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SHORT PAPER

Expression of Steroid Receptors and Calponin in a Cervical Leiomyoma in a Young Pig

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Summary

Thickening of the uterine cervix and bilateral ovarian cystic change was identified in a 6-month-old pig during routine abattoir inspection. Microscopically, the cervical lesion comprised a non-encapsulated mass of densely packed, large and monomorphic spindle cells within the myometrium. Immunohistochemically, the majority of these neoplastic cells expressed the cytoplasmic terminal smooth muscle differentiation marker calponin, the nuclear oestrogen receptor α and the progesterone receptor. The ovarian cysts were classified as follicular cysts. A diagnosis of leiomyoma of the uterine cervix with bilateral ovarian follicular cysts was made. The expression of calponin as a marker of smooth muscle differentiation in tumours of the genital tract of the pig has not previously been reported. The expression of steroid hormone receptors suggests a role for steroid hormones derived from the ovarian follicular cysts in tumourigenesis.

Keywords: calponin; hormone receptors; leiomyoma; pig; uterus

Leiomyomas are the most common tumours of the genital tract of mammals. In the human female, the predilection site is the uterus and 30–50% of women develop leiomyomas during their active reproductive life (Crum *et al*., 2004). In contrast to the high prevalence of human uterine leiomyomas, few animals naturally develop these tumours. In domesticated animals, uterine leiomyomas have been reported more frequently in the dog, representing approximately 0.1% of total neoplasms and affecting middle-aged to older animals (Klein, 2001). The prevalence of uterine leiomyomas in pigs is very low and depends upon the age of the population studied (Munday and Stedman, 2002; Mozzachio *et al*., 2004).

Specific identification of smooth muscle differentiation in leiomyomas can be accomplished by immunohistochemistry using differentiation markers such as desmin, smooth muscle actin and calponin. Calponin is a highly sensitive marker of smooth muscle differentiation in human neoplasms (Zhu *et al*., 2004) and has been successfully applied to tumours from dogs and cats (Martín de las Mulas *et al*., 2002; Millán *et al*., 2007). Oestrogen receptor (ER) and progesterone receptor (PR) expression has been reported in human, feline and canine leiomyomas of the genital tract (Martín de las Mulas *et al*., 2002; Bodner *et al*., 2004; Zhu *et al*., 2004). The present study reports the expression of calponin and steroid hormone receptors in a cervical leiomyoma in a fattening pig.

A 6-month-old Landrace cross Large-White pig, one of a batch sent from a fattening unit to an abattoir, had appeared clinically normal and was of expected slaughter weight. However, during routine post-mortem inspection a thickened uterine cervix was observed. On cut section, a 4 cm rounded, white, firm mass with fibrous texture was observed within the wall of the cervix (Fig. 1). The uterine horns and vagina were unaffected. Multiple 3–4 cm cysts

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Fig. 1. White firm mass with fibrous texture in the wall of the uterine cervix of a 6-month-old female fattening pig.

with serous content were observed in both ovaries. Tissue samples were fixed in 10% neutral buffered formalin and embedded in paraffin wax. Sections (3 μm) were stained with haematoxylin and eosin (HE) and Masson's trichrome. Immunohistochemistry (IHC) was also performed on serial sections using the avidin–biotin–peroxidase complex (ABC) technique (Vectastain, ABC Kit Elite; Vector Corporation, Burlingame, CA, USA). Monoclonal mouse antibodies raised against human calponin (clone CALP; Dako S.A., Saint Just Desvern, Barcelona, Spain) diluted 1 in 400, human ERα (clone 1D5; Dako S.A.) and human PR (clone PRA109; Immunotech, Marseille, France) diluted 1 in 500 were used as previously described (Espinosa de los Monteros *et al*., 2002; Millán *et al*., 2007).

Histological examination revealed the presence of several expansive, non-encapsulated masses within the cervical myometrium. These masses consisted of densely packed cells arranged in woven fascicles and bundles (Fig. 2a). Neoplastic cells stained red by Masson's trichrome and were supported by a moderate quantity of blue fibrous stroma (Fig. 2b). The neoplastic cells were monomorphic, large, spindle-shaped and with abundant eosinophilic fibrillar cytoplasm and a large central cigar-shaped vesicular nucleus. The uterine endometrium and myometrium appeared normal. The cystic structures of the ovary had a thin wall lined by slender cells with eosinophilic cytoplasm and were classified as follicular cysts. Follicles in different stages of maturation were also present. The majority of neoplastic cells displayed cytoplasmic expression of calponin (Fig. 3a) and nuclear expression of ERα and PR (Fig. 3b, c). On the basis of both histopathological and immunohistochemical studies the definitive diagnosis was leiomyoma of the uterine cervix and ovarian follicular cysts.

In pigs, uterine leiomyomas have been observed in 5–10-year-old Vietnamese pot-bellied pigs, which are popular pets (Munday and Stedman, 2002), and pot-bellied pigs (Mozzachio *et al*., 2004). These animals have repeated oestrus cycles and therefore, chronic oestrogen and progesterone stimulation, a situation that has been associated with both uterine leiomyomas and endometrial hyperplasia in women (Flake *et al*., 2003; Crum *et al*., 2004). In the present case the endometrium appeared normal and without alterations in endometrial glands. The prevalence of uterine leiomyoma in a population of slaughter pigs with a mean age of four years was 0.4% (Akkermans and Van Beusekow, 1984) and, to the authors'

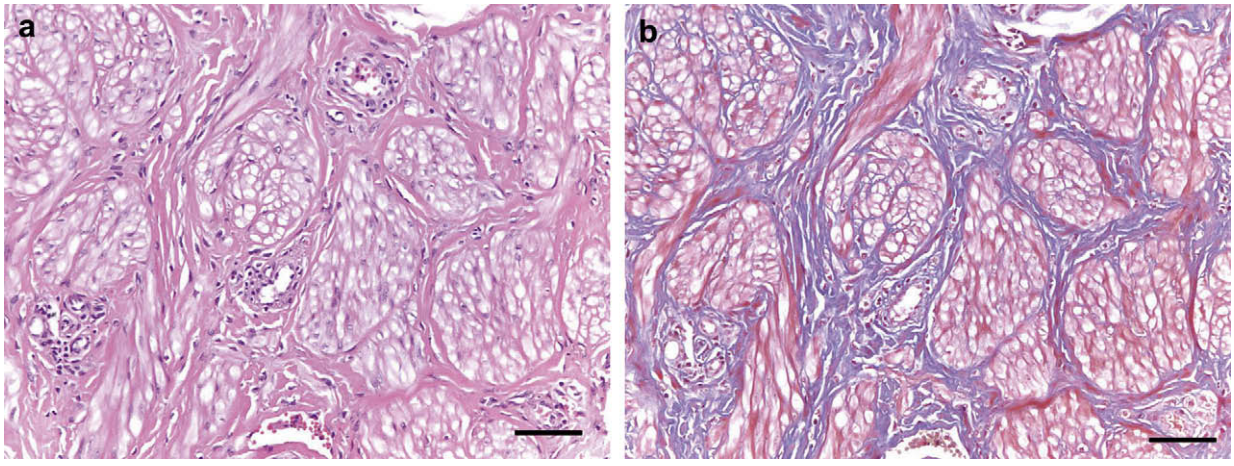

Fig. 2. (a) Microscopical appearance of the uterine cervical leiomyoma. The neoplasm is composed of densely packed cells arranged in woven fascicles and bundles. The neoplastic cells appear monomorphic and spindle-shaped, with abundant eosinophilic cytoplasm and a large central cigar-shaped vesicular nucleus. The cytoplasm contains numerous clear vacuoles. HE. Bar, 30 μm. (b) Abundant red-staining neoplastic cells and moderate blue-staining fibrous connective tissue stroma. Masson's trichrome. Bar, 30 μm.