



# Theriogenology Question of the Month

## History

A 5-year-old Tennessee Walker mare that had never foaled was referred to our veterinary medical teaching hospital for evaluation of the reproductive tract. Although the mare had never been tested for receptivity to a stallion, the owner believed that the mare had normal estrous cycles on the basis of cyclic change in the mare's behavior and urination pattern. As part of the routine prebreeding preparation for a maiden mare, the referring veterinarian had performed a manual vaginal examination and attempted to break down an intact vaginal vestibular membrane (ie, persistent hymenal remnant). The referring veterinarian had been able to digitally excavate several centimeters past the cranial border of the vestibule but was not able to locate the vaginal lumen.

Initial examination at our facility revealed that the perineum had a normal appearance and conformation. Transrectal palpation of the reproductive tract revealed that the left and right ovaries were of normal size and shape with multiple follicles, which was confirmed by transrectal ultrasonography. However, both ovaries were located slightly more cranial and dorsal than anticipated. In addition, the cervix, uterine body, and uterine horns were not palpable transrectally. A vaginal speculum could be inserted approximately 6 cm (2.4 inches) into the vestibule. This comprised the length of the entire vestibule, which was confirmed by simultaneous transrectal ultrasonography. The vestibule was insufflated with air and examined by use of a video

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## ABBREVIATIONS

AMH	Antimüllerian hormone
MIF	Müllerian inhibiting factor



Figure 1—Endoscopic view of the cranial wall of the vestibule of a mare with no transrectally palpable tubular reproductive tract. The vestibule was insufflated with air to aid in the endoscopic examination.

endoscope. A soft tissue septum with 2 lateral blind pouches was visible on the cranial wall of the vestibule, immediately dorsal to the urethral opening (Figure 1). A diagnosis of aplasia of the tubular reproductive tract was made.

## Question

What is the developmental origin of the 2 lateral blind pouches and soft tissue septum? *Please turn the page.*

## Answer

The structures are most likely remnants of the paramesonephric ducts at the point where they fuse with the urogenital sinus.

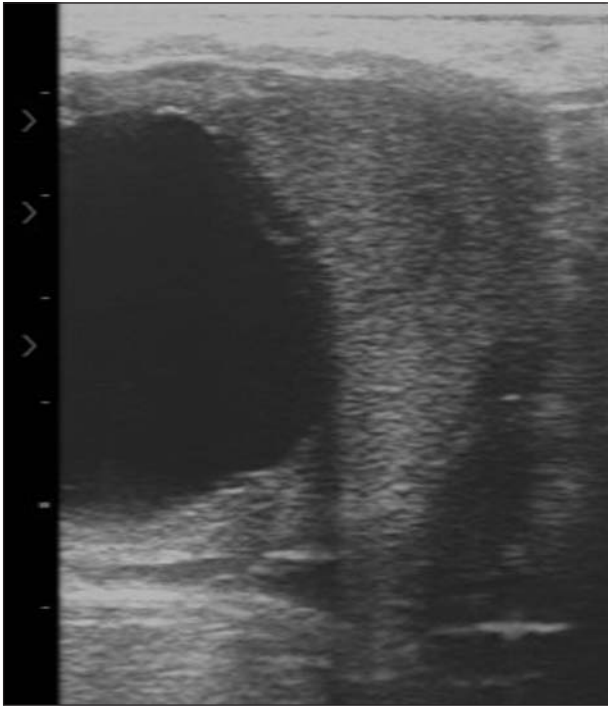


Figure 2—Ultrasonogram of the left ovary of a mare with no evidence of a tubular reproductive tract. Notice that the appearance of the structure is consistent with a corpus luteum. Marks on the left side are at intervals of 1 cm.

## Result

The mare was reexamined at our facility 3 weeks later. Transrectal ultrasonography revealed 3 follicles on the left ovary, each of which was > 3 cm (1.2 inches) in diameter, and several smaller follicles on the right ovary. In addition, a structure consistent in appearance with a corpus luteum was evident on the left ovary (Figure 2). We interpreted this to indicate that the mare could ovulate.

During this examination, the mare was exposed to a teaser stallion. She responded negatively (ears back, tail clamped, elusive movements, and kicking at the stallion). However, serum progesterone concentration was not measured to confirm that the ovarian structure was luteal tissue.

A jugular venous blood sample was collected into sodium heparin and submitted to a laboratory<sup>a</sup> for chromosomal analysis. Results of that analysis revealed a normal karyotype for a mare (64,XX and Sry negative).

## Discussion

In mammals, the phenotypic sex is determined essentially by the testes or the lack of testes in a developing embryo (Figure 3). Masculinization is the result of hormones secreted by the testes (ie, testosterone and AMH). Lack of these hormones results in formation of a female phenotype. A sexually undifferentiated embryo possesses Wolffian (mesonephric) and Müllerian (paramesonephric) ducts, which become the tubular reproductive tract. The genital tubercle, urogenital sinus, and genital swellings become the external genitalia.<sup>1</sup> Anti-Müllerian hormone (previously known as MIF, a member of the activin-transforming growth factor- $\beta$  superfamily) is the earliest substance

secreted by the fetal testes. It has been reported<sup>2</sup> in rats that although AMH is not expressed by females during fetal development, granulosa cells of preantral and small antral ovarian follicles begin expressing AMH shortly after birth and continue to express AMH throughout adulthood. As its name implies, AMH causes regression of the Müllerian ducts, which leaves the Wolffian ducts to develop into the male reproductive tract. Fetal interstitial (Leydig) cells secrete testosterone, which stimulates differentiation of the Wolffian ducts into the epididymides, vas deferens, and accessory sex glands. In females, lack of AMH allows persistence of the Müllerian ducts, which develop into the uterine tubes, uterus, cervix, and vagina. In mares, the medial walls of the 2 ducts fuse to form the uterine body, cervix, and vagina, and the septum between the ducts undergoes atresia. Because there is no testosterone in a female fetus, the Wolffian ducts regress and the vestibule, clitoris, and vulva develop.<sup>1</sup>

Mares with developmental abnormalities of the reproductive tract may have behavioral abnormalities, including stal-

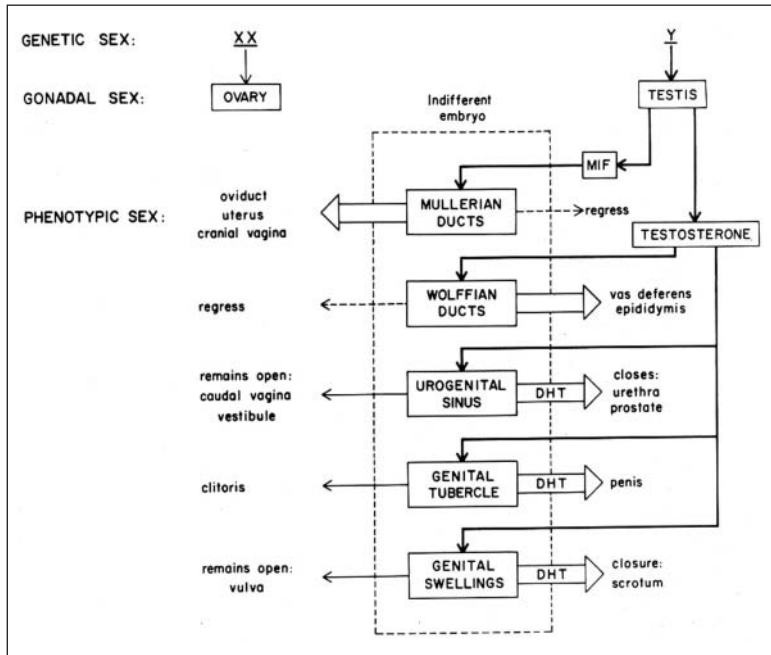


Figure 3—Illustration depicting the roles of MIF (currently known as AMH) and dihydrotestosterone (DHT) on sexual differentiation of the reproductive tract. (Adapted from Meyers-Wallen VN, Patterson DF. Disorders of sexual development in the dog. In: Morrow DA, ed. *Current therapy in theriogenology*. 2nd ed. Philadelphia: WB Saunders Co, 1986;567–574. Reprinted with permission.)

lion-like behavior, or be infertile. The most common congenital abnormality detected is an intact vaginal vestibular membrane (persistent hymenal remnant).<sup>3</sup> A persistent hymen results from failure of the caudal sections of the Müllerian ducts to fuse with the urogenital sinus. Hymenal remnants vary in appearance, depending on whether the hymen is complete or incomplete.<sup>4</sup> A complete persistent hymen can result in accumulation of fluid within the vagina because of impaired drainage. The clinical appearance depends on the amount of fluid that accumulates and may be seen clinically as a cystic structure protruding through the vulvar lips. Other developmental abnormalities reported in mares include hypoplasia of the vagina and vestibule,<sup>3</sup> uterus unicornis,<sup>5</sup> segmental uterine aplasia,<sup>6</sup> and uterus bicolis (double cervix).<sup>7</sup>

It is unclear whether the tubular reproductive tract of the mare reported here was aplastic or atretic. It is possible that the Müllerian ducts failed to develop in this mare. Aplasia of the uterus is extremely rare, except in freemartins and hermaphrodites.<sup>8</sup> Uterine aplasia may involve segments of 1 or both uterine horns, or the uterine horns may not be evident or may develop singly or in association with varying degrees of aplasia of the cervix and vagina.<sup>8</sup> In mice, XX, Sry-negative females that lack the *Lim1* gene have normal ovaries but lack uterine tubes, a uterus, and a cervix.<sup>1</sup> Multiple mutations in retinoic acid receptor genes also result in female mice that are missing all or the caudal portions of the reproductive tract.<sup>1</sup> In the mare described here, it is possible that vestigial remnants of the Müllerian ducts existed that were not found during transrectal palpation or ultrasonography but that were sufficient to supply luteolytic concentrations of prostaglandin F<sub>2α</sub>.

It is also possible that the mare was exposed to AMH during fetal development. The mare could have had an ovotestis despite a normal ultrasonographic appearance of ovaries and the fact she was Sry-negative, as has been reported<sup>9</sup> in a few animals with 64,XX, Sry-negative sex reversal. The lack of Müllerian ducts in this mare could have resulted from testicular tissue that secreted AMH during embryogenesis, which resulted in complete regression (atresia) of the Müllerian ducts. However, an ovotestis almost always results in abnormalities of the external genitalia or behavioral aberrations, which were not evident in the mare.

A male cotwin early during gestation may have led to AMH exposure in this mare. A male cotwin could have secreted AMH and then died and been resorbed before prenatal secretion of testosterone. The mare's dam had a history of twin pregnancies, although a twin fetus was not reported for the pregnancy that resulted in the birth of the mare reported here. However, the mare's dam was not examined to detect pregnancy until 5 months of gestation, at which point there would have been no evidence of a male twin that died before 60 days of gestation. It is interesting that most mares born cotwin to a male are able to reproduce normally.<sup>10</sup>

In a retrospective study<sup>10</sup> of male-female twins recorded by the Arabian Horse Registry of America, 24 of 35 mares born with a male cotwin subsequently gave birth to a foal. Thus, it is generally suggested that in most cases of male-female twins in horses, placental vascular anastomoses develop at a later point in gestation and do not affect the development of the female twin's reproductive tract.

It is possible that vestigial remnants of the Müllerian ducts existed in the mare that were not found during transrectal palpation or ultrasonography but which may have been found during postmortem examination. To our knowledge, this is the first report of an apparent complete aplasia of the tubular reproductive tract in a mare. The actual incidence of this condition in the equine population is unknown because the condition would only be detected in mares examined for breeding management or to determine the cause of infertility.

## Outcome

The mare was considered unsuitable for use in breeding because the tubular reproductive tract had not developed or was too hypoplastic to be detected during transrectal examinations. The mare was retained as a companion animal and used for competitive and pleasure riding.

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