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The Relation between Mixed Mesodermal Tumors and Adenocarcinomas of the Ovary. An Immunopathologic Study

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Abstract—*Mixed mesodermal tumor of the ovary is a rare neoplasm. The diagnosis is made if an ovarian carcinoma contains heterologous sarcomatous differentiation. With the help of an immunopathologic stain for desmin, the intermediate filament protein of muscle cells, twenty cases of ovarian adenocarcinoma were examined for the presence of morphologically unrecognizable heterologous tumor cells. Six of twenty tumors were positive. There was no significant correlation between a number of clinical parameters and desmin positivity. The authors conclude that mixed mesodermal tumor of the ovary may not be a separate entity. Instead, there may be a single group of ovarian epithelial tumors ranging from borderline or well-differentiated carcinomas to poorly-differentiated tumors which may show homologous (carcinosarcoma) or heterologous (mixed mesodermal tumor) differentiation.*

INTRODUCTION

MIXED mesodermal tumors (MMT) and carcinosarcomas of the ovary are considered to be rare tumors [1, 2]. These diagnoses can be made, if in addition to adenocarcinomatous elements, sarcomatous elements are present in an ovarian tumor. These elements may be homologous (carcinosarcomas) or heterologous (MMT) [3].

Ramaekers *et al.* reported recently a case of MMT which could be diagnozed only by intermediate filament protein analysis [4]. In their case, sarcomatous elements had not been recognized by routine light or electron microscopic studies in the primary ovarian localization. With the help of an antidesmin antibody it appeared that the primary tumor had desmin-containing cells. This showed a heterologous sarcomatous differentiation in the tumor, establishing the diagnosis of MMT. The metastases of the tumor showed morphologically recognizable rhabdomyosarcoma.

In the present study we examined 20 ovarian adenocarcinomas for the presence of tumor cells which contained desmin, the muscle-specific type of intermediate filament [5]. We also performed stains for keratin and vimentin and examined twenty endometrial adenocarcinomas and four endocervical adenocarcinomas with stains for the same intermediate filaments. The results are compared to a number of clinical parameters, and suggest that ovarian adenocarcinomas may form a continuous spectrum with carcinosarcomas and mixed mesodermal tumors.

MATERIALS AND METHODS

Twenty cases of ovarian adenocarcinoma, 20 cases of endometrial adenocarcinoma and four cases of endocervical adenocarcinoma, diagnosed in the years 1982–1985, were retrieved from the files of the SSDZ. From these cases, routine formalin-fixed, paraffin-embedded and H- and E-stained sections were reviewed. The tumors were classified according to the WHO classification [3] and graded [6]. Per tumor, one or two sections, showing the highest grade, were selected. Extra sections were cut from the matching blocks and stained with anti-intermediate filament antibodies. The anti-intermediate filament staining technique was performed as follows: 6 μm sections were cut and mounted on chicken protein-coated slides. The sections were deparaffinized, rehydrated, washed in distilled water and treated with 0.3% H_2O_2 in water for 5 min. The sections were washed in PBS for 10 min, preincubated with 0.1% Bovine Serum Albumin (BSA) in PBS for 10 min and then incubated with anti-desmin for 90 min at 37° C. The sera used were polyclonal rabbit anti-intermediate filament antibodies obtained from Eurodiagnostics

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(Apeldoorn, The Netherlands; Desmin lot No. 005, Vimentin lot No. 011, Keratin lot No. 007) in a 1 : 25 dilution in PBS with 0.1% BSA. The production and characterization of these antibodies have been described in detail elsewhere [5]. After 10 min washing in PBS and 10 min preincubation with 0.1% BSA in PBS, the sections were incubated with swine anti-rabbit-peroxidase (Dako, Glostrup, Denmark lot No. 101, dilution 1 : 80) for 30 min and washed in PBS for 20 min.

Color was developed using AEC for 10–30 min. The sections were washed in distilled water and counterstained with haematoxylin. For negative controls, the primary antibody was omitted. A colonic adenocarcinoma was used as keratin-positive control. Internal positive controls for desmin and vimentin were present in all sections (muscle cells of blood vessels and desmin-containing ovarian stroma cells for desmin; other stroma cells for vimentin).

Anti-intermediate filament staining was considered positive if a red precipitate, completely contained within cell boundaries, was observed in cytologically malignant cells. Patient data concerning number of children, number of years after menopause, duration of symptoms and follow-up, were obtained from the patients' medical records.

RESULTS

All 20 cases of ovarian adenocarcinoma, all 20 cases of endometrial adenocarcinoma and all 4 cases of endocervical adenocarcinoma were positive for keratin. Ten cases of ovarian adenocarcinoma (50%), 19 cases of endometrial adenocarcinoma (95%) and 2 cases of endocervical adenocarcinoma (50%) were positive for vimentin. All cases of endometrial and endocervical adenocarcinoma were desmin-negative (see Table one).

Six of 20 cases of ovarian adenocarcinoma showed cells which expressed desmin-intermediate filament (see Fig. 1a and b). Positivity ranged from a few single cells (cases 16 and 17) to multiple groups of tumor cells (cases 15 and 19). Desmin-expressing cells were seen exclusively in poorly differentiated areas of the investigated tumors. The average age of patients with anti-desmin positive tumors was 61.5 years, as compared to 63.4 years for the anti-desmin negative group ($P = 0.93$). There was no significant difference in grade ($P = 0.29$), stage ($P = 0.62$), degree of nulliparity ($P = 0.17$), duration of symptoms ($P = 0.84$) or number of years after menopause ($P = 0.70$) between the two groups of patients.

Six of 14 patients with desmin-negative tumors died of the disease, with a mean survival of 3.5 months. The other 8 patients have a mean follow-up of 18.6 months. One of the 6 patients with desmin-positive tumors died 7 months after diagnosis. The other 5 patients have a mean follow-up of 6.8 months (see Table 2).

In the new sections made for immunopathologic staining, we found a nest of clearly malignant cartilage in one of our cases (No. 18). Elsewhere this tumor also contained desmin-positive cells. The cartilage had not been present in the sections on which the original diagnosis was made. This observation confirmed our diagnosis of MMT.

DISCUSSION

Mesodermal mixed tumors (MMT) of the ovary are reported to be a rare but distinct type of tumor, occurring in older women with a high incidence of nulliparity. These tumors are reported to be in an advanced stage when first diagnosed and to have a poor survival rate [7]. Some authors have noted an association with endometriosis [8, 1]. Azoury [9] considered this tumor to be of paramesonephric origin, while others believe that these tumors arise from the pluripotential epithelial cells of the ovary [7, 8]. This epithelium is also considered to be the source of other types of ovarian adenocarcinomas [4, 7].

Mesodermal mixed tumors are defined histopathologically by the presence of morphologically identifiable sarcoma in association with carcinoma. In the present study, we have extended these criteria to include the immunohistologic demonstration of tumor cells which express a protein characteristic of benign and malignant muscle cells, the intermediate filament desmin. We consider this extended criterium valid for the following reasons.

The differentiation of a cell can be investigated in different ways. One way is to look at its morphology, another way to analyse its proteins. Intermediate filament proteins are tissue-specific pro-

Table 1. Intermediate filament expression of gynecological adenocarcinomas

Tumor cells expressing	Ovarian adenocarcinomas	Endometrial adenocarcinomas	Endocervical adenocarcinomas
Keratin	20/20	20/20	4/4
Vimentin	10/20	19/20	2/4
Desmin	6/20	0/20	0/4

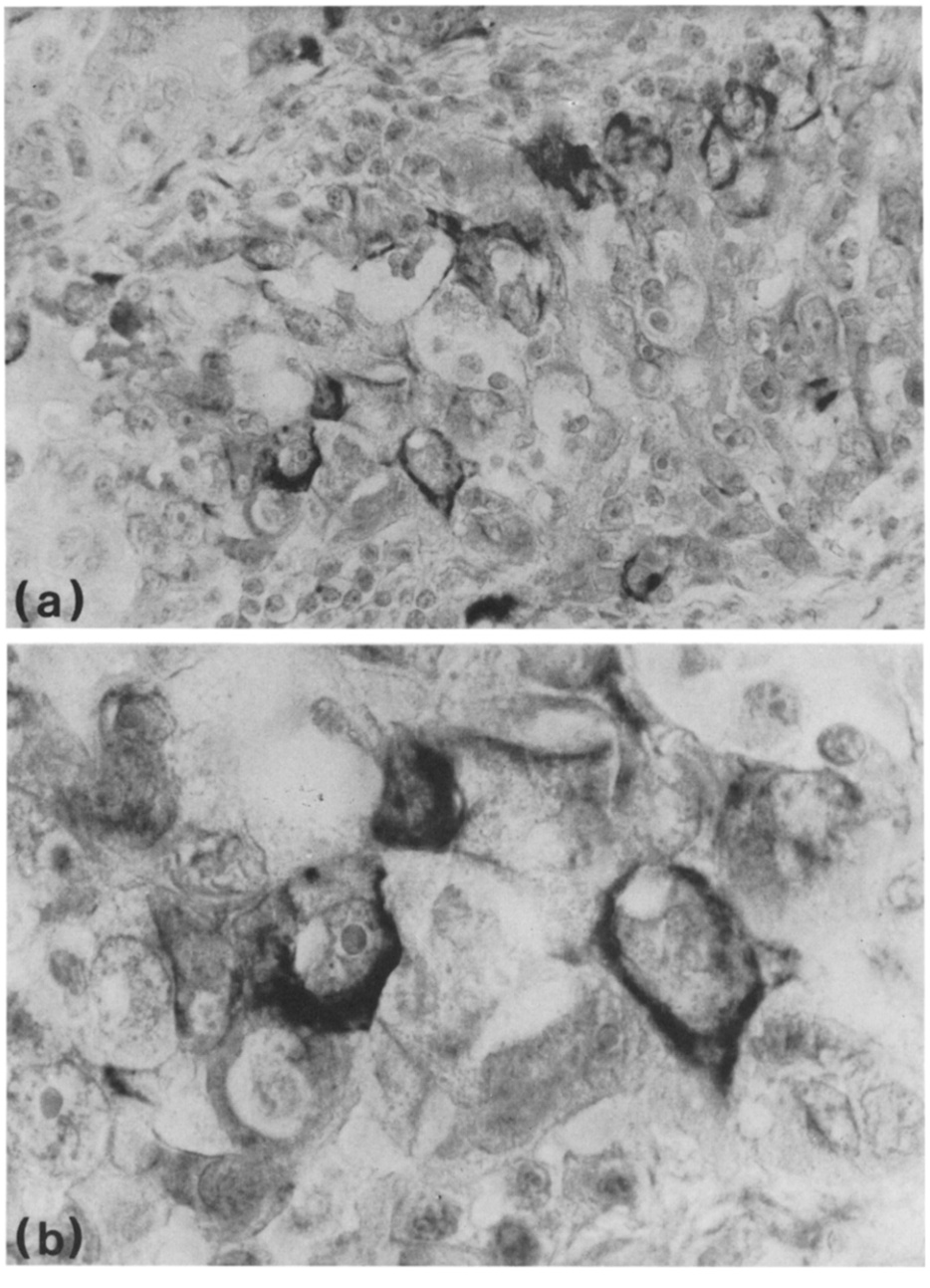

Fig. 1(*a*). *Tumor cells expressing desmin, the intermediate filament of muscle cells, in a poorly-differentiated part of an ovarian adenocarcinoma* (*case No.* 15. *indirect immunoperoxidase, original magnification* × 250).
(*b*) *Detail, showing cytologically malignant characteristics of desmin-expressing cells* (*original magnification* × 1000).

Table 2. Clinical and pathological features of 20 patients with ovarian tumors

No.	Age	Type of adeno-carcinoma *	Grade	Figo stage	Tumor cells expressing desmin	Tumor cells expressing Vimentin	Number of children	Years after menopause	Duration of symptoms (months before diagnosis)	Current status	Follow up (months)
1	68	S	2–3	III	–	+	7	22 jr.	N.A.	dead	1
2	58	U	3	Ic	–	–	0	4 jr.	1	alive	32
3	58	S	1–2	IV	–	–	2	7 jr.	1.5	dead	7
4	63	E	1–2	IV	–	+	3	11 jr.	1	dead	1
†5	71	S	2	Ia	–	+	0	20 jr.	16	alive	30
6	52	M	1–2	Ia	–	–	1	2 jr.	3	alive	15
7	30	M	1	I	–	–	0	0 jr.	N.A.	alive	30
8	82	U	3	IIb	–	–	4	31 jr.	12	dead	5
9	57	S	2	III	–	–	2	13 jr.	1	dead	4
10	77	S	1	Ia2	–	–	0	27 jr.	12	alive	12
11	61	S	1	III	–	+	1	6 jr.	3	alive	12
12	86	U	3	IV	–	–	3	N.A.‡	1	dead	1
13	56	S	2–3	III	–	–	4	3 jr.	N.A.	alive	10
14	54	S	2–3	III	–	–	5	5 jr.	N.A.	alive	8
15	60	S	2–3	III	+	+	4	N.A.	1	dead	7
16	63	S	2–3	III	+	+	2	10 jr.	N.A.	alive	11
17	51	S	1–2	IIc	+	+	1	2 jr.	4	alive	12
18	68	S	2–3	III	+	+	2	16 jr.	2.5	alive	7
19	63	E	2–3	III	+	+	2	10 jr.	1	alive	2
20	64	U	3	III	+	+	3	4 jr.	8	alive	2

*S: serous.
M: Mucinous.
E: Endometrioid.
U: Undifferentiated.
†An endometrium carcinoma was also present.
‡N.A.: Data not available.

teins [5]. Most tissues express only one type of intermediate filament. Coexpression of keratin and vimentin is known to occur in malignant ovarian epithelial neoplasms [10, 11], in renal carcinomas [11, 12] endometrial carcinomas and thyroid carcinomas [11]. In these tumors expression of vimentin is generally not regarded as sarcomatous differentiation. We agree with this because in renal and thyroid neoplasms, a carcinosarcomatous component, comparable to carcinosarcoma or MMT of the endometrium or ovary is not recognized [13,14]. For endometrial and ovarian neoplasms, expression of vimentin cannot be regarded as sarcomatous differentiation because coexpression of keratin and vimentin is present in normal ovarian surface epithelium [10] and normal endometrial epithelium [11].

However, to our knowledge, expression of desmin and keratin, in the same tumor, has not been reported before. Therefore, and because desmin is not present in normal ovarian surface epithelium, we think this expression of desmin should be regarded as myosarcomatous differentiation. This interpretation is supported by the fact that myosarcoma is the second most frequently found heterologous mesenchymal differentiation in MMT [6] and by the desmin-positive tumor (case No. 18) which showed malignant cartilage in newly-cut sections.

Ten of 20 ovarian adenocarcinomas and 19 of 20 endometrial adenocarcinomas were positive for vimentin, which is consistent with the literature [11]. To our surprise, no desmin-expressing tumor cells were found in the endometrial adenocarcinomas. Perhaps the relation between ovarian MMT and adenocarcinoma, on the one hand, and endometrial MMT and adenocarcinoma on the other, is not the same. Another explanation may be that myosarcomatous differentiation is easier to recognize morphologically in endometrial adenocarcinomas than in ovarian adenocarcinomas.

Six of 20 ovarian tumors initially diagnosed as adenocarcinomas contained morphologically malignant cells with immunohistochemically demonstrable desmin, a chemical marker of myosarcomatous differentiation. At least two possible conclusions can be drawn from these results. The first is that a number of ovarian adenocarcinomas may be classified by immunohistochemical criteria as MMT, because they contain desmin-positive tumor cells. This means that they show, by immunohistochemical criteria, heterologous sarcomatous

differentiation. In our data there is no significant difference between the patients with desmin-positive tumors and the patients with desmin-negative tumors as regards age distribution, degree of parity, duration of symptoms or survival. This means there is no support from clinical evidence to regard these desmin-positive tumors as a separate entity.

It may then follow that ovarian tumors form a single unified spectrum arising from the ovarian epithelium. At one end of the spectrum of these tumors are the borderline and well-differentiated tumors and at the other end the poorly differentiated tumors including tumors with sarcomatous differentiation, the carcinosarcomas and MMT. In this context it is not difficult to imagine that, with the aid of immunopathologic stains, more tumors will show sarcomatous differentiation than is seen with routine histology alone. Further support for this concept comes from the fact that desmin-positive cells in our material were always located in the poorly-differentiated parts of the tumors.

Because of limited follow-up and a limited number of patients, further work is needed to clarify this unifying concept of epithelial ovarian tumors. However, this concept could lead to a better understanding of ovarian tumorigenesis as well as to a refined grading with a better therapeutic and prognostic value.

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