suggested that sertoliform endometrioid adenocarcinoma should be considered as well-differentiated carcinoma (grade 1), despite the presence of a solid, sex cord-like proliferation, because it has a relatively good prognosis when limited to the ovary.[6)13)] According to these suggestions, it may be better to diagnose our case as grade 1 endometrioid adenocarcinoma based on the histological features in the purely endometrioid portion since the carcinoma was limited to the ovary. However, we are afraid that these suggestions cannot be simply applied to our case because of the higher MIB-1 LI and more mitotic figures in the granulosa cell tumor-like solid portions compared to those in the purely endometrioid adenocarcinoma portion (50% vs 10% and 12 mf/30 hpf vs 2 mf/30 hpf, respectively). Although high Ki67/MIB-1 LI and mitotic count did not have any impact on recurrence or on disease-related death in adult-type granulosa cell tumors of the ovary,[19] high Ki67 LI (>35%) had independent prognostic influence in endometrial carcinomas of endometrioid type.[20] Prognostically significant information was provided by mitotic count also in ovarian carcinomas including endometrioid adenocarcinomas.[21] Thus, careful follow-up is needed in our case.

Estradiol-producing tumors include granulosa cell tumor, thecoma, and Sertoli-Leydig cell tumor, and endometrioid adenocarcinoma is not a hormone-producing tumor. In our case, the high estradiol level decreased to the normal level after removal of the ovarian tumor because of the presence of stromal hyperthecosis in the tumor stroma. Stromal hyperthecosis is a disorder in which luteinized cells are scattered singly and in small nests or nodules throughout a typically hyperplastic ovarian stroma.[3] The luteinized stromal cells (stromal hyperthecosis) exhibit positive staining for α-inhibin and calretinin.[22)-24)] When hormonal manifestations are present in postmenopausal patients, they are usually estrogenic, and the estrogenic effects are mediated through peripheral aromatization of androgens produced by luteinized stromal cells in hyperthecosis.[25)26)]

In conclusion, diagnosis of carcinomas in frozen sections is important, but accurate diagnosis is sometimes difficult especially in their rare variants and needs awareness of the variants together with multiple sampling and use of cytologic preparations. Co-presence of hyperthecosis should be considered also as a cause of estrogenic manifestations in postmenopausal patients.

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