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Original Contribution

Undifferentiated endometrial carcinoma: A selected immunohistochemical panel including PAX-8 and E-cadherin for aiding distinction from other endometrial carcinomas

Xiaofei Zhang*, Minghua Yu

Department of Surgical Pathology, Women's Hospital, School of Medicine, Zhejiang University, Zhejiang, PR China

1. Introduction

Undifferentiated endometrial carcinoma (UEC) is a rare and clinically aggressive malignant uterine epithelial variant with no differentiation by 2014 World Health Organization classification [1]. The incidence of UEC was reported to range between 1% [2] and 9% [3,4] in the limited published studies. The discrepancy is largely due to unfamiliarity with this disease entity, although a few UEC case series have been published in recent years, describing its morphologic features and immunohistochemical staining profile. A portion of UEC is associated with a differentiated component, mainly International Federation of Gynecology and Obstetrics (FIGO) grade 1/2 endometrioid carcinoma, whereas FIGO grade 3 endometrioid carcinoma (G3EC), serous carcinoma (SC) [5], or clear cell carcinoma [4] may also occur concurrently, upon which it is named as dedifferentiated endometrioid adenocarcinoma (DEAC). The concurrent presence of a differentiated component and a solid component in UEC might easily be misdiagnosed as a FIGO G2EC or G3EC when not considering this tumor. The distinction of UEC/DEAC from other endometrial carcinomas has important clinical significances as UEC/DEAC has a poorer outcome, and even an undifferentiated component of as low as 20% is associated with an aggressive outcome [6].

A series of immunophenotypes have been studied to clarify the characteristic immunohistological expression of UEC and to distinguish it from its mimics, such as G3EC (the most common misdiagnosis), SC with a solid growth pattern, and carcinosarcoma (malignant mixed Müllerian tumor (MMMT)). However, no unique immunohistologic marker has been found to identify UEC to date. PAX-8, an epithelial marker for tumors of Müllerian origin, is reported to be lost in about 80% of UEC cases [5,7], and appears to be a credible marker in distinguishing UEC from other endometrial carcinomas. E-cadherin, which is associated with epithelial-to-mesenchymal transition, is reported to be lost in more than half of the UEC cases [8].

In this present study, we examined the clinicopathologic features and evaluated a panel of immunohistochemical markers in our UEC/DEAC cases. The aim was to determine an effective immunomarker panel for clinical use in the distinction of UEC from other endometrial carcinomas.

2. Materials and methods

2.1. Samples

The database of the Department of Pathology at the Women Hospital, School of Medicine, Zhejiang University was searched for cases of UEC. The database on the Pathology Information System Registry was searched for cases diagnosed as UEC, G3EC, and SC from 2005 to 2018. We collected patient clinical information, including age, presenting symptoms, surgery type, tumor stage, adjuvant therapy, and follow-up.

Hematoxylin and eosin slides were reviewed for all cases. Morphologic features, including the growth pattern (solid sheets, nests, and alveolar patterns), size of the tumor cell, presence of rhabdoid morphology, lymphovascular invasion, concomitant components (e.g., endometrioid, serous, or sarcomatous) and their proportion, necrosis, and myxoid background, were examined.

2.2. Immunohistochemistry analysis

Immunohistochemical staining was performed on formalin-fixed, paraffin-embedded tissue sections using the Envision™ method. Positive and negative controls were used for each test. The immunohistochemical staining was performed with antibodies against the following markers: CKAE1/AE3, CK8/18, epithelial membrane antigen (EMA), estrogen receptor (ER), progesterone receptor (PR), p16, p53, PAX-8, E-cadherin, and the mismatch repair protein (MMRP) markers MLH1, MSH2, MSH6, and PMS2. Details on the antibodies used are provided in Table 1. All markers of the immunohistochemical panel were also examined in G3EC ($n = 20$) and SC ($n = 11$) control groups. Staining for p16 was considered positive if strong nuclear and

* Corresponding author at: Department of Surgical Pathology, the Affiliated Women's Hospital, School of Medicine, Zhejiang University, 1 Xueshi Road, Hangzhou, Zhejiang Province 310006, PR China.

E-mail address: pathozxf@zju.edu.cn (X. Zhang).

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Table 1
Information on antibodies used for immunohistochemistry.

Antibodies	Vendor	Clone	Dilution
Cytokeratin AE1/AE3	MXB	AE1, AE3	1:500
Cytokeratin 8/18	Gene Tech	5D3	1:120
EMA	MXB	E29	1:500
ER	Gene Tech	SP1	1:200
PR	Gene Tech	SP2	1:400
PAX-8	Gene Tech	ZR-1	1:250
E-cadherin	Gene Tech	EP700Y	1:1000
P16	Gene Tech	G175-405	1:150
P53	MXB	DO-7	1:300
MLH1	Gene Tech	ES05	1:50
MSH2	Gene Tech	25D12	1:50
MSH6	Gene Tech	EP49	1:400
PMS2	Gene Tech	EP51	1:50

ER indicates estrogen receptor; PR, progesterone receptor.

cytoplasmic staining was seen in > 75% of the tumor cells; otherwise, it was considered negative. Positivity for p53 was defined as either a diffuse and strong nuclear staining in > 75% of the tumor cells or a complete loss of staining; otherwise, it was interpreted as negative. Loss of nuclear staining in any of the four MMRPs with appropriate positive internal control was interpreted as a positive result. For all other immunomarkers, the staining proportion was graded as negative (< 5% of cells stained), focal (5%–25% of cells stained), patchy (26%–75% of cells stained), and diffuse (> 75% of cells stained).

2.3. Statistical analysis

All statistical analyses were performed using SPSS software version 23.0 (SPSS Institute Inc., Cary, NC). The Pearson "χ^2" (chi square) test or the Fisher exact test for small sample sizes was used to analyse expression rates of different markers in UEC, G3EC and SC.

P value of < 0.05 was considered to be significant.

3. Results

3.1. Clinicopathologic characteristics of UEC

Six cases of pure UEC and 5 of DEAC were identified between 2005 and 2018. These 11 UEC/DEAC cases comprised 1% of all endometrial carcinomas (11/1086) diagnosed during this period. Twenty cases of G3EC and 11 cases of SC were selected as the controls. The 11 patients with UEC were all postmenopausal, with an average age of 61 years. Postmenopausal bleeding presented as the same complaints. None of the curettage specimens in the UEC group were correctly diagnosed. Six of the 11 (55%) UEC/DEAC cases were at the advanced stages when diagnosed.

The patients with UEC presented with aggressive features, including lymphovascular invasion (82%) and lymph node metastasis (54%). All patients underwent a total abdominal hysterectomy, bilateral salpingo-oophorectomy, and pelvic lymphadenectomy. Seven patients underwent an additional paraaortic lymphadenectomy and 3 had an omentectomy in UEC/DEAC. Nine patients with UEC/DEAC received chemoradiation therapy after surgery. Follow-up information was available for all 11 UEC/DEAC cases. Five of the patients with UEC/DEAC died at 1 month, and 6, 6, 9, and 35 months, respectively, after surgery, and the remaining 6 patients were free of disease, with a follow-up time of 1–89 months (median 9 months) (Table 2).

3.2. Morphologic feature of UEC/DEAC

Of the 11 cases, 6 (55%) were composed purely of UEC, and the 5 other cases were mixed with differentiated carcinoma (DEAC). Three cases of DEAC were associated with FIGO G2EC, with the undifferentiated carcinoma component constituting 80%, 80%, and 90% of the total tumor volume, respectively. The remaining 2 DEAC cases had combined SC, which constituted 50% and 90% of the total tumor volume, respectively. The differentiated component in all 5 DEAC cases was superficial, whereas the undifferentiated carcinoma was deep or adjacent to it. The transition between the 2 components was abrupt, with a sharp border (Fig. 1A). Diffuse sheets (a characteristic growth pattern of the undifferentiated carcinoma component) were found in 8 cases (73%) (Fig. 1B,C), alveolar combined with solid sheets in 2 cases, and a focal nest pattern in 1 case. Medium-sized monotonous tumor cells were observed in 7 cases, medium-sized cells admixed with focal large cells in 3 cases, and mainly large cells in 1 case. Focally rhabdoid cells were seen in 5 cases (45%) (Fig. 1D), whereas a myxoid background was seen in 3 cases (27%) (Fig. 1E). Geographic necrosis and active mitotic and apoptotic figures were seen in all the UEC cases (Fig. 1F).

Although solid sheets or nests may be the predominant structures in G3EC, at least focal conspicuous glandular or trabecular structures could be seen. Moreover, tumor cells that had transitioned from the glandular area to the solid area of G3EC could also be found, and they shared a similar large-sized cytomorphology with large nuclei. Mitosis and necrosis were variable, often less than those in UEC. Rhabdoid cells and a myxoid background could hardly be seen in G3EC. With regard to SC, although it may be associated with the solid component, the characteristics of papillae, slit-like compressed spaces, and architectural–cytologic dyssynchrony may intrigue its diagnosis.

3.3. Immunohistochemical features

The results of the immunohistochemical studies on UEC and analysis of different markers in G3EC and SC are summarized in Tables 3 and 4, respectively. The immunohistochemical expression of the undifferentiated component in UEC/DEAC, G3EC, and SC was as follows: of the 11 UEC cases, pan-cytokeratin was positive in 6 cases (55%), CK8/18 in 9 cases (82%) (Fig. 2A), and EMA in 9 cases (82%). EMA and CK8/18 were the only positive epithelial markers in 2 different cases (9%). In all UEC cases, although positive expression for at least 1 of the 3 epithelial markers was found, it mostly showed focal strong staining. In contrast, diffuse and strong immunoreactivity was found in 90% of the G3EC and 100% of the SC cases. The difference in positivity expression of the tumor cells had statistical significance ($P = 0.000$). The ER was negative in most UEC cases (82%) except for 2 cases, whereas 2 of the 20 G3EC cases (10%) and 3 of the 11 SC cases (27%) were negative for this receptor (P = 0.000 and 0.03, respectively). Almost all UEC cases (91%) except for 1 case were negative for the PR, whereas 7 of the G3EC cases (35%) and 5 of the SC cases (45%) had lost the expression of this receptor. The difference in PR expression between UEC and G3EC had statistical significance ($P = 0.003$), whereas that between UEC and SC did not ($P = 0.063$).

Eight of the UEC cases (73%) completely lost the expression of PAX-8 (Fig. 2B), but the expression was strongly and diffusely positive in a single case (9%), and weakly and patchy positive in another 2 cases. In contrast, 2 of the G3EC cases (10%) showed negativity for PAX-8, 17 cases (85%) showed diffuse and strong positivity, and 1 case (5%) showed focal weak staining. Ten of the SC cases (91%) exhibited diffuse and strong positivity for PAX-8, and only 1 case had lost the PAX-8 expression. A total loss of E-cadherin expression was found in 8 of the UEC cases (73%) (Fig. 2C), whereas 1 case showed patchy positive staining and the remaining 2 cases exhibited focal positivity. Meanwhile, most of the G3EC (95%) and SC cases (82%) showed diffuse positivity for E-cadherin, and only 1 of the G3EC cases (5%) and 2 of the SC cases (18%) showed negative E-cadherin immunoreactivity. The differences in expression of PAX-8 and E-cadherin between UEC and G3EC, and between UEC and SC, were statistically significant ($P < 0.05$). Of the 11 cases of UEC, 7 cases showed negativity for both PAX-8 and E-cadherin, 1 case with diffuse positivity for PAX-8 was

Table 2
Clinical and pathologic features of UEC cases.

#	Age (yr)	Presenting Symptoms	Stag/ FIGO	Surgery	CTX	RTX	Status	OS (months)	Morphology (differentiated component %)	Curettage specimenDX
1	64	AB	IIIC	TAH + PLN	NO	NO	DOD	6	UEC	NO
2	63	VD	IIIC2	TAH + BSO + OM PLN + PALN	NO	YES	DOD	10	UEC with SC (50%)	SC
3	62	PMB	IA	TAH + BSO + PLN	NO	YES	FOD	33	UEC with SC (90%)	G3 EC
4	74	PMB	IIIC2	TAH + BSO + PLN + PALN	NO	NO	DOD	6	UEC	CS
5	65	PMB	IIIC1	TAH + BSO + PLN + PALN	NO	YES	DOD	12	UEC	G3 EC
6	59	PMB	IB	TAH + BSO + PLN + PALN	NO	NO	FOD	10	UEC with EC (G2 20%)	G3 EC
7	54	PMB	IA	TAH + BSO + PLN + PALN	YES	NO	FOD	4	UEC with EC (G1 20%)	G3 EC
8	52	PMB	IIIC	TAH + BSO + PLN + OM	YES	YES	AWD	2	UEC	PDC
9	60	PMB	II	TAH + BSO + OM PLN + PALN	NO	YES	FOD	1	UEC	PDC
10	60	PMB	IIIB	TAH + BSO + PLN	YES	NO	LFU	NA	UEC	G3 EC
11	61	PMB	IIB	TAH + BSO + PLN	YES	YES	FOD	89	UEC with EC (G2 10%)	CS

AB, abdominal distention; AWD, alive with disease; BSO, bilateral salpingo-oophorectomy; CTX, chemotherapy; CS, carcinosarcoma; D, dead; DEC, de-differentiated endometrial carcinoma; DX, diagnosis; EC, endometrioid carcinoma; FOD, free of disease; HYS, hysterectomy; LFU, lost follow-up; NA, not available; OM, omentum; OS, overall survival; PALN, para-aortic lymph nodes; PDC, Poorly differentiated carcinoma PLN, pelvic lymph nodes; PMB, postmenopausal bleeding; RTX, radiotherapy; SC, serous carcinoma; SO, salpingo-oophorectomy; TAH, total abdominal hysterectomy; VD, vaginal discharge.

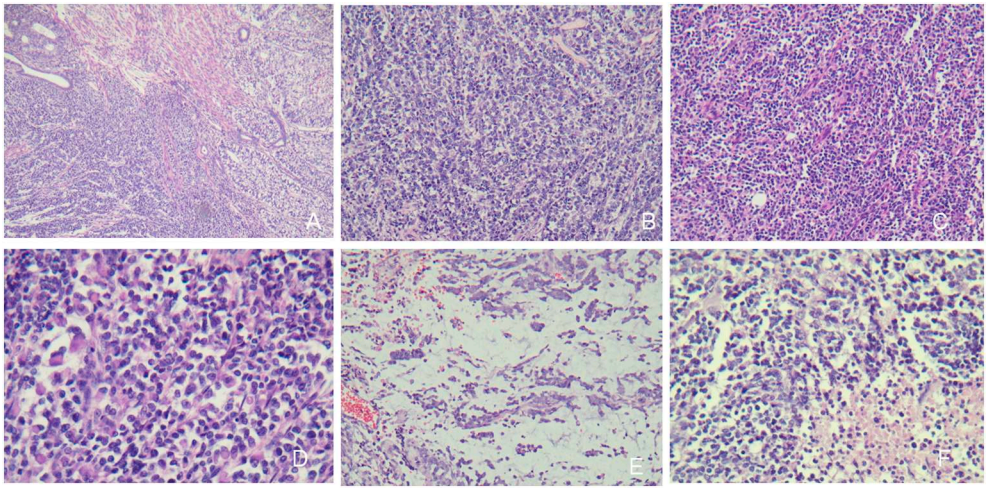

Fig. 1. Morphologic feature of UEC/DEAC. (A) Examples of the abrupt transition between a well-differentiated endometrioid adenocarcinoma and an adjacent undifferentiated endometrial carcinoma. (B, C) An undifferentiated endometrial carcinoma is composed of patternless solid sheets of discohesive tumor cells with intervening delicate fibrovascular septae. (D) Focally rhabdoid cells were found in a few cases. (E) A myxoid background was seen in a few cases. (F) Monotonous discohesive cells with high-grade nuclei, frequent mitoses, and necrosis.

Table 3
The Immunohistochemical profile of the undifferentiated endometrial carcinoma (UEC).

No	AE1/AE3	CK8/18	EMA	ER	PR	PAX-8	E-Cad	P16	P53	MLH1/PMS2	MSH2/MSH6
1	N	FPS	FPS	N	N	N	N	P	p	R/R	R/R
2†	N	FPS	FPS	N	N	DP	N	P	p	R/R	R/R
3†	FPS	FPS	FPS	PP	N	PP	PP	P	p	R/R	R/R
4	N	N	FPS	N	N	N	N	N	p	R/R	R/R
5	N	FPS	N	N	N	N	FP	N	N	L/L	R/R
6*	FPS	FPS	FPS	PP	FP	PP	FP	P	N	L/L	R/R
7*	N	FPS	FPS	N	N	N	N	P	P	R/R	R/R
8	FPS	FPS	N	N	N	N	N	N	N	L/L	R/R
9	FPS	N	FPS	N	N	N	N	P	N	R/R	R/R
10	FPS	FPS	FPS	N	N	N	N	N	N	L/L	R/R
11*	FPS	FPS	PPS	N	N	N	N	N	N	L/L	R/R

DP, diffuse positive; FP, focal positive; L, lost; N, negative; O, other component; P, positive.
PP, pathy positive; R, retained; Vim, Vimentin; S, strong; W, weak.
* Undifferentiated carcinoma mixed with endometrioid carcinoma.
† Undifferentiated carcinoma mixed with serous carcinoma.

negative for E-cadherin, the other 2 cases with patchy positivity for PAX-8 showed patchy and focal positivity for E-cadherin.

Six of the UEC cases (55%) were positive for p16 (Fig. 2D), compared with 6 of the G3EC cases (30%) and 10 of the SC cases (91%) being positive for this marker. Five of the UEC cases (45%) showed diffuse and strong staining for p53, and 6 (55%) showed wild-type staining. With regard to the G3EC cases, 5 (25%) showed diffuse and strong positivity for p53. Ten of the SC cases (91%) showed diffuse and strong positivity for p53. None of the differences in p16 and p53 expression between UEC and G3EC, and between UEC and SC, had statistical significance ($P > 0.05$).

Five of the UEC cases (45%), 7 of the G3EC cases (35%), and 1 of the SC cases (9%) showed a loss of MLH1 and PMS2 immunoreactivity, whereas all UEC, G3EC, and SC cases showed nuclear positivity for

Table 4
Immunohistochemical analysis of different markers in undifferentiated endometrial carcinoma (UEC), FIGO Grade 3 Endometrial Carcinoma (G3EC) and serous carcinoma (SC).

	UEC {Component)		G3EC 20 cases		SC 11 cases		UEC/G3EC	UEC/SC
	11cases [n]		(solid area) [n]		[n]		P	P
AE1/AE3 + CK8/18 + EMA	FP 10	DP 0	FP 1	DP 19	FP 0	DP 11	0.000	0.000
ER	N 9	P 2	N 2	P 18	N 3	P 8	0.000	0.03
PR	N 10	P 1	N 7	P 13	N 5	P 6	0.003	0.063
PAX-8	N 8	P 3	N 2	P 18	N 1	P 10	0.001	0.008
E-Cad	N 8	P 3	N 1	P 19	N 2	P 9	0.000	0.03
P16	N 5	P 6	N 14	P 6	N 1	P 10	0.255	0.149
P53	N 6	P 5	N 15	P 5	N 1	P 10	0.423	0.063
MLH1/PMS2	L 5	R 6	L 7	R 13	L 1	R 10	0.705	0.149
MSH2/MSH6	L 0	R11	L 0	R 20	L 0	R 11		

MSH2 and MSH6. Although it seemed that more cases of UEC had lost the expression of MMRP, the difference was not statistically significant ($P > 0.05$).

4. Discussion

UEC is a rare, high-grade endometrial carcinoma. Although we have learned increasingly more about the morphologic features and immunophenotype of this disease entity, it is still underrecognized. Considering its highly aggressive behavior [3,9], UEC must be distinguished from other endometrial carcinomas, such as endometrioid carcinoma, especially when associated with a more differentiated component, mostly FIGO grade 1/2 endometrioid carcinoma (DEAC), SC, and MMMT (carcinosarcoma). The incidence of UEC in our hospital is 1%, in accordance with the reported incidences of 0.7%–2% [2,10], but much lower in comparison with other reported incidences of 4% and 9% [3,7]. Some factors related to this discrepancy are the rarity of this tumor (leading to it limited recognition) and the absence of characteristic immunohistologic markers (making identification of UEC more difficult), which may lead to underrecognition, and even overrecognition, of UEC.

Misinterpretation frequently occurs with UEC, where only 18% of cases have been correctly diagnosed, according to the study of Altrabulsi and Al-Hussaini [3,7]. In a study by Laura and colleagues, out of 22 consultation cases, 6 were misdiagnosed as G3EC, 5 as poorly differentiated/high-grade carcinoma, and 4 as carcinosarcoma [9]. UEC could hardly be identified on biopsy, owing to the limited sample and misinterpretation, especially for DEAC, when concurrent with differentiated glandular carcinoma. The biopsy diagnosis in 2 of our 5 DEAC cases was G3EC, and was SC in 1 case, as concurrency with the evident SC component led to misdiagnosis of the UEC as a solid SC. A similar misinterpretation occurred with the rest of our DEAC cases, where when evidence of a carcinoma with a myxoid background and dispersive tumor cells with sheet-like growth mimicking a sarcoma was found, the diagnosis of MMMT had been made. With respect to the 6 cases of pure UEC, 3 were diagnosed as a poorly differentiated cancer, 1 as a G3EC, and 1 as a sarcoma. The features related to the distinction of UEC from G2EC and G3EC have been mentioned in our Results section and by others previously [3,9]. With regard to sarcomas, the inclusion of the components of carcinosarcoma and pure sarcoma always consists of pleomorphic spindle-cell proliferation [11]. The cancer component in MMMT is usually a high-grade carcinoma, most frequently SC, in contrast to a low-grade endometrioid type in DEAC [11]. MMMT typically occurs in older women, whereas UEC can occur in young patients. Moreover, a proportion of DEAC appears to be associated with microsatellite instability, in contrast to MMMT, which does not seem to be related to this feature [9,11].

In addition to the morphologic manifestations, a panel of

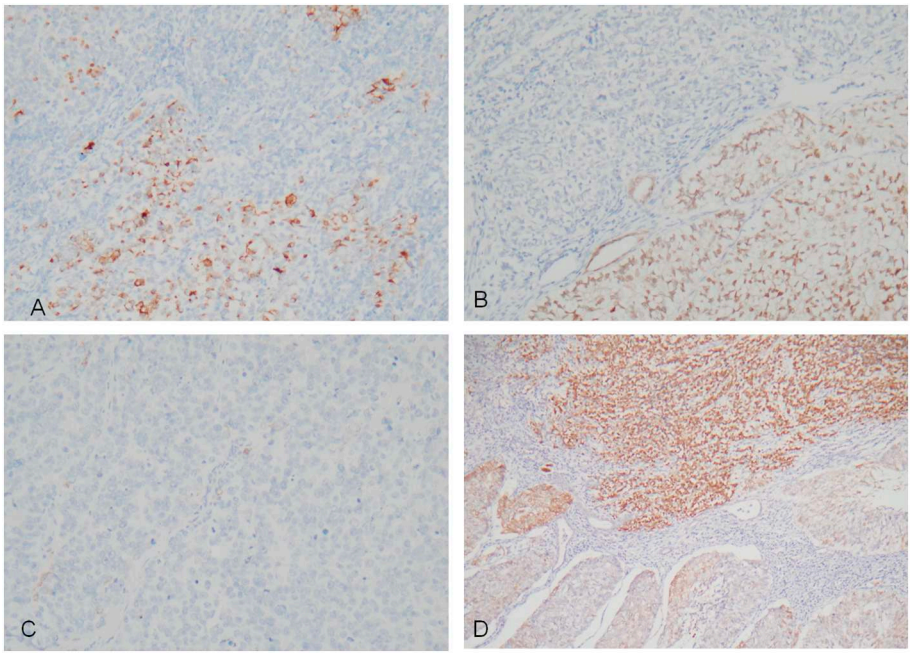

Fig. 2. Immunohistochemical features of UEC/DEAC. (A) CK8/18, showing patchy staining in undifferentiated endometrial carcinoma (UEC). (B) High-power views of PAX-8 staining in dedifferentiated carcinoma, showing strong staining in the endometrioid component and negative staining in the undifferentiated carcinoma (UEC) component. (C) Loss of E-cadherin expression in UEC. (D) Patchy staining for p16 in the endometrioid component, and strong and diffuse staining in the UEC component.

immunophenotypes may also aid in distinguishing UEC from other endometrial malignancies. The expression of at least one of the cytokeratin cocktail, including EMA, is a robust marker in supporting the epithelial nature of the tumor. The reported positive expression is mostly focal and strong [7,9,10,12,13]. However, in a study by Ramalingam [5], 54% of the UEC cases showed either patchy or diffuse expression of the keratin cocktail, and those UEC cases showed focal expression in a previous study [3]. Ramalingam attributed this discrepancy to the different antibody dilution and retrieval methods used [5]. Our results are similar to those of previous reports, with the positive staining pattern in all UEC cases focal and strong. We also found CK8/18 and EMA was the only positive markers in 2 cases, suggesting the epithelial origin of the tumor. It is necessary to use > 1 cytokeratin cocktail, especially CK8/18, including EMA to clarify the epithelial differentiation. The possibility of UEC must be considered whenever there is a case of endometrial neoplasms made up of sheets of epithelioid cells that are negative for epithelial markers.

PAX-8 belongs to the paired box gene transcription factors, and has crucial roles in the embryonic development of the kidney, Müllerian organs, and thyroid. It could be used as a sensitive and specific marker for tumors of renal, Müllerian, or thyroid origin, despite their primary and metastatic sites [14-16]. PAX-8 expression was found to be lost in most of the UEC cases in recent studies [5, 7 , 12], compared with that in G3EC, SC, clear cell carcinoma, and carcinosarcoma, all high-grade endometrial tumors that have been shown to retain the expression of PAX-8 [14,15,17]. The characteristic loss of PAX-8 expression makes it a reliable immunohistologic marker to differentiate UEC from other high-grade endometrial tumors, although a small number of UEC cases reported positivity in the above studies; that is, 17% (6/35), 13% (2/15), and 8% (1/13), respectively [5,7 , 12]. Among those positive cases reported, 4/9 UEC cases showed diffuse and strong PAX-8 immunoreactivity. We report similar findings herein, where most of our UEC cases were negative (73%) for PAX-8, with positive staining in only 3 cases. In contrast, PAX-8 was positive in 95% of the G3EC and 91% of the SC cases. Given the usual absence of PAX-8 in UEC, the findings of a metastatic undifferentiated carcinoma with negativity for PAX-8 should take the endometrial origin into consideration, especially for those with a differentiated endometrial carcinoma history that has progressed to local recurrence or distant metastasis with an undifferentiated carcinoma component [5).

E-cadherin, encoded by the CDH1 gene [18] and associated with epithelial-to-mesenchymal transition, is a master regulator of cell adhesion and polarity, and is always reduced or absent in poorly differentiated carcinomas [8,18]. The reduced or absent expression might be related to the poor prognosis, by enhancing the metastatic potential of undifferentiated endometrial carcinomas [19]. Laura and colleagues found that 67% of UEC cases had lost the expression of E-cadherin, in contrast to 7%, 20%, and 41% in the G1/2EC, G3EC, and SC cases, respectively, and suggested that the immunohistochemical analysis of E-cadherin could help distinguish UEC from G3EC [8]. However, this suggestion has been challenged by Ramalingam, who thought that E-cadherin was not of value in routine diagnosis owing to its variable expression in UEC [5]. We found that 73% (8/11) of our UEC cases showed loss of E-cadherin expression, 1 with focal weak and 2 with patchy weak staining, whereas most of our G3EC (95%, 19/20) and SC (82%, 9/11) cases showed diffuse positive staining, except for 1 G3EC case and 2 SC cases showing negative staining. Our results were similar to those of previous studies [7, 9–10 , 13], but the result for the SC cases contrasted with those published by Laura and colleagues, who found that 41% of their SC cases were negative for E-cadherin [8]. Combining the expression of PAX-8 and E-cadherin, we found that 7 of the UEC cases lost the expression of both markers, and 1 case with diffuse positivity for PAX-8 showed loss of E-cadherin expression. From these results above, we approved the suggestion that the analysis of E-cadherin expression might aid in distinguishing UEC from other subtypes of endometrial carcinomas.

Because most of our UEC cases were negative for ER and PR (82% and 91%, respectively), the combination of PAX-8 and ER negativity could be very helpful in distinguishing UEC from other subtypes of endometrial carcinoma. We found that 2 cases of UEC with patchy positivity for ER showed patchy positive expression of PAX-8, and the 2 cases of G3EC with loss of PAX-8 were all negative for ER. We speculate that there might be some relationship between the expressions of these two biomarkes, although the reason for the loss of PAX-8 expression in UEC remains unknown.

Whereas a strong positivity for both p16 and p53 is a useful immunohistologic marker for distinguishing between SC and G3EC, these staining patterns are not specific for UEC [20]. In our UEC cases, 55% and 45% showed strong and diffuse positivity for p16 and p53, respectively, while, the difference between UEC and G3EC, UEC and SC had no statistical significance. Nevertheless, Ramalingam suggested that p16 and p53, combined with PAX-8, might be helpful in differentiating UEC from SC with a solid pattern [5]. The positive expression of PAX-8 combined with the diffuse and strong positive expression of p16 and p53 will in favour of SC.

Frequent DNA MMRP alterations have been reported in UEC, most of them with loss of MLH1/PMS2 [5,7,9]. The loss of MSH2/MSH6 is rare, whereas MSH2 is the most common mutation seen in endometrial carcinomas related to hereditary nonpolyposis colorectal cancer or Lynch syndrome [21]. The deficient expression of MMRP in UEC is almost sporadic, which means it is secondary to the hypermethylation of MLH1 rather than being germline mutations related to Lynch syndrome [9,22]. However, Laura and colleagues pointed out that at least a proportion of UECs are associated with Lynch syndrome and suggested that all UEC cases should be tested for MMRPs. We had similar findings to those of previous reports [5,7,9], where 5 of our UEC cases (45%) were deficient for MLH1/PMS2 and none for MSH2/MSH6. However, none of our cases were tested for MLH1 hypermethylation.

5. Conclusions

In conclusion, UEC is a rare highly aggressive tumor that is frequently misdiagnosed. We suggest that the use of a panel of immunohistochemical stains including epithelial markers (pan-cytokeratin, CK8/18, EMA), ER, PR, PAX-8, and E-cadherin, will aide in the differential diagnosis of UEC from other endometrial carcinomas. The loss of expression of PAX-8, E-cadherin in suspected endometrial carcinoma will serve as robust indication of UEC.

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