

Mature teratoma arising from an undescended testis in a horse: comparison between ultrasonographic and morphological features

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Scant information is available on the ultrasonographic appearance of different testicular tumours in the stallion. Preoperative ultrasound imaging and gross and microscopic features of a testicular teratoma in a horse is described. An asymptomatic 4 years old cryptorchid Arabian horse was admitted for orchiectomy. Combined transabdominal and inguinal ultrasound examination revealed a large complex ovoid mass, containing both solid and cystic elements with internal echoes, located dorsally to the superficial inguinal ring. Two main hypoechoic cavities divided by a linear hyperechoic septum were evident, with a hyperechoic circular structure inside the lumen of the largest one. A tumour of the undescended testis was suspected. Ultrasound findings guided a provisional diagnosis of teratoma that was confirmed by histology following surgical removal of the tumour. Due to the correspondence between ultrasonographic and morphological features, trans-abdominal ultrasonography was conclusive in the preoperative planning. (Folia Morphol 2016; 75, 2: 211–215)

Key words: equine testicular tumour, ultrasound imaging, morphology, cryptorchidism

INTRODUCTION

Teratomas are the most common neoplasm of the colt and young stallion and occur frequently in cryptorchid testes [4, 20, 21]. They are benign and slow growing tumours composed of multiple tissues, derived from different germ layers, not native to the area in which they occur [12]. Testicular teratomas in the horses are asymptomatic and frequently cause complications when a cryptorchid horse is submitted to orchiectomy [8].

In cryptorchid horses, combined transabdominal and inguinal ultrasonography provides an easy diagnosis of abdominal cryptorchidism, allows the evaluation of abnormalities, such as tumours or cysts,

and could enhance the choice of an appropriate surgical approach for their removal [17]. However, to our knowledge, scant information is available on the ultrasonographic appearance of different testicular tumours in the stallion [3, 5, 22] and imaging findings of intra-abdominal mature teratomas in undescended testis have been very rarely reported in the horse [8].

Ultrasound imaging of a testicular teratoma in a 4 years old Arabian horse is described. Ultrasound findings were peculiar and similar to that reported in humans. In horses, imaging findings reflect morphology and physical properties of the multiple tissue of the testicular teratoma and can allow a provisional diagnosis that must be confirmed by histopathology.

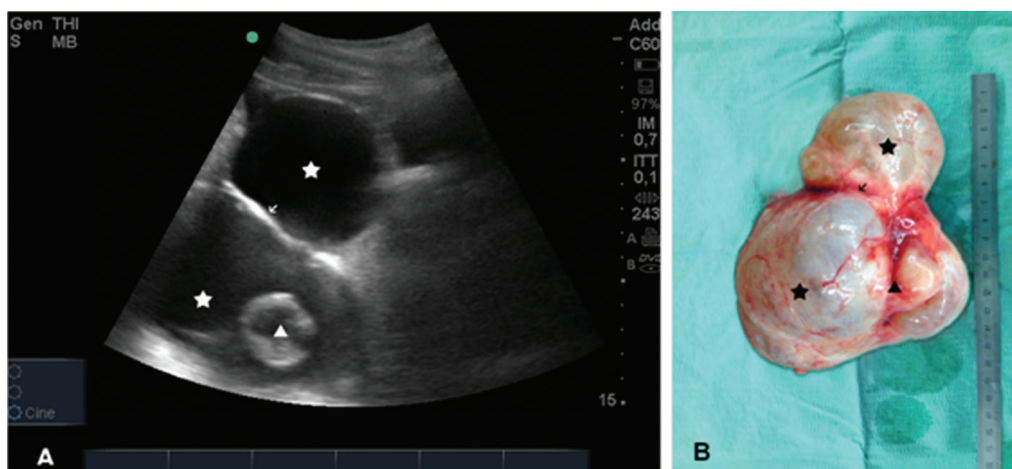


Figure 1. **A.** Ultrasonography showing two main hypoechoic cavities (white star) divided by a linear hyperechoic septum (white arrow), with a hyperechoic circular structure inside the lumen of the largest one (white triangle). **B.** Two large cystic structures (black star) divided by a septum (black arrow) and a more compact area corresponding to a solid mass of greyish-white cartilaginous and bony tissue (black triangle) (height: 17.7 cm; width: 12.5 cm).

MATERIALS AND METHODS

History and clinical finding

A 4-year-old cryptorchid Arabian horse was admitted for orchiectomy. An empty right scrotum and a left scrotum containing a palpable testis were observed at the physical examination. Combined transabdominal and inguinal ultrasound examination, performed with portable ultrasound (Sonosite® M-turbo®, convex probe 3–5 MHz, © 2013 FUJIFILM SonoSite, Inc.), revealed the presence of a large complex ovoid mass, containing both solid and cystic elements with internal echoes, located dorsally to the superficial inguinal ring. Two main hypoechoic cavities divided by a linear hyperechoic septum were evident, with a hyperechoic circular structure inside the lumen of the largest one (Fig. 1A). Superficial inguinal lymph nodes were normal and transabdominal ultrasonography of abdominal viscera did not reveal any other significant abnormality.

A tumour of the undescended testis was suspected and a laparoscopy was advised, but the owner refused to carry the horse to an equipped facility.

Treatment

Feed was withheld 12 h before general anaesthesia. Before the induction of anaesthesia, benzyl procaine penicillin/dihydrostreptomycin (Gellipen®, Intervet Productions S.r.l., Aprilia, Latina, Italy; 22,000 U/kg intravenously [IV]) and phenylbutazone (Fenilbutazone®, ATI s.r.l., Ozzano Emilia, Bologna, Italy; 4.4 mg/kg IV) were administered. After acepromazine (Prequillan®, FATRO SpA, Ozzano Emilia, Bologna, Italy; 0.025 mg/kg) and

romifidine (Sedivet®, Boehringer Ingelheim Div.Veter Milano, Italy; 100 µg/kg) premedication, anaesthesia was induced by a combination of diazepam (Valium®, Roche Spa, Milano, Italy; 0.05 mg/kg) and ketamine (Ketavet100®, Intervet Productions S.r.l., Aprilia, Latina, Italy; 2.2 mg/kg IV) and maintained on isoflurane in 100% oxygen. The horse was positioned in dorsal recumbency. After aseptic preparation, a skin incision was made directly over the right superficial inguinal ring and the inguinal fascia was digitally separated. The inguinal extension of *gubernaculum testis* was easily identified and traction on this ligament allowed the vaginal process to be everted into the vaginal canal. The vaginal process was longitudinally incised by Metzenbaum scissors. The spermatic cord was completely identified and exteriorised, but to pull out the testis was impossible. Thus, a celiotomic approach was performed. The spermatic cord was emasculated by the inguinal approach and the testis was pulled out by the abdominal incision.

The left testis was removed by a classical scrotal approach.

The horse recovered uneventfully after surgery and was discharged from the hospital 2 weeks later.

Diagnosis

The left testis appeared normal at the gross examination. The right testis showed an epididymis-like structure and two large cystic structure divided by a septum (height: 17.7 cm; width: 12.5 cm) (Fig. 1B). A volume of 400 mL of fluid, straw yellow in colour was aspirated from the lumen of the cyst. Within

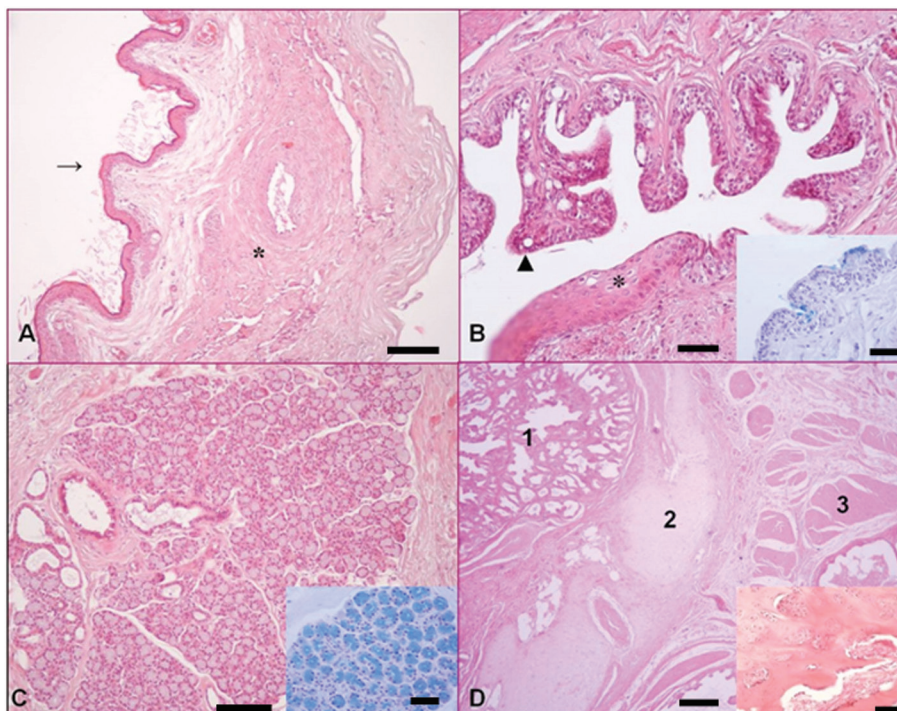


Figure 2. **A.** Haematoxylin and eosin (H&E), original magnification 10×: teratoma, cyst wall. Squamous epithelium (arrow) and vascularised connective stroma (asterisk); **B.** H&E, original magnification 20×: teratoma. Transition between a pseudostratified ciliated epithelium (arrow head) and pluristratified epithelium (asterisk). Particular: alcian blue periodic acid-Schiff (PAS) stain, original magnification 40×: in blue are stained the acidic mucopolysaccharides secreted by goblet cells of the pseudostratified epithelium; **C.** H&E, original magnification 10×: teratoma, mucin producing glands. Particular: Alcian blue PAS stain, original magnification 40×: in blue, acidic mucopolysaccharides secreted by mucin producing glands; **D.** H&E, original magnification 10×: trabecular glandular structure (1), cartilaginous tissue (2) smooth muscle cells (3). Particular: H&E, original magnification 20×: teratoma, trabecular bone and cartilage.

the wall of the cyst, a more compact area was seen, showing solid masses of greyish-white cartilaginous and bony tissue on the cut section.

Sections of the tumour and of the left testis were fixed in 10% formalin, dehydrated, in a graded series of ethanol and embedded in paraffin. After fixation, samples containing bony tissue were decalcified for 3 days with a 4% trichloroacetic acid solution and then embedded in paraffin. The paraffin blocks were cut into 6 µm thick sections and stained with haematoxylin and eosin (H&E) for a general morphological examination and with the histochemical stain alcian blue periodic acid-Schiff (PAS) to differentiate between neutral and acidic mucin producing glands.

Microscopic examination of the cystic walls of the right testis revealed the presence of a squamous pluristratified epithelium. The more compact area was characterised by various tissues derived from all germ layers including: squamous pluristratified epithelium (ectodermal), myelinated and non-myelinated nervous bundles (neuroectodermal), trabecular bone

tissue, cartilage, smooth muscle fibres (mesodermal), ciliated pseudostratified epithelium and acidic mucin producing glands (endodermal) (Fig. 2). Therefore, a final diagnosis of testicular teratoma was made.

Seminiferous tubules in the left testis had diffusely distended lumen with severely reduced spermatogenesis. A relative prominence of interstitial Leydig cells, interstitial fibrosis and presence of intraluminal spermatidic multinuclear giant cells were observed as well. Overall, these findings were consistent with a diagnosis of testicular degeneration.

DISCUSSION

Different techniques have been proposed for the diagnosis of cryptorchidism and location of the testis. Hormonal assays could help to identify cryptorchidism, but they do not locate the errant testis. Furthermore human chorionic gonadotropin (hCG) stimulation test cannot be used in horses of less than 18 months of age and during winter, while plasma oestrogen levels are not performed in horses

younger than 3 years and in donkeys [7]. Palpation *per rectum* of the abdominal testis and transrectal ultrasonography are considered sensitive and reliable techniques when performed by experienced clinicians, but they are potentially dangerous and inappropriate in nervous, young horses and in ponies [11].

It was demonstrated that transabdominal ultrasound examination is helpful part of diagnostic investigation of equine abdomen [2]. Furthermore, combined transabdominal and inguinal ultrasonography is reported as a highly sensitive technique for evaluation of cryptorchid testis. It is safe and allows to identify the site of the testis and to imagine the cryptorchid testis, showing enlargement and morphological alterations, potentially associated to testis neoplasia, so that the surgeon can program the most suitable technique [6]. Abdominal undescended testes have a characteristic appearance with uniform grey stroma and bright acoustic signal of the tunica albuginea. The presence of hyperechoic albuginea and either a central vein or the epididymis of abdominal testes help to discriminate from other abdominal structures such as empty small intestinal loops, the density of which was sometimes identical to that of a cryptorchid testes [18].

Surgical removal of cryptorchid testicular teratoma has been described in horse using laparoscopy, celiotomy, inguinal and flank approach and equine surgeons need to examine cryptorchid horses accurately to choose the best and safest technique [8, 13, 14]. In this case, on the basis of the sonographic findings, laparoscopy was proposed but the owner refused to transfer the horse to a facility equipped for laparoscopy, due to logistic and economic reasons. The removal of testicular teratomas by an inguinal approach, without a pre-surgical diagnosis, implies a considerable elongation of surgery and anaesthesia, but in this case it took a short time, thanks to a correct planning of the surgery.

Cribb and Bourè [8] describe removal of testicular teratoma by laparoscopy in a standing horse, after an unsuccessful inguinal approach. An accurate transabdominal ultrasound examination could have avoided the first anaesthesia and surgery.

In this case, the primary differential diagnosis for the ultrasonographic findings was a teratoma. Differential diagnoses were seminoma, Sertoli cell tumour, Leydig cell tumour, teratocarcinoma, and embryonal carcinoma [1, 4, 10, 15, 19].

In humans, ultrasound is used as a screening modality for testicular tumours due to its high sensitivity for lesion detection. Ultrasound alone is not capable of differentiating specific cell types within a testicular mass but may be able to narrow the differential diagnosis significantly, because it reflects the morphology of the examined tissues [9]. In the horse, seminoma are characterised by a diffusely heterogeneous appearance. The testicle appears diffusely hypoechoic with ill-defined regions of hyperechogenicity giving the appearance of hypoechoic nodules throughout the parenchyma. Anechogenic band, representing a pseudocapsule, could be seen surrounding the testicle [3]. In cases of malignant mixed sex cord-stromal tumour, ultrasonographic evaluation of the testes revealed diffuse heterogeneous parenchyma with multiple hypoechoic nodular areas [5, 22].

Teratomas are composed of mature or immature components derived from pluripotent primordial cells of all 3 germ layers. Thus ectodermal (skin derivatives and neural tissue), mesodermal (bone, fat, cartilage, and muscle), and endodermal (gastrointestinal and bronchial epithelium, thyroid) tissues in most cases coexist. Furthermore, cystic components, commonly represented by greasy fluid made of sebum, keratin, and hair surrounded by a capsule of varying thickness, may be found. Specific imaging findings, indicative of a teratoma, reflect the physical properties of these tissues. In humans, the ultrasound features of testicular teratomas are typical, as well. The classic sonographic appearance of a human mature teratoma is a simple or complex cystic mass with a densely echogenic tubercle called *Rokitansky nodule*, projecting into the cyst lumen, along with concomitant areas of diffuse or localised echogenicity, representing fat or sebaceous material that produces sound attenuation [16]. Similar findings were seen in this horse. Two hypoechoic structures divided by a linear hyperechoic septum dorsally to the superficial inguinal ring, and a hyperechoic circular structure, analogue to a *Rokitansky nodule* inside the lumen of the largest cavity were observed. These findings were suggestive of teratoma.

Microscopic alterations of the normotopic testis, similar to those observed in the left testis of this case, can be present in horses affected by testicular teratoma. The observed lesions might be related to unknown aetiological factors that would have influenced the intrauterine growth of both gonads. Thus, bilateral orchiectomy is the advisable treatment.

CONCLUSIONS

In conclusion, the ultrasonography proved to be the most important step in presurgical evaluation of cryptorchid horses and sonographic findings of intra-abdominal testicular teratomas were specific and overlapping the images observed in human medicine.

Even if only the histological examination makes possible to put the final diagnosis, an undescended testis, coupled with an abdominal mass, should very strongly suggest the diagnosis of an intra-abdominal testicular tumour. As teratomas include tissues derived from all embryologic germ layers, ultrasound imaging results in specific sonographic findings. These imaging modalities can aid in determining the extent of disease, as well as provide information for alternative diagnoses.

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