Prognostic significance of fascin expression in endometrioid carcinoma

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Summary

Purpose of investigation: Actin bundling protein fascin has been previously associated with tumor progression in human cancers. We evaluated whether fascin also plays a role in endometrioid carcinomas.

Methods: Cases of 28 proliferative and hyperplastic endometrium and 43 endometrioid carcinomas were examined by immunohistochemistry using antihuman fascin antibody.

Results: Weak fascin expression in glandular epithelium was observed in 39% of non-neoplastic samples and various degrees of fascin expression were observed in 74% of neoplastic samples. The number of positively stained samples and intensity of epithelial staining were significantly higher in endometrioid carcinoma compared to the non-neoplastic group ($p < 0.001$). The number of positively stained samples and total fascin scores of stroma were significantly higher in proliferative and hyperplastic endometrium biopsies compared to the endometrioid carcinoma ($p < 0.001$). Higher grade endometrioid carcinoma cases had significantly increased total epithelial fascin scores (.042, $p < 0.05$). There was also a significant difference between tumor grade and patient survival (.040, $p < 0.05$). There was a significant correlation between microvessel count and disease-free survival ($r = .412$, $p = .006$). In the proliferative and hyperplastic endometrial biopsies microvessels stained homogeneously in all cases (28/28), but in the endometrioid carcinoma group eight out of 43 cases showed heterogeneous fascin staining of microvessels. The difference was significant (.019, $p < 0.05$).

Conclusions: Our study supported the dynamic role of actin bundling protein fascin in generating and maintaining endometrial neoplasms. It also showed that in the development of neoplasia, stromal fascin expression decreases but epithelial fascin expression up-regulates.

Key words: Endometrium; Endometrioid carcinomas; Fascin; Angiogenesis; Prognosis.

Introduction

The actin cytoskeleton is directly involved in cell locomotion, cytokinesis, cell-cell and cell-matrix interactions, vesicular and organelle transport and the establishment and maintenance of cell morphology. Fascin is an actin-regulatory protein which provides mechanical support to cellular protrusions and stress fibers. Neurons, glial cells, dendritic cells and endothelial cells express fascin [1-4]. Down-regulation or up-regulation of fascin expression is demonstrated in several types of human neoplasms, such as breast cancers, ovarian tumors, pancreas carcinomas, lung carcinomas, anaplastic large cell lymphomas, Hodgkin diseases, and skin tumors including melanomas [5-13]. In studies, fascin expression in breast cancer was restricted to high-grade tumors, which were more proliferative and metastatic. The up-regulation of fascin in estrogen and progesterone receptor negative breast cancers suggests that fascin may have a fundamental role in the acquisition of malignant tumorigenic phenotypes [5-6].

Endometrioid type (Type 1) endometrial carcinomas are related to unopposed estrogen stimuli. Normal and hyperplastic endometrial biopsies and endometrioid carcinomas can be a good model to examine fascin expression in patients who have different levels of estrogenic activity [14]. In this study, our objective was to evaluate the tissue distribution of fascin in biopsy samples representative of proliferative, hyperplastic endometrium and endometrioid carcinoma, with the aim of investigating the role of actin bundling protein fascin in endometrioid carcinoma progression and angiogenesis.

Materials and Methods

Tissue samples for this study were retrieved from the files of the Pathology Department of Osmangazi University Hospital between the years 1988 and 2000. Seventy-one endometrial biopsy samples representing proliferative endometrium (n = 10), simple hyperplasia (n = 10), complex hyperplasia (n = 8) and endometrioid carcinoma (n = 43) were included in the study. All of the biopsy samples were reexamined histopathologically. Presence or absence of inflammation, necrosis, metaplastic changes, grades of malignant samples, lymphatic invasion and lymph node metastasis were recorded. All malignant tumors were staged according to International Federation of Gynecology and Obstetrics (FIGO) criteria. Patient characteristics and clinicopathologic findings were obtained from hospital records. Stage and grade of endometrioid carcinoma samples are given in Table 1.

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Table 1. — *Stage and grade of endometrioid carcinomas.*

Variable	Endometrioid carcinoma
Stage	*no.*
IA	3
IB	13
IC	7
IIA	3
IIB	1
IIIA	6
IIIC	9
IVB	1
Grade	*no.*
1	6
2	28
3	9
Total samples	43

Immunohistochemistry

Paraffin-section blocks of the most representative sections of invasive tumor were selected from the study samples. The paraffin blocks were cut into 4 μm sections and immunohistochemical assays for the expression of fascin using liquid mouse monoclonal antihuman fascin antibody (Novocastra, NCL-L-FASCIN, USA) were performed. Tissue sections were deparaffinized in xylene, rehydrated in alcohol solutions, and placed in 0.5% hydrogen peroxide in methanol for 10 min to block endogenous peroxidase activity. Rehydration was completed by placing them in absolute alcohol and finally in water. The slides were treated with a boiling solution of freshly prepared 0.05 M-citrate buffer, pH 6.0 for 5 min in a pressure cooker. The sections were reacted overnight with the primary antibodies at a dilution of 1:300 in buffer. They were rinsed in phosphate buffered saline (PBS) before being treated with biotinylated universal secondary antibody for 10 min. After further rinsing, the slides were treated with avidin-biotin-peroxidase complex (Novocastra, Novostain Universal Detection Kit) and rinsed again. Immunostaining was accomplished by incubating them with 3 amino-9-ethylcarbazole (AEC) for 7 min and then the slides were rinsed in distilled water and counter-stained with Mayer's hematoxylin. Sections of human tonsil were used as positive controls. Capillary endotheliums were also used as endogenous positive controls. As a negative control, the primary antibody was replaced by PBS.

Assessment of fascin-stained slides

The positivity of endometrial glandular cells/tumor cells was categorized in three groups: ≤ 10% as 1, < 11-50% as 2 and 51-100% as 3. The intensity of immunostaining was scored on a three point scale: 1 = weak; 2 = moderate; 3 = intense. A weighed score for each tumor specimen was the sum of the percentage score and the intensity score and was defined as 'total epithelial fascin score'. Stromal staining was also assessed with both intensity score as 1 to 3 and percentage score as 1 to 2 (1-50% = 1, 51-100% = 2). A weighed score of each specimen was obtained from the sum of two scores and was defined as 'total stromal fascin score'. The role of fascin in endothelial cell migration and angiogenesis was investigated by counting endometrial and intratumoral microvessels in areas with the highest vascularity. The microvessel count was performed with a 15-cm monitor of a digital camera (Nikon Digital Sight, DS-5M-L1) on each area at a x 200 field (x 20 objective and x 10 ocular). The frame area for each captured image was the same. Each tumoral area was selected in the peripheral portions of the tumor including approximately equal portions of tumor and stroma for each tumor. The homogeneous or heterogeneous staining pattern of fascin expression in microvessels were assessed in both the non-neoplastic and neoplastic group.

Statistical analyses

All statistical analysis were performed using SPSS (Statistical Package of Social Services, Chicago, IL, USA) for Windows version 11.5. Data were analyzed according to the Mann-Whitney test, Pearson chi-square test, Fisher's exact chi-square test, continuity correction, T test and ANOVA. Probability values less than 0.05 were considered statistically significant.

Results

Mean ages of patients with proliferative endometrium, simple hyperplasia, complex hyperplasia and endometrioid carcinoma were 37, 43, 48 and 60, respectively. Table 1 shows the distribution of grades and stages of endometrioid carcinoma patients according to FIGO criteria.

Localization of fascin in non-neoplastic endometrial specimens

Proliferative endometrium

Stroma of proliferative endometrium stained diffuse and homogeneously in all samples (Figure 1). The total stromal fascin scores ranged from 3 to 5. Glandular epithelium stained for fascin in four (40%) samples and the total fascin immunoreactivity was found to be occasional and weak (the sum of scores = 2). Microvessel endothelium stained homogeneously in all samples. Mean microvessel count was 42.2 ± 4.04.

Simple hyperplasia

Fascin staining in stroma was seen in all samples. The total stromal fascin scores ranged from 2 to 4 (Figure 2). Glandular epithelium stained weakly (the sum of scores = 2) in one out of 10 samples. Microvessel endothelium stained homogeneously in all samples. Mean microvessel count was 62.6 ± 7.5.

Complex hyperplasia

Fascin staining in stroma was seen in all samples. The total stromal fascin scores ranged from 2 to 5. Glandular epithelium stained with fascin in six of eight samples. Staining was weak and is associated with the change in nuclear polarization. The total score ranged from 2 to 3. In one case, areas showing squamous metaplasia stained with fascin (Figure 3). Microvessel endothelium stained homogeneously in all samples. Mean microvessel count was 54.62 ± 4.43. Minimal lymphocytic inflammation was seen in one of 28 samples in the non-neoplastic control group.

Localization of fascin in endometrioid carcinoma

Fascin staining in tumoral stroma was seen in seven of 43 samples (16%). The total stromal fascin scores ranged from 2 to 3. The epithelial portion of tumor stained with fascin in 32 of 43 samples (74%). The total score ranged

g. 1

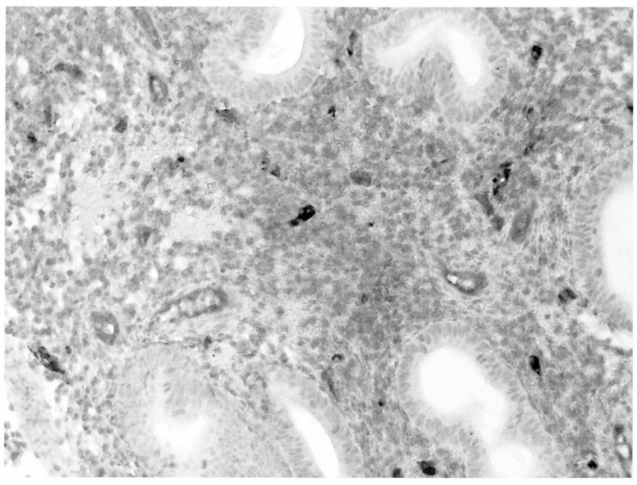
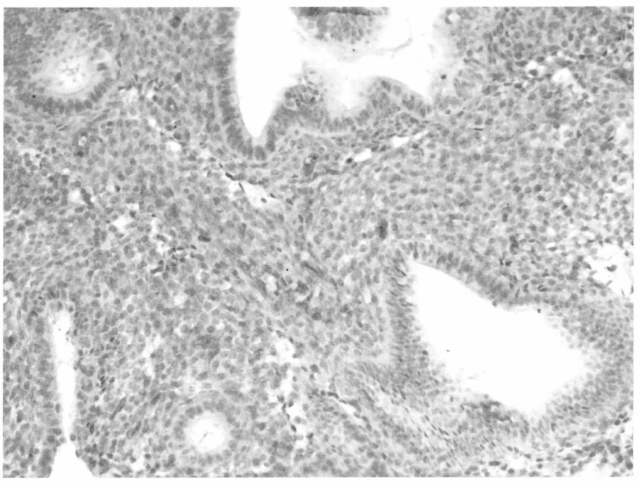


Fig. 2



g. 3

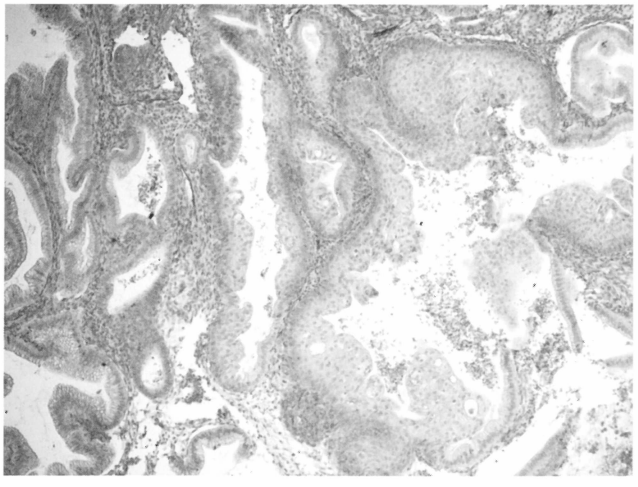


Fig. 4

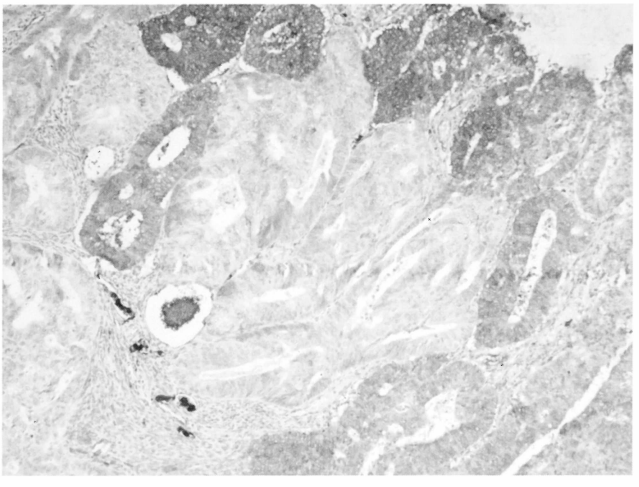


Figure 1. — Proliferative endometrium. Diffuse and homogeneous fascin staining of stroma and microvessels (fascin immunohistochemistry x 400).
Figure 2. — Simple hyperplasia. Stroma and microvessels stained positively for fascin, whereas glandular epithelium did not stain (fascin immunohistochemistry x 400).
Figure 3. — Complex hyperplasia. Positive fascin staining in stroma, microvessels and basal portion of metaplastic squamous epithelium (fascin immunohistochemistry x 400).
Figure 4. — Endometrioid carcinoma. Glandular epithelium partially stained for fascin. Tumor cells in a lymphatic vessel and a few dendritic cells in stroma intensely stained for fascin (fascin immunohistochemistry x 400).

from 2 to 5 (Figure 4). Eleven carcinoma samples (25%) did not stain with fascin. Microvessel endothelium stained homogeneously in 35 samples (81%) and heterogenously in eight (19%) samples. Mean microvessel count was 51.11 ± 3.7. In tumoral stroma fascin-stained dendritic cells were seen in 34 of 43 samples (79%). Metaplasia was observed in 21 (49%) samples and stained with fascin.

Comparison of samples with endometrioid carcinomas and benign endometrial biopsies are given in Table 2. Total epithelial fascin scores of samples were significantly higher in the endometrioid carcinoma group compared to proliferative and hyperplastic endometrium ($p < 0.001$). Stromal fascin labeling was positive in all proliferative and hyperplastic endometrium biopsies whereas 16% of carcinoma samples showed positive stromal fascin labeling which was significant ($p < 0.001$). Higher grade endometrioid carcinoma samples had significantly increased total epithelial fascin scores (.042, $p < 0.05$). There was no statistically significant difference betweeen tumor grade and total stromal fascin expression (.758, $p > 0.05$). Comparison of grade and survival of patients with fascin staining of endometrioid carcinoma patients are given in Tables 3 and 4. The length of follow-up ranged from one month to 97 months. The mean follow-up interval was 34.86 ± 4.24 months. Mean folow-up intervals of grade 1 tumors (n = 6), grade 2 tumors (n = 28), grade 3 tumors (n = 9) in endometrioid carcinoma patients were 61.00 ± 11.55, 29.85 ± 4.73 and 33.00 ± 9.61 months, respectively. There was a significant difference between tumor grade and patient survival (.040, $p < 0.05$). Grade 1 tumors had significant difference in patient outcome compared to grade 2 ($p = .012$) and grade 3 ($p = .050$) tumors. Lymphatic invasion was

Table 2. — *Comparison of samples with endometrioid carcinomas and benign endometrial biopsies.*

Variable	Carcinoma n = 43	Benign endometial biopsies n = 28	p	Test
Total epithelial fascin score:				
no. (%)	32 (74%)	11 (39%)		Mann-
Mean rank	42.95	25.32	.000*	Whitney U
Total stromal fascin score:				
no. (%)	7 (16%)	28 (100%)		Mann-
Mean rank	23.21	55.64	.000*	Whitney U
Fascin staining of microvessels:				Fisher's
no. (%)				Exact
Homogeneous	35 (81%)	28 (100%)	.019*	Chi-square
Heterogeneous	8 (19%)	0		
Mean microvessel count	51.11 ± 3.70	53.03 ± 3.59	.902	T test

*Statistically significant

Table 3. — *Comparison of grade and mean disease-free survival and total epithelial fascin scores in endometriod carcinoma.*

Variable	Endometrioid carcinoma						p
Survival	Mean disease-free survival						.040*[a]
Grade 1 (n = 6)	61.00 ± 11.55						
Grade 2 (n = 28)	29.85 ± 4.73						
Grade 3 (n = 9)	33.00 ± 9.6						
Survival in grade 1 tumors	Mean difference						
Grade 2 tumors	31.14 ± 11.82						.012*
Grade 3 tumors	28.00 ± 13.84						.050*
Total epithelial fascin score (ranged 0-5)	0	1	2	3	4	5	.42*[b]
Grade 1 (n = 6)	0	0	4	0	2	0	
Grade 2 (n = 28)	6	0	9	11	1	1	
Grade 3 (n = 9)	4	1	1	2	1	0	

*Statistically significant; a: ANOVA; b: Exact Pearson's chi-square test.

Table 4. — *Comparison of histopathologic findings and patient outcome in endometrioid carcinoma.*

Disease-free survival	Endometrioid carcinoma			r	p
Lymphovascular space involvement	*Mean survival (months)*				
Presence: 34	34.23 ± 4.75				.778[a]
Absence: 9	37.22 ± 9.90				
Mean microvessel count	Pearson's correlation			.412	.006*
Local cellular immune response (Dendritic cells & inflammatory cells)					
Absence: 9	35.88 ± 8.06				
Mild: 10	28.70 ± 9.59				
Moderate: 15	43.73 ± 8.72				
Intense: 9	25.88 ± 4.07				
Overall survival (n = 22)		DOD no. = 12	Alive no. = 10		
Local cellular immune response	negative	3	2		.667[c]
	+	4	1		
	++	4	6		
	+++	1	1		
Total epithelial fascin scores					
Mean rank					.180[d]
	13.04	9.65			
Total epithelial fascin scores	*Pearson Correlation*			-.089	.569

*Statistically significant; **DOD: died of disease; a: T test; b: ANOVA; c: Pearson chi-square; d: Mann-Whitney U test.

observed in 79% (34 of 43) of samples. There was no difference in patient outcome with presence or absence of vascular invasion (p = .778). Local inflammatory immune response (dendritic cells and lymphocytic infiltration) was observed in 34 of 43 samples (79%). Inflammatory response was mild in ten samples, moderate in 15 samples and intense in nine samples. There was no significant difference in disease-free survival and overall survival with presence or absence of local cellular immune response and its severity (.677, $p > 0.05$). Mean microvessel count of the non-neoplastic and neoplastic group were 53.035 ± 3.59 and 51.116 ± 3.70, respectively. There was no significant difference in terms of mean microvessel count between the two groups (.902, $p > 0.05$). Disease-free survival moderately correlated with mean microvessel count in endometrioid carcinoma patients (r = .412, p =.006). In the proliferative and hyperplastic endometrial biopsies microvessels stained homogeneously in all samples (28/28), but in the endometrioid carcinoma group, eight of 43 samples showed heterogenous fascin staining of microvessels. The difference between the two groups was significant (.019, $p < 0.05$).

Discussion

Among all cancers in women, endometrial carcinoma ranks fourth behind cancers of the breast, colorectum and lung. A number of constitutional factors have been generally identified in women who develop endometrial cancer, including obesity, diabetes, hypertension, nulliparity and late menopause which suggest an underlying endocrine-metabolic disorder. Worldwide, the variable incidence of endometrial cancer is most strongly associated with total fat consumption. Most of the metabolic abnormalities observed in endometrial carcinoma patients are related to age and obesity. The risk of endometrial carcinoma is also related to the chronicity of unopposed estrogen exposure, as well as to the level of circulating estrogens [14, 15]. We observed a statistically significant difference among the fascin labeling pattern of normal proliferative endometrium, hyperplastic endometrium and endometrioid cancer. The fascin effect on endometrium may be related to the estrogen level. Unopposed estrogen stimulus decreases the amount of endometrial stroma, whereas it enhances proliferation of endometrial glandular cells. We demonstrated that proliferative and hyperplastic endometrial stroma had strong fascin labeling whereas neoplastic endometrial stroma had decreased fascin immunoreactivity ($p < 0.001$). Inversely, it was shown that as the development of neoplasia epithelial fascin expression up-regulated ($p < 0.001$). Endometrium and endometrial cancers can be a good model to research the molecular mechanisms which up-regulate or down-regulate the fascin.

Local immune/inflammatory cell reaction which is not found to be correlated with survival and age of patients is a common finding in endometrial carcinomas [16, 17]. However, inflammatory cell reaction contributes greatly to cell-matrix interaction. Actin-regulatory proteins, such as fascin, direct the site-specific assembly and disassembly of actin filaments, which are directly involved in cell motility, cell-cell and cell-matrix contacts, and the establishment and maintenance of cell morphology [18]. Specifically, fascin colocalizes with actin in stress fibers and cellular protrusions, structures that require strong mechanical

support [1-4]. We observed that homogeneous and intense fascin immunoreactivity is seen in normal endometrial stroma and microvessels and fascin-actin interaction supports normal functions of endometrium.

β-catenin, which is a cytoplasmic partner of E-cadherin, links cadherins to α-catenin and actin cytoskeleton and mediates strong cell-cell adhesion in the adherens junction. β-catenin is an important element of the Wnt (adenomatous polyposis coli/T cell factor/catenin) signal transduction pathway which has been implicated in embryogenesis and carcinogenesis, including the development of endometrial carcinomas. It was demonstrated that low-grade endometrioid adenocarcinomas showed strong β-catenin and weak E-cadherin expression. β-catenin expression is strongly associated with the histologic subtype [19-21]. Recent studies suggest that fascin's association with β-catenin is related to the microenvironment of the neoplasm and not to the gene function [22].

The role of fascin in generating and maintaining a dynamic tumorigenic phenotype has been supported by recent studies. Wong [23] and Guan *et al.* [24] demonstrated that glucocorticoids down-regulate fascin and TGF-α reverses the steroid induced down-regulation of fascin and abrogates the glucorticoid stimulation of tight junction formations in rat mammary epithelial tumor cells. Jawhari *et al.* [22] showed that up-regulation of fascin is associated with aggregation of growing cells and results in glandular differentiation as manifested by polarization of nuclei toward the basal surface of cells and the organization of cells around a central lumen in colonic carcinoma cell lines. Fascin overexpression has been demonstrated in various tumors [5-12]. Goncharuk *et al.* [13] demonstrated that local aggressive and rarely metastatic skin cancers such as basal cell carcinoma were associated with diffuse and intense fascin immunoreactivity, whereas those cancers with risk of metastasis such as melanoma were associated with weak or down-regulated fascin expression. Down-regulation of fascin and loss of cell-cell, cell-matrix adhesions have also had an important role in malignant tumor progression in some tumors, such as melanomas. Shonukan *et al.* [25] demonstrated that there are high levels of neurotrophin expression in the normal tissue adjacent to brain metastases of melanoma. It was suggested that interaction between fascin and neurotrophin provides a direct link between the NGF signaling pathway and neurotrophin-mediated melanoma cell movement by down-regulation (dephosphorilation) of fascin. The TNF/NGF pathway is associated with alterations in the regulation of apoptosis and resistance of tumor cells to therapy [26]. In this study, we demonstrated that neoplastic endometrial stroma had a low level of fascin immunoreactivity, whereas fascin expression up-regulated in neoplastic epithelium and higher grade endometrioid carcinoma samples had a significantly high total epithelial fascin score (.042, $p < 0.05$). Our study supports the importance of actin bundling protein fascin in generating and maintaining a dynamic tumorigenic phenotype in endometrial neoplasms.

The second aim of this study was to investigate the role of fascin in endothelial cell migration and angiogenesis in normal and hyperplastic endometrium, and endometrioid carcinoma. The induction of angiogenesis was mediated by specific angiogenic molecules released by the tumor and by activated macrophages [27]. Recently, a study from our institution demonstrated the prognostic value of angiogenesis in endometrial carcinoma. In this study, factor VIII-related antigen was used as a marker [28]. Microvessels stained homogeneously with fascin in all samples of proliferative and hyperplastic endometrium. Heterogenous fascin expression of microvessels was found in eight of 43 endometrioid carcinomas. The difference between the two groups was significant (.019, $p < 0.05$). Heterogeneous fascin staining of microvessels was found to be associated with loss of fascin staining in the stroma of tumors despite enchanced epithelial staining. This feature may be explained with de novo microvessel formation in these patients instead of microvessel regeneration and proliferation by endothelial cell migration. Down-regulation of fascin in microvessel endothelium may enchance microcirculatory disturbances in tumor and peritumoral tissue and may have an important role in tumor progression. As far as we know, this is the first study showing the role of fascin in endothelial cell migration and angiogenesis in endometrioid cancers.

Various cell signaling pathways related to endocrine and metabolic functions which are activated by ageing and obesity may have a central role in the progression of endometrioid carcinoma [14, 15].

Our study supported the importance of actin-bundling protein fascin in generating and maintaining a dynamic tumorigenic phenotype and the prognostic significance of fascin in endometrioid carcinoma. Further studies are necessary to reveal the down-regulating molecular mechanisms of fascin in endometrial carcinoma.

References

[1] Adams J.C., Clelland J.D., Collet G.D.M., Matsumura F., Yamashiro S., Zhang L.: "Cell-matrix adhesions differentially regulate fascin phosphorylation". *Mol. Biol. Cell,* 1999, *10*, 4177.

[2] Cohan C.S., Welnhofer E.A., Zhao L., Matsumura F., Yamashiro S.: "Role of the actin bundling protein fascin in growth cone morphogenesis: localization in filopodia and lamellipodia". *Cell. Motil. Cytoskeleton,* 2001, *48*, 109.

[3] Duh F.M., Latif F., Weng Y. *et al.*: "cDNA cloning and expression of the human homolog of the sea urchin fascin and Drosophila signed genes which actin-bundling protein". *DNA Cell Biol.,* 1994, *13,* 821.

[4] Ishikawa R., Yamashiro S., Kohama K., Matsumura F.: "Regulation of actin binding and actin bundling activities of fascin by caldesmon coupled with tropomyosin". *J. Biol. Chem.,* 1998, *273*, 26991.

[5] Grothey A., Hashizume R., Ji H. *et al.*: "c-erbB-2/Her-2 upregulates fascin, an actin-bundling protein associated with cell motility, in human breast cancer cell lines". *Oncogene,* 2000, *19*, 4864.

[6] Guvakova M.A., Boettiger D., Adams J.C.: "Induction of fascin spikes in breast cancer cells by activation of the insulin-like growth factor-1 receptor". *Int. J. Biochem. Cell Biol.*, 2002, *34*, 685.

[7] Hu W., McCrea P.D., Deavers M., Kavanagh J.J., Kudelka A.P., Verschraegen C.F.: "Increased expression of fascin, motility associated protein, in cell cultures derived from ovarian cancer and in borderline and carcinomatous ovarian tumors". *Clin. Exp. Metastasis*, 2000, *18*, 83.

[8] Maitra A., Iacobuzio-Donahue C., Rahman A. *et al.*: "Immunohistochemical validation of a novel epithelial and a novel stroma marker of pancreatic ductal adenocarcinoma identified by global express microarrays: sea urchin fascin homolog and heat shock protein 47". *Am. J. Clin. Pathol.,* 2002, *118*, 52.

[9] Pelosi G., Pastorino U., Pasini F. *et al.*: "Independent prognostic value of fascin immunoreactivity in Stage I non small cell lung cancer". *Br. J. Cancer,* 2003, *88*, 537.

[10] Pelosi G., Barisella M., Pasini F. *et al.*: "CD117 immunoreactivity in Sage I adenocarcinoma and squamous cell carcinoma of the lung: relevance to prognosis in a subset of adenocarcinoma patients". *Mod. Pathol.,* 2004, *17*, 711.

[11] Pelosi G., Pasini F., Sonzogni A. *et al.*: "Prognostic implications of neuroendocrine differentiation and hormone production in patients with Stage I non small cell lung carcinoma". *Cancer,* 2003, *97*, 2487.

[12] Kempf W., Levi E., Kamarashev J. *et al.*: "Fascin expression in CD30-positive cutaneous lymphoproliferative disorders". *J. Cutan. Pathol.*, 2002, *29*, 295.

[13] Goncharuk V.N., Ross J.S., Carlson J.A.: "Actin-binding protein fascin expression in skin neoplazi". *J. Cutan. Pathol.,* 2002, *29*, 430.

[14] Mutch D.G. Uterine cancer. In: Scott J.R., Gibbs R.S., Karlan B.Y., Haney A.F. (eds.), Danforth's Obstetrics and Gynecology, Philadelphia: Lippincott Williams &Wilkins, 2003, 951.

[15] Yen S.S.C.: "Neuroendocrinology and reproduction". In: Straus J.F. III, Barbieri R.L. (eds.), Yen and Jaffe's Reproductive Endocrinology. Philadelphia: Elsevier Saunders, 2004, 3.

[16] Silverberg S.G., Sasano N., Yajima A.: "Endometrial carcinoma in Miyagi Prefecture, Japan: histopathologic analysis of a cancer registry-based series and comparison with samples in American women". *Cancer,* 1982, *49*, 1504.

[17] Dundar E., Tel N., Ozalp S.S., Isiksoy S., Kabukcuoglu S., Bal C.: "The significance of local cellular immune response of women 50 years of age and younger with endometrial carcinoma". *Eur. J. Gynaecol. Oncol.*, 2002, *23*, 243.

[18] Puius Y.A., Mahoney N.M., Almo S.C.: "The modular structure of actin regulatory proteins". *Curr. Opin. Cell Biol.,* 1998, *10*, 23.

[19] Schlosshauer P.W., Ellenson L.H., Soslow R.A.: "β-catenin and E-cadherin expression patterns in high grade endometrial carcinoma are associated with histological subtype". *Mod. Pathol.*, 2002, *15*, 1032.

[20] Palacios J., Catasus L., Moreno-Bueno G.M., Matias-Guiu X., Prat J., Gamallo C.: "β-and γ-catenin expression in endometrial carcinoma. Relationship with clinicopathological features and microsatellite instability". *Virchows Arch.,* 2001, *438*, 464.

[21] Moreno-Bueno G., Gamallo C., Perez-Gallego L., de Mora J.C., Suarez A., Palacios J.: "β-catenin expression pattern, γ-catenin gene mutations and microsatellite instability in endometrioid ovarian carcinomas and synchronous endometrial carcinomas". *Diag. Mol. Pathol.,* 2001, *10*, 116.

[22] Jawhari A.U., Buda A., Jenkins M. *et al.*: "Fascin, an actin bundling protein, modulates colonic epithelial cell invasiveness and differentiation in vitro". *Am. J. Path.,* 2003, *162*, 69.

[23] Wong V., Ching D., McCrea P.D., Firestone G.L.: "Glucocorticoid down-regulation of fascin protein expression is required for steroid-induced formation of tight junctions and cell-cell interactions in rat mammary epithelial tumor cells". *J. Biol. Chem.,* 1999, *274*, 5443.

[24] Guan Y., Woo P.L., Rubenstein N.M., Firestone G.L.:"Transforming growth factor-α abrogates the glucocorticoid stimulation of tight junction formation and reverses the steroid-induced down-regulation of fascin in rat mammary epithelial tumor cells by a Ras-depentent pathway". *Exp. Cell Res.*, 2002, *273*, 1.

[25] Shonukan O.T., Bagayogo I., McCrea P.D., Chao M., Hempstead B.: "Neurotropin-induced melanoma cell migration is mediated through actin-bundling protein fascin". *Oncogene*, 2003, *22*, 3616.

[26] Müller M., Kramer P.H.: "Integrated cell function: apoptosis". In: Arias I.M., Boyer J.L., Chisari F.V., Fausto N., Schachter D., Shafritz D. (eds.). The Liver Biology and Pathobiology, Philadelphia: Lippincott Williams &Wilkins, 2001, 187.

[27] Wagatsuma S., Konno R., Sato S., Yajima A.: Tumor angiogenesis, hepatocyte growth factor, and c-Met Expression in endometrial carcinoma. *Cancer,* 1998, *82,* 520.

[28] Ozalp S., Yalcin O.T., Acikalin M., Tanir H.M., Oner U., Akkoyunlu A.: "Microvessel density (MVD) as a prognosticator in endometrial carcinoma". *Eur. J. Gynaecol. Oncol.*, 2003, *24*, 305.

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