

Uterine Adenocarcinoma with Prominent Desmoplasia in a Geriatric Miniature Pig

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ABSTRACT. A 10-year-old miniature sow was died, showing inappetence and weight loss. Grossly, neoplastic enlargement of the uterus was found. Histopathologically, the lesions consisted of acinar, ductular and cystic proliferations of mono- and multilayered epithelial cells; these cells reacted immunohistochemically strongly with three different cytokeratin antibodies, and occasionally to vimentin. Myofibroblastic desmoplastic cells, positive to α -smooth muscle actin, were present among neoplastic cells. Metastatic lesions were seen in the lungs and liver. Based on these findings, a diagnosis of uterine adenocarcinoma with marked desmoplasia was made. This case is the second report of uterine adenocarcinoma in the miniature pig.

KEY WORDS: desmoplasia, miniature pig, uterine adenocarcinoma.

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Uterine adenocarcinomas are uncommon neoplasms in most animals with the exception in rabbits and cattle [6, 13]. Woman cases are the most common invasive cancers of the genital tract [3]. To our knowledge, there are two cases of uterine adenocarcinomas reported in English so far in sow; a 3-year-old crossbred Chester White-Hampshire-Duroc sow without metastases [15], and a 16-year-old Vietnamese pot-bellied pig with visceral metastases [5]. Detailed pathological examinations such as immunohistochemistry were not made for these cases. We encountered a sow bearing a uterine adenocarcinoma. Because uterine adenocarcinoma is extremely rare in sow [15], in the present study we examined histopathological and immunohistochemical characteristics of this case.

A 10-year-old sow (47.3 kg body weight) of the miniature pig strain, kept in the Adventure World Zoo (Wakayama, Japan) as an exhibit, was found dead after several weeks of inappetence and gradual deterioration of body condition. The sow had never farrowed. At necropsy, the right horn of the uterus was markedly enlarged with irregularly small nodular lesions, whitish to yellow in color and up to 1 cm in diameter, in the thickened wall (Fig. 1), and the endometrium had variously-sized cysts containing brownish fluid. Multiple nodules, up to 2 cm in diameter and pale tan or yellow in color, were found in the lungs and liver (Fig. 2). The right kidney was contracted. Tissues from the uterus, lungs, liver, ovaries and kidneys were fixed in 10% neutral buffered formalin, embedded in paraffin, cut at 4 μ m in thickness and stained with hematoxylin and eosin (HE); some sections were stained with alcian blue (pH 2.5), Azan-Mallory and periodic acid-Schiff (PAS) stains. For immunohistochemistry, following primary monoclonal antibodies were used: anti-pan-cytokeratin (CK) (predilution; Dako,

Carpinteria, CA, U.S.A.), anti-CK19 (1:100; Novocastra Laboratories Ltd., Newcastle, U.K.), anti-AE1/AE3 (predilution; Dako), anti-vimentin (1: 400; Dako) and anti- α -smooth muscle actin (α -SMA) (1:100; Dako). Then, these slides incubated with the primary antibody were reacted with a horseradish peroxidase-conjugated anti-mouse secondary antibody (Histofine simplestain MAX-PO(M) kit; Nichirei, Tokyo, Japan), and positive reactions were visualized with 3, 3'-diaminobenzidine as chromogen. Mouse non-immune serum instead of the primary antibodies served as negative control. Sections were counterstained lightly with hematoxylin.

Histopathologically, neoplastic proliferation of epithelial cells, arranged in acinar, ductular, cystic and solid growth patterns, was seen diffusely or focally in the uterus, mainly in the endometrium. The acinar, ductular and cystic structures were rimmed by irregularly mono- or multilayered, flattened, cuboidal or columnar cells; these neoplastic cells showed prominent cellular pleomorphism and lost polarity, being sometimes desquamated into the lumina (Fig. 3). Furthermore, the neoplastic cells had nuclei with irregularly dispersed or condensed chromatin and prominent nucleoli, apparently eccentric nuclei. Mitotic figures were occasionally seen. Some neoplastic cells had vacuoles in the cytoplasm, reacting positively to PAS and alcian blue stains; this indicated mucin production. Similar neoplastic proliferation was also seen deep in the thickened wall (smooth muscle layers) of the uterus and ovaries, indicating invasive growth into surrounding tissues. Metastatic lesions, seen in the lungs and liver as multiple nodules, exhibited the same morphologic features of neoplastic cells as seen in the uterus. Necrosis and dystrophic calcification were sporadically seen in the uterus and ovaries. No squamous differentiation of neoplastic cells was seen in any lesions. Immunohistochemically, neoplastic cells gave a strong positive reaction to three different anti-CK antibodies (pan-CK, CK19 and AE1/AE3) (Fig. 4), and occasional neoplastic cells reacted to vimentin (Fig. 5). Intriguingly, collagen

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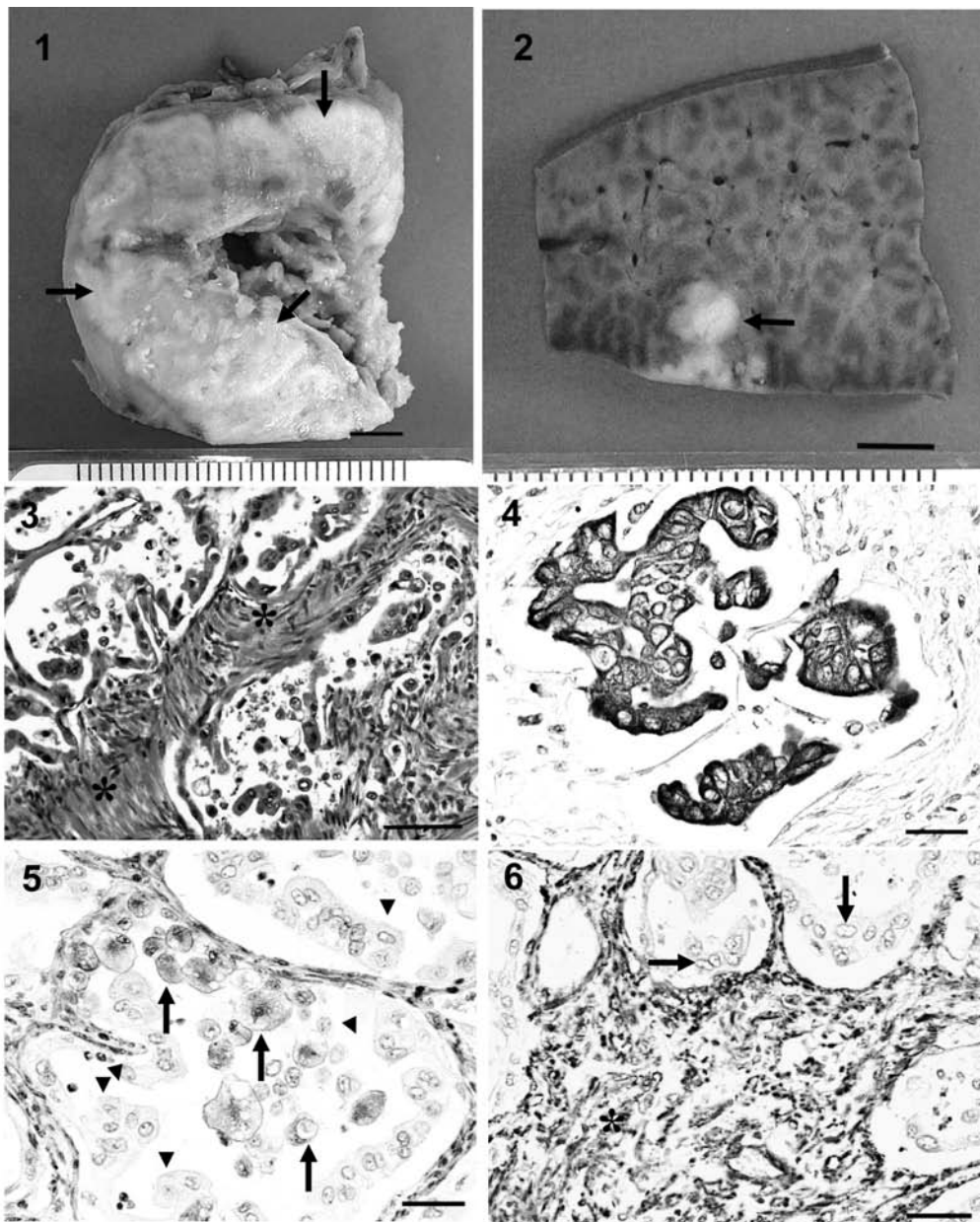


Fig. 1. Neoplastic enlargement of the uterus in a geriatric sow, with small whitish nodular lesions (arrows) on the cut surface of the wall. Bar = 0.5 cm.

Fig. 2. Whitish metastatic lesions (arrow) seen in the cut surface of the liver. Bar = 0.5 cm.

Fig. 3. Neoplastic epithelial cells, arranged as an irregularly lining layer or desquamated in the lumen, in the desmoplastic tissues (asterisks) in uterus; cellular pleomorphism and nuclear atypia are seen in these neoplastic cells. HE stain. Bar = 100 μ m.

Fig. 4. Multilayered, cuboidal neoplastic epithelial cells forming irregular acini seen in the uterus; these cells react strongly to cytokeratin (AE1/AE3). Immunohistochemistry. Bar = 50 μ m.

Fig. 5. Acini rimmed by neoplastic epithelial cells seen in the lung as a metastatic lesion. Some neoplastic epithelial cells react to vimentin (arrows), but there are also neoplastic cells which do not react to vimentin (arrow-heads). Immunohistochemistry. Bar = 50 μ m.

Fig. 6. Desmoplastic tissue in a metastatic lesion in the lung having many spindle cells reacting to α -smooth muscle actin, indicating the presence of myofibroblasts (asterisk). Arrows indicate neoplastic epithelial cells forming acini. Immunohistochemistry. Bar = 50 μ m.

fiber proliferations, so-called desmoplasia, were often seen around neoplastic cells in the primary and metastatic lesions; such desmoplastic areas contained acinar and ductular structures or sporadic solitary growth of neoplastic cells, giving an appearance of scirrhous carcinoma. The desmoplastic areas were stained blue by the Azan-Mallory methods, and spindle cells present in these areas reacted to vimentin and α -SMA (Fig. 6), suggestive of myofibroblastic nature of desmoplastic cells [14]. Single neoplastic cell or clusters of neoplastic cells forming ambiguous acini were detected in the lymphoid vessels in the uterus, lungs, liver and ovaries, indicating lymphogenic metastasis. The contracted kidney was considered to be due to ureter obstruction by the enlarged uterus.

Differential diagnoses should be made for adenocarcinomas generating from the ovaries, stomach and intestines, as well as for peritoneal malignant mesotheliomas. On necropsy, neoplastic large nodules suggestive of a primary lesion were not found in the ovaries, stomach and intestines. Additionally, although peritoneal dissemination is characteristic for ovarian adenocarcinomas [4] and malignant mesotheliomas [1], metastatic lesions in the serosa of abdominal organs and the peritoneum were not found in the present sow. Histopathological characteristics of the current uterine tumor were corresponding generally to those of the uterine adenocarcinomas of cow [6], as well as a Vietnamese pot-bellied pig [5]. The present study showed that neoplastic cells reacted to three different CKs; CKs are intermediate filaments present in epithelial cells, comprising generally 20 different polypeptides [10]; AE1/AE3 antibody and pan-CK antibody stain with a broad of epithelial cells, even in undifferentiated epithelial cells, and reactivity to CK19 antibody is specific for glandular epithelial cells and basal cells of the stratified squamous epithelium [9, 10]. Vimentin antibody is well known to stain with regenerating renal tubular epithelial cells [16]. Some neoplastic cells of the present case reacted to vimentin, suggestive of immature nature of neoplastic epithelial cells. Recently, it has been reported that desmoplasia seen in scirrhous carcinomas in the salivary gland and stomach is formed by myofibroblasts; the cells may be induced by transforming growth factor- β 1 produced by neoplastic cells [11]. Desmoplasia, a characteristic of the present tumor, might be induced by the same mechanisms, because many α -SMA-positive myofibroblasts were seen in the desmoplastic areas. Such a unique finding has never been reported in uterine adenocarcinomas in domestic animals, except cattle cases in which desmoplasia is occasionally found [6, 13].

The etiology of uterine adenocarcinomas in domestic animals has not fully been investigated. Risk factors responsible for woman uterine adenocarcinomas include the obesity, anovulatory cycles, and incompatible estrogen replacement therapy, as well as the presence of estrogen secreting ovarian tumors or cysts [3]. It has been considered that endometrial hyperplasia might be closely linked to endometrial adenocarcinomas in women, because animals with hyperestrogenism undergo excessive endometrial estrogen

receptor stimulation which leads to endometrial hyperplasia [2, 3, 7]; finally, the endometrial hyperplasia progresses towards endometrial adenocarcinoma [8]. In the present sow, cystic endometrial hyperplasia was seen. The endometrial hyperplasia has been reported in dogs and cats as an aging change, but the lesion has not been described to predispose to develop endometrial adenocarcinomas [13]. The anaplastic type of woman endometrial adenocarcinomas develops without hyperestrogenism [3]. The present sow was nulliparous, but there was no information on sex-hormone milieu. The etiology of uterine adenocarcinomas in animals should be further investigated by accumulating cases of uterine adenocarcinomas. Recently, miniature pigs have been housed as a laboratory animal or pet. There are several breeds of miniature pigs such as Yucatan, Hanford and Sinclair in the United States, Gottingen in Europe, and Ohmini, CLAWN and NIBS in Japan [12]. They have characteristics of body weights ranging from 20 to 80 kg, and longevity ranging from 10 to 15 years. It is important to investigate non-neoplastic and neoplastic lesions with age, to establish the biological properties of the pig strain.

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