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Original contribution

High frequency of *POLE* mutations in synchronous endometrial and ovarian carcinoma

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Summary Synchronous endometrial and ovarian carcinomas) represent 5% to 10% of endometrial or ovarian carcinomas. We assessed genetic alterations (in *PTEN*, *CTNNB1*, *POLE*, etc) and evaluated correlations with patient outcomes to determine the utility of clonality analyses for differentiating between metastases and concurrent primary tumors and for determining whether genetic alterations in synchronous tumors are predictive of biological behavior. Genomic DNA was isolated from formalin-fixed, paraffin-embedded tissues and frozen tissues from patients with synchronous endometrial and ovarian carcinomas. Samples were obtained from the Department of Obstetrics and Gynecology at the Shimane University School of Medicine between 2003 and 2017. Sanger sequencing was used to analyze the mutational status of the coding exons in *TP53*, *PTEN*, *POLE*, *PIK3CA*, *KRAS*, and *CTNNB1* using previously published primers. All patients lived, and 3 had disease recurrence. The frequencies of somatic mutations in *TP53*, *PTEN*, *CTNNB1*, *KRAS*, and *POLE* were 3 (37.5%), 2 (25.0%), 3 (37.5%), 0 (0.0%), and 5 (62.5%) of 8 cases in ovarian tumors and 3 (37.5%), 2 (25.0%), 3 (37.5%), 1 (12.5%), and 5 (62.5%) of 8 cases in endometrial tumors, respectively. The frequencies of *POLE* and *CTNNB1* mutations were higher than those in previous reports. A clonal relationship was determined by genomic analyses in 3 of 6 cases that were initially diagnosed as primary independent tumors. We confirmed that these 3 cases were indicated metastatic tumors because the lesion of mutation was the same. This information, provided by the sequencing-based strategy, could be useful for hypothesizing a patient's prognosis and deciding on treatment.

1. Introduction

Synchronous endometrial and ovarian carcinomas (SEOs) have been reported in 5% to 10% of endometrial or ovarian carcinoma [1,2]. Whether these SEOs are 2 independent

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primary tumors or metastatic disease has critical implications for prognosis and patient management [3-5]. Typically, patients with SEOs (endometrioid/endometrioid) have a good prognosis, even if they are in an advanced stage, and clinical trials have been performed to evaluate treatment with adjuvant therapy; however, the reasons for a good prognosis have not been established.

Several recent studies have identified synchronous tumors that exhibit clonality by deep sequencing and parallel sequencing approaches [6-8]. Endometrioid ovarian and endometrioid endometrial tumors also share some molecular features, including microsatellite instability, *TP53* loss, and nuclear β-catenin expression (indicative of *CTNNB1* activating mutations). In this study, mutations in various genes (eg, *POLE*, *PTEN*, *KRAS*, and *PIK3CA*) were evaluated in patients with SEOs to determine the association between mutations and patient survival and to characterize tumor profiles using a molecular approach. In particular, the objectives of the current study were to determine the clonal relationship between simultaneously diagnosed endometrial and ovarian carcinomas to identify primary or metastatic carcinomas.

Table 1 Scully's classification [9]

Independent primary tumors
1. Histologic dissimilarity of tumors
2. No or only superficial myometrial invasion of endometrial tumor
3. No vascular space invasion of endometrial tumor
4. Atypical endometrial hyperplasia additionally present
5. Absence of other evidence of spread of endometrial tumor
6. Ovarian tumor unilateral (80%–90% of cases)
7. Ovarian tumors located mainly in parenchyma
8. No vascular space invasion, surface implants, or predominant hilar location in the ovary
9. Absence of other evidence of spread of ovarian tumor
10. Ovary endometriosis present

2. Materials and methods

2.1. Tissue samples

Nine patients diagnosed as having SEOs receiving surgery and systematic chemotherapy were included in the study. Diagnoses were confirmed by a pathologist at our institution. He was trained as a gynecopathologist at our institution. All patients were primarily treated with cytoreductive surgery and adjuvant platinum and taxane chemotherapy (carboplatin CBDCA ; [AUC5], paclitaxel PTX ; 175 mg/m^2, or docetaxel DTX ; 70 mg/m^2). All patients received 6 to 12 courses of this regimen. When patients had disease recurrence, the second-line chemotherapy regimen included doxorubicin hydrochloride (pegylated liposomal PLD;) and carboplatin for patients with platinum-sensitive ovarian carcinoma and gemcitabine hydrochloride GEM;, doxorubicin hydrochloride (pegylated liposomal), topotecan hydrochloride TPT ; alone, and bevacizumab and BEV ; for patients with platinum-resistant carcinoma.

Tissue samples were obtained from the Department of Obstetrics and Gynecology at the Shimane University School of Medicine between 2003 and 2017. The acquisition of tumor tissues was approved by the Shimane University Institutional Review Board (No. 20070305-1). The diagnoses were confirmed by a surgical pathologist before the tumor samples were harvested for experiments. After appropriate explanation, all patients provided written informed consent for the procedure and for participation in the study.

Of 9 cases, 8 cases were diagnosed as dual primary tumors by Scully's classification (Table 1) [9]. Six pairs of SEOs classified as endometrioid type and 2 pairs of SEOs with different histologic types were obtained. Only 1 case was first diagnosed as a metastatic tumor.

2.2. DNA isolation

Tumor DNA was extracted from formalin-fixed, paraffin-embedded tissues according to protocols for the isolation of total DNA. Briefly, the cell pellet was obtained by centrifugation for 5 minutes at 300*g*. Samples were lysed by adding 20 μL of proteinase K and 0.2 mL of buffer AL and incubated at 56°C for 10 minutes. Then, 0.2 mL of 96% to 100% ethanol was added for precipitation. The sample mixture was loaded onto the DNeasy Mini Spin Column, QIAGEN, GERMANY. After 2 wash steps, the DNA solution was eluted and the aliquot was used for further analysis. Matched normal DNA was extracted from the nonneoplastic myometrium or peripheral blood.

2.3. Mutational analysis of ovarian carcinoma tissues by Sanger sequencing

Nucleotide sequencing was used to analyze the mutational status of *TP53*, *PTEN*, *CTNNB1*, and *POLE* in tumor cells isolated from ovarian and endometrial carcinomas. The analysis focused on exons that have been reported to harbor most mutations in each gene. The primer sequences and the polymerase chain reaction (PCR) protocol have been described in previous reports [10-18].

DNA was extracted and amplified by PCR using primers for exon 2 of *KRAS*, exons 1 to 9 of *PTEN*, exons 9 and 20 of *PIK3CA*, exons 1 to 9 of *TP53,* exons 9 to 14 of *POLE,* and exon 3 of *CTNNB1*. Primers are shown in Supplementary Table 1.

All mutations identified in tumors were confirmed by independent PCR and Sanger sequencing in the specific tumors

Table 2 Clinicopathological factors of SEO tumors firstly diagnosed as independent tumors

Case	Patient age (y)	DSH	First diagnosis	Other malignant disease	Family history of malignant disease	Surgery	Adjuvant therapy	Patient outcome
1	53	OC T3c endometrioid EC T1b endometrioid	Independent	None	None	mRH-BSO, PLND, PAND, omentectomy, appendectomy	Chemotherapy (TC3 + CAP6) Radiation (WP + PAN)	NED 15 y 8 mo
2	65	OC T1C endometrioid EC T1a endometrioid		Gastric ca. Colon ca.	Father: renal ca. Mother: gastoric ca.	STH-BSO, gastrectomy	Chemotherapy (TC6)	AWD 5y2m
3	40	OC T1c endometrioid EC T1a endometrioid		None	Father: gastoric ca. Mother: gastoric ca. Brother: gastoric ca.	STH-BSO, PLND, PAND, omentectomy	Chemotherapy (TC6)	NED 8 y 1 mo
4	62	OC T1a endometrioid EC T1a endometrioid		None	Father: pancreas ca. Mother: gastoric ca.	STH-BSO	Chemotherapy (TC6)	NED 1 y 5 mo
5	51	OC T1b endometrioid EC T2 endometrioid		None	None	mRH-BSO, PLND, PAND	Chemotherapy (TC6)	NED 1y 4 mo
6	59	OC T3b endometrioid EC T1b endometrioid		None	None	STH-BSO, omentectomy	Chemotherapy (TC6 + BEV → BEV maintenance)	NED 5 mo
7	49	OC T3c serous EC T1b endometrioid		None	Mother: lung ca. Sister: endometrial ca. Sister: breast ca.	STH-BSO, omentectomy → IDS	Chemotherapy (TC4) → (IDS) → (DC8)	AWD 4 y 9 mo
8	53	OC T1a mucinous EC T1b endometrioid		Lung ca.	None	STH-BSO, PLND, PAND	Chemotherapy (TC6)	NED 12 y 8 mo
9	72	EC 4B serous	Metastatic	None	None	STH-BSO, omentectomy → IDS	Chemotherapy (TC4) → (IDS) → (DC8)	AWD 2 y 9 mo

Abbreviations: AED, alive with disease; BEV, bevacizumab; BSO, bilateral salpingo-oophorectomy; ca., cancer; CAP, cyclophosphamide + adriamycin + cisplatin; EC, endometrial carcinoma; DSH, disease, stage, histology; IDS, interval debulking surgery; mRH, modified radical hysterectomy; NED, no evidence of disease; OC, ovarian carcinoma; PAND, para-aortic lymphadenectomy; PLND, pelvic lymphadenectomy; STH, simple transabdominal hysterectomy; TC, paclitaxel + carboplatin; WP, whole pelvis; PAN, paraaortic lymphnode.

and paired normal tissues to determine their somatic nature. Sequencing was performed using the ABI BigDye Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems, Foster City, CA). A sequencing analysis was performed to detect mutations in benign tissues, such as blood, from each patient. It was necessary to determine whether the same mutations occurred in tumor tissues and benign tissues to ascertain germline mutations.

3. Results

3.1. Clinical and pathological features

The clinical and histologic features of 9 SEOs are described in Table 2 and Fig. 1. The median age of the patients was 54 years (range, 40-65 years). A total of 8 cases were clinically diagnosed by the pathologist as dual primary carcinomas according to the Scully's classification [9]. Among these 8 cases, 6 cases were classified as synchronous independent tumors that had the same histology in endometrial and ovarian tumors. There were 2 cases (cases 2 and 3) that were suspected of Lynch syndrome because of their family history and previous disease history (Amsterdam II criteria). There were 3 cases (independent tumor cases 2 and 7 and metastasis tumor case 1) that showed disease recurrence, but all patients received second-line chemotherapy and no patients had died during the study period.

3.2. Identification of *TP53*, *PTEN*, *CTNNB1*, *KRAS*, and *POLE* mutations

We examined 8 cases of synchronous carcinomas of the uterus and ovary to determine whether conventional gross and histologic parameters are correlated with molecular genetic alterations. The mutations were confirmed by Sanger sequencing of DNA from tissues of the ovarian tumor and endometrial tumor in the same patient.

Six cases exhibited the same histologic subtype of endometrioid carcinoma in each of the SEOs. The histologic and mutational statuses of *TP53*, *PTEN*, *CTNNB1*, *KRAS*, and *POLE* in all SEO tumors are summarized in Table 2. Somatic mutations in *POLE* were identified in 6 (75.0%) of 8 patients. *POLE* mutations were detected in 5 (62.5%) of 8 patients in both the ovarian tumor and endometrial tumor. Somatic mutations in *CTNNB1* were also identified in 4 (50.0%) of 8 patients. The frequencies of *POLE* and *CTNNB1* mutations were higher than those reported previously [16-18].

The clonal relationship determined by genomic analyses did not agree with the clinicopathological criteria in 3 of 6 cases. In case 3, there was an A insertion at the same location in exon 3 of *CTNNB1* in both tumors. In case 4, there was a point mutation at the same location in exon 13 of *POLE*. In case 6, there were 2 point mutations at the same location in exon 9 of *POLE* and exon 3 of *CTNNB1* (Table 3, Fig. 2A). In these 3 cases, the results indicated that the lesions are clonally related; thus, we obtained a final diagnosis of metastatic tumors.

There was no mutation at the same location in case 1, 2, or 5 (Table 4).

The mutation status of metastatic tumors determined by pathology also showed 3 point mutations at the same location in exon 4 of *TP53*, exon 1 of *PTEN*, and exon 12 of *POLE* (Table 5, Fig. 2B).

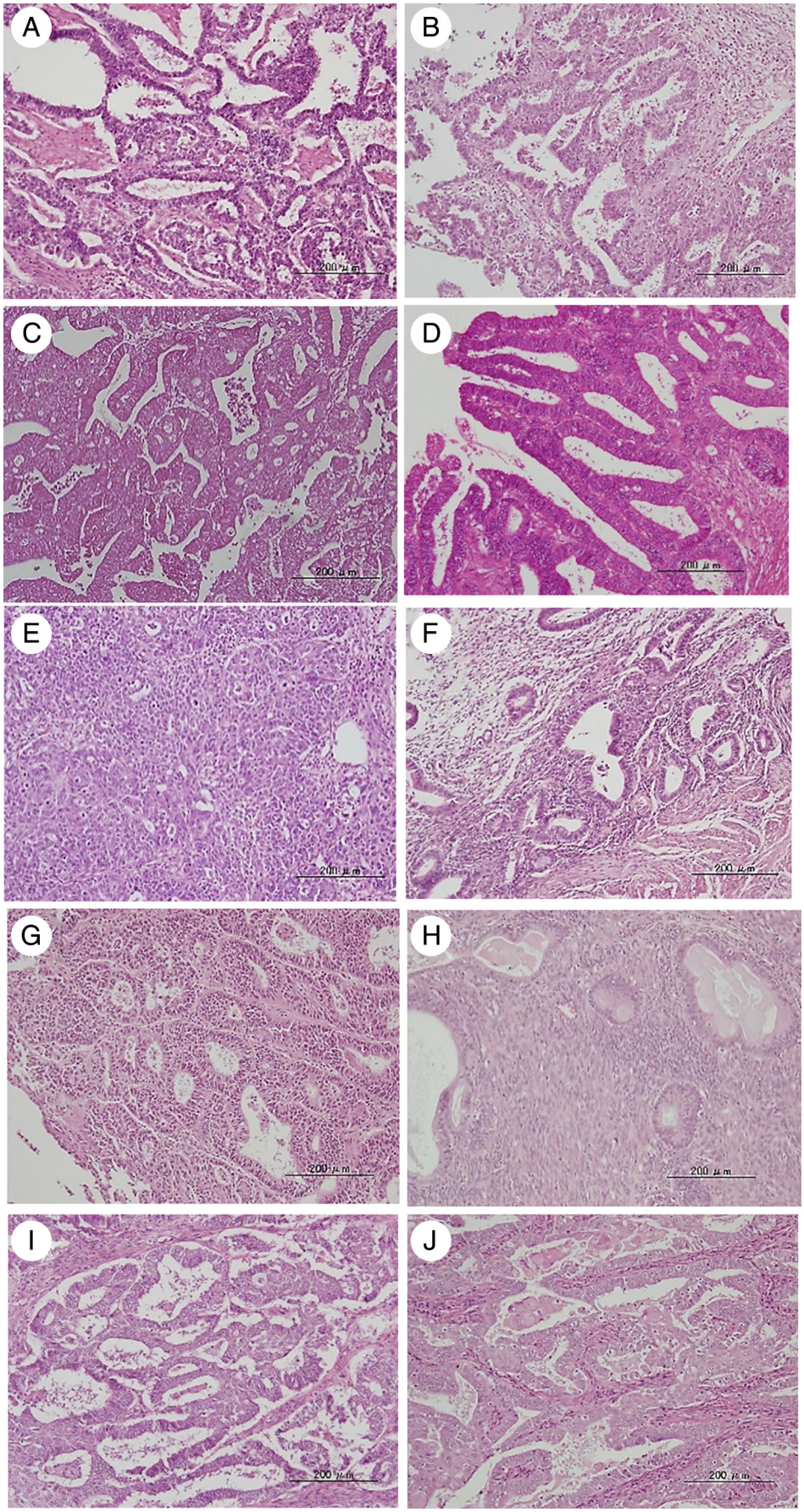
3.3. Clinical features of SEOs with *POLE* mutations

A χ^2 test showed that *POLE* mutations were not related to clinicopathological factors or other gene mutations in SEO tumors (Table 6).

4. Discussion

A clonal relationship between SEOs histologically diagnosed as independent endometrioid endometrial carcinomas and ovarian carcinomas has been reported in recent studies; some of these tumors were finally diagnosed as metastatic tumors by exome sequencing [6-8]. These reports demonstrated that histologic diagnosis alone is not sufficient to determine whether SEOs are primary or metastatic tumors. We also observed 3 cases that were initially diagnosed as independent primary tumors but were actually metastatic tumors based on an examination of gene alterations. In these 3 cases, we assessed gene alterations by Sanger sequencing. We were unable to perform deep sequencing and whole-genome sequence analyses, as in previous reports [6-8]; accordingly, we could not confirm the clonality of the 3 synchronous tumors in detail. In the future, a deep sequencing and whole-genome sequencing approach is preferable; however, our results clearly indicated that the same mutation was present in 2 tumor types within a patient. This evidence for the same mutation in both tumors, indicating that they were metastatic tumors, was obtained by Sanger sequencing, suggesting that this is a low-cost method to confirm the clonality of SEOs.

Finally, all patients with SEOs had a good prognosis, consistent with previous reports [19-22], despite having 2 or more malignant diseases and occasionally having advanced disease. Some previous reports have shown that microsatellite instability is related to a good prognosis [19,20,22]. In the current study, a high frequency of *POLE* mutations was observed; these mutations are widely known as good prognostic factors in endometrial carcinomas [18]. Previous reports have also identified *POLE* mutations in SEOs, but the frequency and the relationship between *POLE* mutations and prognosis were unclear [6]. In the current study, patients with *POLE* mutations did not show significantly longer progression-free survival compared with patients without *POLE* mutations. We investigated the *POLE* mutation that would influence tumor mutation burden, to increase the patient's prognosis.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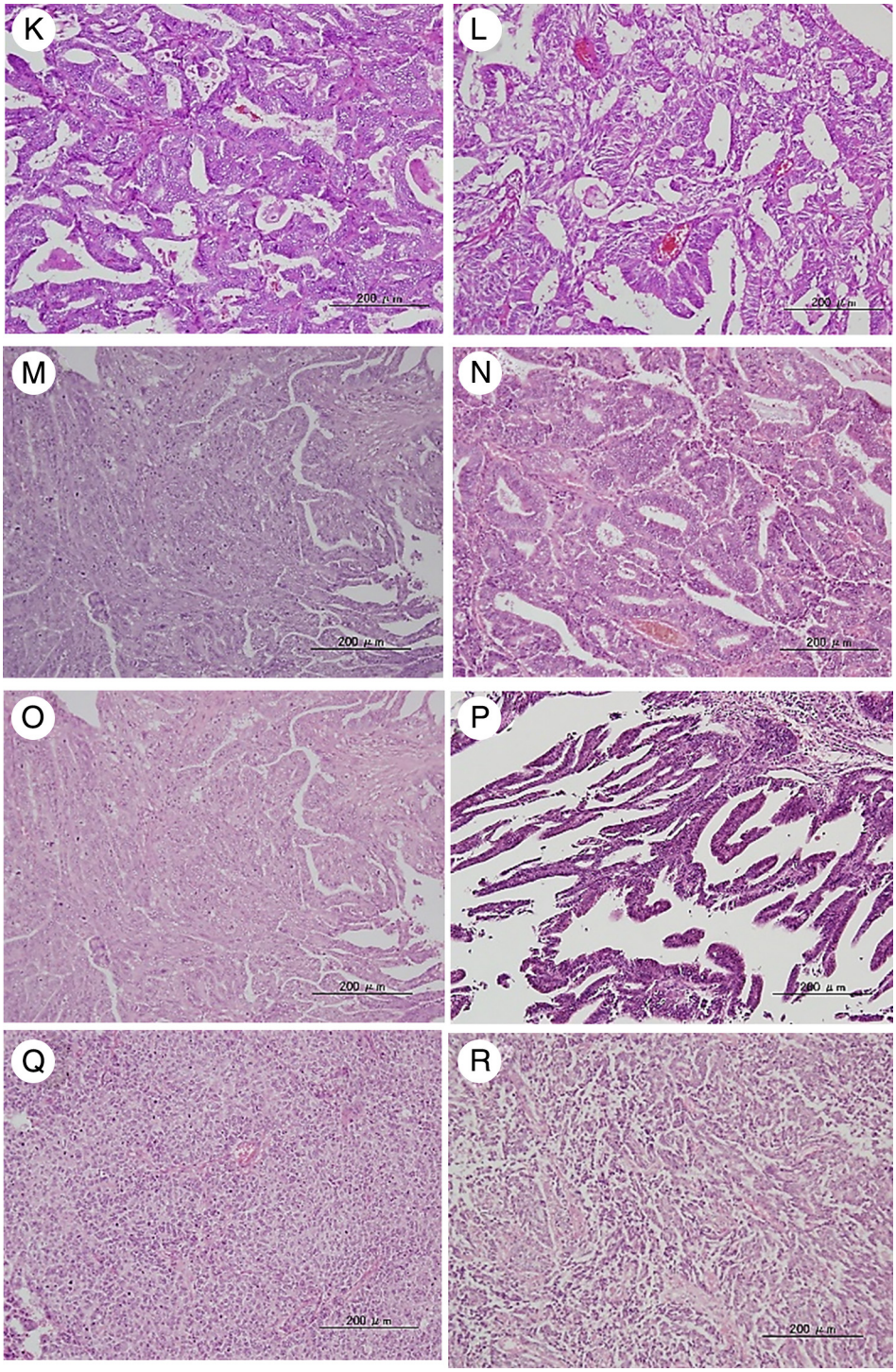

Fig. 1 Hematoxylin and eosin staining of the specimens in patients with SEO tumors. Dual primary cases (AB, CD, IJ) and metastatic cases (EF, GH, KJ, QR). Dual primary cases. Ovarian endometrioid and uterine endometrioid: case 1 (A and B), case 2 (C and D), case 5 (I and J); ovarian serous and uterine endometrioid: case 7 (M and N); ovarian mucinous and uterine endometrioid: case 8 (O and P). Metastatic cases. Ovarian endometrioid and uterine endometrioid: case 3 (E and F), case 4 (G and H), case 6 (K and J); high-grade serous carcinomas in the uterus and ovary: case 9 (Q and R).

Table 3 The mutation status of SEO tumors: same histologic type (endometrioid type)

Case	Patient age (y)	First diagnosis (pathological diagnosis)	*TP53* mutation	*KRAS* mutation	*PTEN* mutation	*CTNNB1* mutation	*POLE* mutation	Final diagnosis (genetic analysis)
1	53	OC 3C (E) Synchronous	W	W	W	W	W	Synchronous
		EC 1B (E) independent (S)	W	W	W	W	W	independent (S)
2	65	OC 1C (E)	W	W	W	c.110_111 insA	W	S
		EC 1B (E)	W	W	W	W	W	
3	40	OC 1C (E)	p. S215 T	W	W	c.203_204 [a] insA	W	Metastatic
		EC 1A (E)	W	W	W	c.203_204 [a] insA	p.A341T	(M)
4	62	OC 1A (E)	W	W	p. Y16C	W	p.Q390H [c]	M
							p.T434 H [c]	
		EC 1A (E)	W	W	W	W	p.Q292E	
							p.Q390H [c]	
							p.T434H [c]	
							c.1155_1156 insA	
5	51	OC 1B (E)	p. S215 T	W	W	W	p.H350Q	S
		EC 2 (E)	W	W	p. C71T	c.203_204 insC	p.E396V	
6	59	OC 4B (E)	W	W	W	p.S29C [b]	p.D287N [d]	M
		EC 4B (E)	W	W	W	p.S29C [b]	p.D287N [d]	
							p.N293D	

[a] In case 3, there was an A insertion at the same location in exon 3 of *CTNNB1* in both tumors.
[b] In case 6, there was a point mutation at the same location in exon 3 of *CTNNB1*.
[c] In case 4, there was a point mutation at the same location in exon 13 of *POLE*.
[d] In case 6, there was a point mutation at the same location in exon 9 of *POLE*.

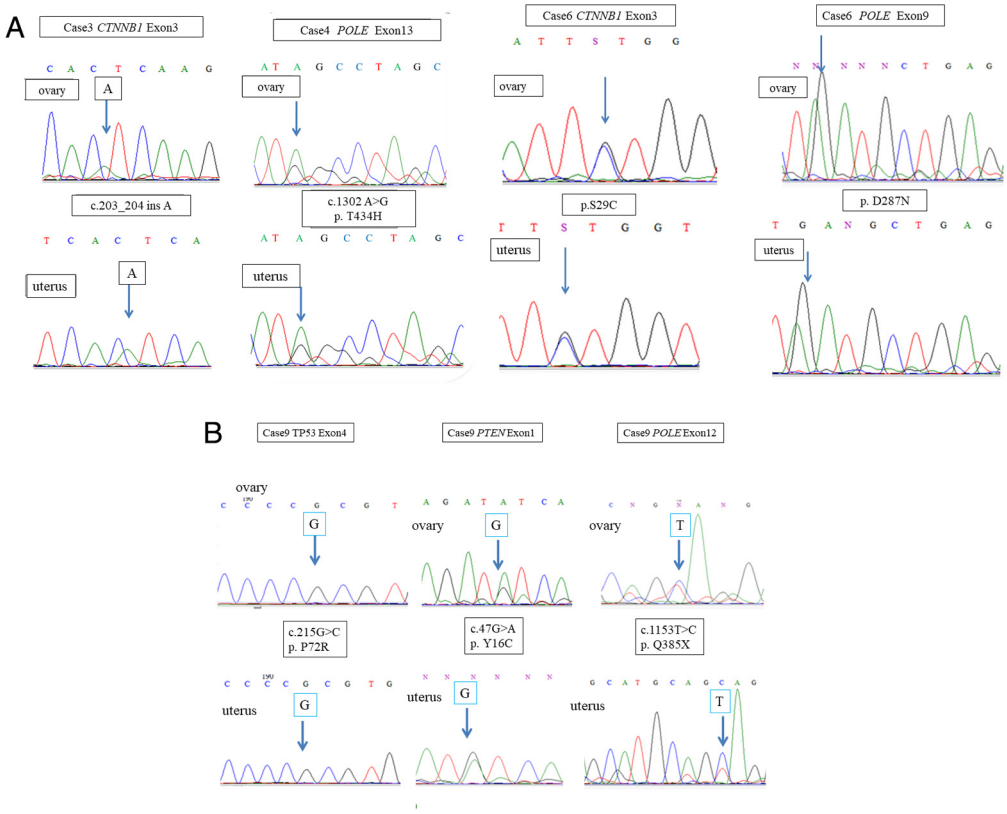

Fig. 2 Somatic mutations were identified at the same location in ovarian and uterine tumors. Mutations in SEO tumors are illustrated. Mutations were confirmed by Sanger sequencing using DNA from the ovarian tumor and endometrial tumor in the same patient. A, Somatic mutations were identified in the same point between ovarian tumor and uterine tumor (cases 3, 4, and 6). In case 3, there was an A insertion at the same location in exon 3 of *CTNNB1*. In case 4, there was a point mutation at the same location in exon 13 of *POLE*. In case 6, there were 2 point mutations at the same locations in exon 3 of *CTNNB1* and exon 9 of *POLE*. B, Somatic mutations were identified at the same points between metastatic tumors. In case 9, there were 3 point mutations at the same locations in exon 4 of *TP53*, exon 1 of *PTEN*, and exon 12 of *POLE*.

Table 4 The mutation status of metastatic tumors by pathological diagnosis

Case	Patient age (y)	First diagnosis (pathological diagnosis)		*TP53* mutation	*KRAS* mutation	*PTEN* mutation	*CTNNB1* mutation	*POLE* mutation	Final diagnosis (genetic analysis)
9	72	EC4B (S)	Metastatic (M)	p.P72R [a]	p.V14E	p.Y16C [b] p.R120K	p.T68 N	p.Q390H p.Q385X [c] p.R372L	Metastatic (M)
				p.P72R [a]	W	p.Y16C [b]	W	p.Q385X [c]	

[a] There was a point mutation at the same location in exon 3 of *CTNNB1*.
[b] There was a point mutation at the same location in exon 3 of *PTEN* in both tumors.
[c] There was a point mutation at the same location in exon 3 of *POLE* in both tumors.

The main limitation of this study was the small number of SEO cases, which led to no significant difference in terms of patient prognosis.

Recent studies have evaluated whether tumor mutation burden is associated with the frequency of somatic mutations [23-25]. In SEOs, if there is an association between *POLE* mutations and a good prognosis via the mutation burden of tumors, checkpoint blockade immune therapies might show a good response. In future studies, we will investigate associations between the overexpression of PD-1/PD-L1 or loss of MMR

Table 5 The mutation status of SEO tumors: other histologic type

Case	Patient age (y)	First diagnosis (pathological diagnosis)		*TP53* mutation	*KRAS* mutation	*PTEN* mutation	*CTNNB1* mutation	*POLE* mutation	Final diagnosis (genetic analysis)
7	49	OC 3C (S)	Synchronous independent (S)	p.L25Q	W	p.Y16C	W	p.F285I p.T483I c.1219_1220 insA	Synchronous independent (S)
		EC 1B (E)		W	W	p.E40Q p.A120E	W	p.Q390H p.E396V	
8	53	OC 1A (M)		W	W	W	W	p.K284E	
		EC 1B (E)		W	p.G12D	W	W	W	

Table 6 The factor of risk of recurrence for patients with SEO tumors (n = 9)

Factors	Patients (n = 9)	*POLE* mutation		*P*
		Negative, n (%)	Positive, n (%)	
Age (y)				
<50	2	0 (0)	2 (100.0)	.391
≥50	7	2 (28.6)	5 (71.4)	
FIGO stage				
I, II	5	4 (80.0)	1 (20.0)	.858
III, IV	4	1 (25.0)	3 (75.0)	
Recurrence				
No	5	1 (20.0)	4 (80.0)	.858
Yes	4	1 (25.0)	3 (75.0)	
TP53				
Negative	7	2 (28.6)	5 (71.4)	.391
Positive	2	0 (0)	2 (100.0)	
KRAS				
Negative	8	2 (25.0)	6 (75.0)	.571
Positive	1	0 (0)	1 (100.0)	
PTEN				
Negative	5	2 (40.0)	3 (60.0)	.151
Positive	4	0 (0)	4 (100.0)	
CTNNB1				
Negative	4	1 (25.0)	3 (75.0)	.858
Positive	5	1 (20.0)	4 (80.0)	

proteins in SEOs and the response to checkpoint blockade immunotherapies to establish useful biomarkers for these tumors.

In the future, clinical sequencing will be an important strategy for the diagnosis and treatment decisions for SEOs. It will be possible to determine not only the correct tumor origin but also the tumor characteristics, such as the active immune microenvironment. This information is useful for decision making for these patients.

Conflicts of interest

The authors declare no conflicts of interest.

Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.humpath.2018.11.001.

References

[1] Chui MH, Ryan P, Radigan J, et al. The histomorphology of Lynch syndrome associated ovarian carcinomas: toward a subtype-specific screening strategy. Am J Surg Pathol 2014;38:1173-81.
[2] Heitz F, Amant F, Fotopoulou C, et al. Synchronous ovarian and endometrial cancer—an international multicenter case-control study. Int J Gynecol Cancer 2014;24:54-60.
[3] Zaino R, Whitney C, Brady MF, De Geest K, Burger RA, Buller RE. Simultaneously detected endometrial and ovarian carcinomas—a prospective clinicopathologic study of 74 cases: a gynecologic oncology group study. Gynecol Oncol 2001;83:355-62.
[4] Soliman PT, Slomovitz BM, Broaddus RR, et al. Synchronous primary cancers of the endometrium and ovary: a single institution review of 84 cases. Gynecol Oncol 2004;94:456-62.
[5] Ramus SJ, Elmasry K, Luo Z, et al. Predicting clinical outcome in patients diagnosed with synchronous ovarian and endometrial cancer. Clin Cancer Res 2008;14:5840-8.
[6] Schultheis AM, Ng CK, De Filippo MR, et al. Massively parallel sequencing-based clonality analysis of synchronous endometrioid endometrial and ovarian carcinomas. J Natl Cancer Inst 2016;108:djv427.
[7] Anglesio MS, Wang YK, Maassen M, et al. Synchronous endometrial and ovarian carcinomas: evidence of clonality. J Natl Cancer Inst 2016;108:djv428.
[8] Chao A, Wu RC, Jung SM, et al. Implication of genomic characterization in synchronous endometrial and ovarian cancers of endometrioid histology. Gynecol Oncol 2016;143:60-7.
[9] Scully RE, Young RH, Clement PB. Tumors of the ovary, maldeveloped gonads, fallopian tube, and broad ligament. Atlas of Tumor Pathology. Armed Forces Institute of Pathology: Bethesda, MD; 1998.
[10] Singer G, Stohr R, Cope L, et al. Patterns of p53 mutations separate ovarian serous borderline tumors and low- and high-grade carcinomas and provide support for a new model of ovarian carcinogenesis: a mutational analysis with immunohistochemical correlation. Am J Surg Pathol 2005;29:218-24.
[11] Leitao MM, Soslow RA, Baergen RN, Olvera N, Arroyo C, Boyd J. Mutation and expression of the TP53 gene in early stage epithelial ovarian carcinoma. Gynecol Oncol 2004;93:301-6.
[12] Sato N, Tsunoda H, Nishida M, et al. Loss of heterozygosity on 10q23.3 and mutation of the tumor suppressor gene PTEN in benign endometrial cyst of the ovary: possible sequence progression from benign endometrial cyst to endometrioid carcinoma and clear cell carcinoma of the ovary. Cancer Res 2000;60:7052-6.
[13] Ali-Fehmi R, Khalifeh I, Bandyopadhyay S. Patterns of loss of heterozygosity at 10q23.3 and microsatellite instability in endometriosis, atypical endometriosis, and ovarian carcinoma arising in association with endometriosis. Int J Gynecol Pathol 2006;25:223-9.
[14] Singer G, OldtIIIrd R, Cohen Y, et al. Mutations in BRAF and KRAS characterize the development of low-grade ovarian serous carcinoma. J Natl Cancer Inst 2003;95:484-6.
[15] Ho CL, Kurman RJ, Dehari R, Wang TL, Shih IM. Mutations of BRAF and KRAS precede the development of ovarian serous borderline tumors. Cancer Res 2004;64:21-8.
[16] McConechy MK, Ding J, Senz J, et al. Ovarian and endometrial endometrioid carcinomas have distinct CTNNB1 and PTEN mutation profiles. Mod Pathol 2014;27:128-34.
[17] Hoang LN, McConechy MK, Köbel M, et al. Polymerase epsilon exonuclease domain mutations in ovarian endometrioid carcinoma. Int J Gynecol Cancer 2015;25:1187-903.
[18] Cancer Genome Atlas Research Network, Kandoth C, Schultz N, et al. Integrated genomic characterization of endometrial carcinoma. Nature 2013;497:67-73.
[19] Kelemen LE, Rambau PF, Koziak JM, Steed H, Köbel M. Synchronous endometrial and ovarian carcinomas: predictors of risk and associations with survival and tumor expression profiles. Cancer Causes Control 2017;28:447-57.
[20] Garg K, Shih K, Barakat R, Zhou Q, Iasonos A, Soslow RA. Endometrial carcinomas in women aged 40 years and younger: tumors associated with loss of DNA mismatch repair proteins comprise a distinct clinicopathologic subset. Am J Surg Pathol 2009;33:1869-77.
[21] Djordjevic B, Hennessy BT, Li J, et al. Clinical assessment of PTEN loss in endometrial carcinoma: immunohistochemistry outperforms gene sequencing. Mod Pathol 2012;25:699-708.
[22] Kobayashi Y, Nakamura K, Nomura H, et al. Clinicopathologic analysis with immunohistochemistry for DNA mismatch repair protein expression in synchronous primary endometrial and ovarian cancers. Int J Gynecol Cancer 2015;25:440-6.
[23] Le DT, Uram JN, Wang H, et al. PD-1 blockade in tumors with mismatch-repair deficiency. N Engl J Med 2015;372:2509-20.
[24] Billingsley CC, Cohn DE, Mutch DG, Stephens JA, Suarez AA, Goodfellow PJ. Polymerase ε (POLE) mutations in endometrial cancer: clinical outcomes and implications for Lynch syndrome testing. Cancer 2015;121:386-94.
[25] Dogan A, Schultheis B, Rezniczek GA, et al. Synchronous endometrial and ovarian cancer in young women: case report and review of the literature. Anticancer Res 2017;37:969-78.