Review of Ovarian Abnormalities in the Mare

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The clinical signs and treatments of ovarian abnormalities in the mare are reviewed. Clinical techniques that may be used in the diagnosis and differentiation of ovarian abnormalities are also presented. Author's address: Dept. of Clinical Sciences, Colorado State University, Ft. Collins, CO 80523. © 1998 AAEP.

1. Introduction
Pathologic conditions of the equine ovary can be divided into abnormalities that cause the ovaries to become larger than normal, that cause them to become smaller than normal, or that cause an alteration in normal ovarian physiology or function. The purpose of this paper is to review the clinical signs, diagnostic procedures, and treatments of ovarian abnormalities in the mare.

2. Materials and Methods
This paper reviews scientific and clinical literature concerning ovarian abnormalities in the mare. Clinical techniques that may be used in the diagnosis and differentiation of ovarian abnormalities are listed in Table 1.

3. Results
A. Normal Ovaries
Equine ovaries are kidney shaped and have a prominent depression on their ventral border, the ovulation fossa, through which all ovulations take place. The size of the ovaries varies with season, age, and stage of the estrous cycle.1,2 A majority of the ovary is covered with visceral peritoneum or serosa. The ovulation fossa is lined by germinal epithelium. Adipose tissue and adrenocortical nodules are commonly found under the serosal surface of the equine ovary.2 Tunica albuginea, a thick connective tissue layer, is located beneath the serosa. Ovarian architecture of the horse is unique in that the cortical zone, which contains the follicles and corpora lutea, is located in the interior of the ovary, while the medullary or vascular zone is located in the superficial region.1

Follicles are composed of two main cell types, i.e., granulosa and theca cells. Granulosa cells line the inside of the follicle. The oocyte is surrounded by a layer of granulosa cells called the cumulus oophorus and is attached to the internal follicular wall at the hillock. Theca cells surround the outside of the follicle and are separated from granulosa cells by a basement membrane. Granulosa cells produce the protein hormone inhibin and, in conjunction with theca cells, produce estradiol. Theca cells are responsible for the production of androgens.

Ovulation occurs in response to a surge of luteinizing hormone secreted by the anterior pituitary. The rupture of the follicle results in hemorrhage from blood vessels in the theca and formation of an intermediate structure, the corpus hemorrhagicum. A corpus luteum subsequently forms from the corpus hemorrhagicum following invasion of granulosa and
luteal cells. Luteinization of the granulosa and theca cells results in the development of large and small luteal cells, respectively. The corpus luteum of the nonpregnant mare secretes progesterone until prostaglandins released by the endometrium cause destruction of the luteal cells, or luteolysis.

B. Enlarged Ovaries

The differential diagnosis for unilaterally or bilaterally enlarged ovaries in the mare includes pathologic conditions such as ovarian tumors and ovarian hematomas and physiologic conditions such as the ovarian enlargement that normally occurs during pregnancy.

1. Granulosa Cell Tumor

The most common ovarian tumor in the mare is the granulosa cell tumor (GCT). Granulosa cell tumors are almost always unilateral, slow growing, and benign. An examination of the affected ovary by using transrectal ultrasonography often reveals a multicystic or honeycombed structure (Fig. 1), but the tumor may also present as a solid mass or as a single large cyst (Figs. 2 and 3). The contralateral ovary is usually small and inactive, although mares with a GCT on one ovary and a functional contralateral ovary have been reported. Behavioral abnormalities such as prolonged anestrus, aggressive or stallionlike behavior (Fig. 4), and persistent estrus or nymphomania may be expressed in affected mares.

Granulosa cell tumors are hormonally active, and clinical diagnostic assays for the detection of a GCT include the measurement of inhibin, testosterone, and progesterone (see Appendix A for endocrine laboratories). Inhibin is elevated in approximately 90% of the mares with a GCT. It has been hypothesized that inhibin produced by the GCT is responsible for the inactivity of the contralateral ovary through the suppression of pituitary follicle-stimulating hormone release. Serum testosterone levels may be elevated if a significant theca cell component is present in the tumor (i.e., a granulosa-theca cell tumor, or GTCT). Testosterone is elevated in approximately 50-60% of affected mares and is usually associated with stallionlike behavior. Progesterone concentrations in mares with a GCT are almost always below 1 ng/ml, since normal follicular development, ovulation, and corpus luteum formation do not occur.

Therefore, measurements of inhibin levels >0.7 ng/ml, testosterone levels >50-100 pg/ml, and progesterone levels of <1 ng/ml are suggestive of a granulosa cell tumor in a nonpregnant mare (Table 2).

Granulosa cell tumors are usually surgically removed if the tumor affects follicular development on the contralateral ovary, causes behavioral abnormalities, or is a source of colic. Surgical approaches for tumor removal include colpotomy, flank and ventral midline laparotomy, and laparoscopy.

Ovulation from the remaining ovary will occur approximately 6-8 months after tumor removal. Attempts at inducing follicular development and

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Table 1. Procedures Used to Evaluate Ovarian Abnormalities

Fig. 1. Sonograph of a GCT in a mare.

Fig. 2. GCT composed of multiple small cysts.
ovulation in the remaining ovary within 1 month after tumor removal by the administration of gonadotropin-releasing hormone (GnRH) has not been successful. However, the administration of equine follicle-stimulating hormone, in the form of equine pituitary extract, has been successful in inducing an ovulation from the contralateral ovary after the surgical removal of a GCT that resulted in a pregnancy.\textsuperscript{a}

2. Cystadenoma

The most common tumor of the surface epithelium of the equine ovary is the cystadenoma. Cystadenomas occur unilaterally, and the contralateral ovary is normal. The ultrasonographic appearance of the affected ovary may include one to many cystlike structures (Fig. 5). In general, these tumors are rare and benign and are not considered to be hormonally active, although reports of mares with cystadenomas with elevated plasma testosterone concentrations have been made.\textsuperscript{3,7}

The treatment of choice for an ovarian cystadenoma is surgical removal. The decision to remove the affected ovary does not have to be made immediately, as the tumor is slow growing and has not been reported to metastasize. However, if the tumor does continue to enlarge, the mare may exhibit episodes of abdominal pain.

3. Teratoma and Dysgerminoma

Teratomas and dysgerminomas are rare ovarian tumors of germ cell origin.\textsuperscript{8,9} Teratomas are considered to be benign, while dysgerminomas are potentially malignant. Both are unilateral, hormonally inactive, and associated with normal contralateral ovaries. Germ cell tumors may contain hair, bone, muscle, and other tissues (Fig. 6). They do not alter the behavior of the mare and do not interrupt the estrous cycle.

The treatment of choice for germ cell tumors is surgical removal. This is especially true for the dysgerminoma, because of its potential for metastasis.

4. Ovarian Hematoma

Hematomas are one of the most common causes of unilateral ovarian enlargement.\textsuperscript{10,11} Hematomas result from excessive hemorrhage into the follicular lumen following ovulation, and they are essentially greatly enlarged corpora hemorrhagica (Fig. 7). The contralateral ovary is normal in size and function, and the mare continues to cycle normally. No behavioral abnormalities are noted and endocrine patterns of the mare are normal.

Hematomas of the ovary usually do not require treatment. The hematoma will gradually reduce in size over a period of several weeks, and in most instances the ovary returns to normal function. However, occasionally a hematoma may destroy the ovarian germinal tissue, rendering the affected ovary nonfunctional.

5. Cystic Ovaries

The presence (or absence) of cystic ovaries in the mare, as described in dairy cattle, has been a subject of debate for years. Persistent anovulatory follicles...
do occur in the mare and are discussed in a later section.

A case of bilateral polycystic ovaries was recently diagnosed in a 6-year-old Andalusian mare presented to Colorado State University. The ovaries were each approximately 15 cm in diameter and had remained enlarged for almost 2 years. Ultrasonographic evaluations and measurements of serum hormone concentrations were performed every 1–3 months. A laparoscopic ovarian biopsy was eventually performed on each ovary, and a histologic diagnosis of polycystic ovaries was made independently by pathologists at two institutions. The ovaries were subsequently removed, and a further histologic evaluation confirmed the previous diagnosis of polycystic ovaries.

Cysts within the region of the ovulation fossa (Fig. 8) and cysts adjacent to and within the oviductal tissue are common in the mare and generally arise from structural remnants of the müllerian or wolfian ducts. Fossa cysts and parovarian cysts may be found in a high percentage of mares as incidental findings. These cysts are generally not associated with reduced fertility unless they obstruct the process of ovulation or oocyte transport into and through the oviduct. No treatment is necessary for fossa cysts or parovarian (fimbrial) cysts that are not interfering with ovulation or oocyte transport.

6. Ovarian Enlargement During Pregnancy

Pregnant mares have bilateral ovarian enlargement after secondary corpora lutea begin to form at approximately day 40.1 Pregnant mares may show estrus, aggressive behavior, or stallionlike behavior. Testosterone concentrations increase to greater than 100 pg/ml by days 60–90 of pregnancy and reach peak concentrations at approximately day 200 of pregnancy (Fig. 9).12 Testosterone concentrations subsequently decline to basal levels by the time of foaling. The major source of testosterone is the fetal gonads, which attain a size that is larger than the ovary of the mare at 7–8 months of gestation. The decline in size of the fetal gonads is associated with a decrease in testosterone concentrations in the pregnant mare. Pregnancy must be ruled out be-
for considering the possibility of a granulosa cell tumor in a mare with bilaterally enlarged ovaries, elevated testosterone, and stallionlike behavior.

No treatment is warranted for the behavioral changes that may occur secondary to the normal physiologic increase in testosterone during pregnancy. Management practices should be implemented to ensure that a pregnant mare with aggressive or stallionlike behavior does not injure a person or another horse. The ovaries of the pregnant mare should obviously not be removed.

C. Small Ovaries
The differential diagnosis for bilaterally small or inactive ovaries in the mare include pathologic conditions such as chromosomal abnormalities, Equine Cushing’s Disease, exogenous hormone treatment, and malnutrition and physiologic conditions such as prepuberty, advanced age, seasonal anestrus, and postpartum anestrus.

1. Chromosomal Abnormalities
The normal chromosome number of the domestic horse (Equus caballus) is 64, and it consists of 62 autosomes and two sex chromosomes. The karyotype of the normal mare and stallion are 64,XX and 64,XY, respectively. Horses of all domestic breeds have the same number, size, and shape of chromosomes.

Chromosomal abnormalities, especially of the sex chromosomes, have been associated with infertility in the horse. The prevalence of sex chromosome abnormalities in the mare has been reported to be <3%. A chromosomal abnormality may be suspected in a mare of breeding age with primary infertility and gonadal hypoplasia.

The most commonly reported chromosomal abnormality of the horse is 63,X gonadal dysgenesis, in which only a single sex chromosome is present. The condition may occur when the sex chromosome pair fails to separate during meiosis, producing one gamete without a sex chromosome and another with two sex chromosomes. The equine condition is analogous to Turner's syndrome in humans. The 63,X (or XO) condition has been detected in most domestic horse breeds, including draft and miniature breeds.

Horses with gonadal dysgenesis develop as phenotypic females because of the absence of a Y sex chromosome. Affected horses are often small in size for their age and breed, have small ovaries lacking follicular development, and have endometrial gland hypoplasia (Fig. 10). The uterus and cervix are generally small and flaccid. The external genitalia is female, but the vulva may be smaller than normal and there is no ditoral hypertrophy. XO mares may exhibit anestrous or irregular estrous behavior and occasionally stand to be mated. True XO mares are considered to be sterile. However, mares with a mosaic or chimeric karyotype (63,XO/64,XX) are not always small in stature and some have been reported to produce a foal. Mosaic mares account for approximately 15–30% of all cases of gonadal dysgenesis. Numerous other chromosomal abnormalities have also been reported in the mare.

Chromosome analysis (karyotyping) can be performed on any tissue with actively dividing cells. A fresh blood sample collected into acid citrate dextrose or heparin may be sent by overnight courier to a laboratory specializing in animal karyotyping (see Appendix A).

An examination of peripheral blood smears for sex chromosome appendages, or drumsticks, on polymorphonuclear neutrophils (PMN’s) can be used as a screening procedure to detect a reduced number of chromosomes. Drumsticks appear as a lobe on the nucleus of PMN’s and are present in approximately 10% of PMN’s from normal mares and are absent in stallions and geldings. An examination of peripheral blood smears will reveal an absence of drumsticks in XO mares.

No treatments are possible to correct a chromosomal abnormality.

2. Age-Related Ovarian Dysfunction
Ovulatory dysfunction has been identified as a cause of subfertility in mares approximately 20 years of age or older. Older mares may have a longer inter-ovulatory interval than younger mares because of a longer follicular phase. A lengthening of the follicular phase in association with elevated gonadotropin concentrations may indicate impending reproductive senescence in older mares. Complete ovulation failure or ovarian senescence has been observed in aged mares and may be due to an insufficient number of primordial follicles. In addition, older mares may experience a delay in their initial ovulation of the year by an average of 2 weeks.

No effective treatments are currently available in the mare for promoting follicular growth in senescent ovaries. Predisposing factors for subfertility in older mares such as poor perineal conformation and ineffective uterine clearance should be addressed.

3. Exogenous Hormone Treatment
Anabolic steroid administration may effect both estrous behavior and ovarian function. The treat-
ment of mares with low doses of anabolic steroids may cause aggressive or stallionlike behavior, while high doses may inhibit ovarian activity and result in failure of follicular development and ovulation.\textsuperscript{19} The administration of anabolic steroids to prepubertal fillies may result in clitoral hypertrophy. The use of anabolic steroids should be avoided in fillies and mares intended to be used for breeding.

Progestins are commonly given to cycling mares for the suppression of estrus or synchronization of ovulation. Mares may continue to ovulate during progestin administration, especially if treatment is started late in the luteal phase. A high incidence of persistent corpus luteum formation has been noted for mares ovulating during progestin treatment.\textsuperscript{20}

4. Equine Cushing’s Disease

Mares with hypertrophy, hyperplasia, or adenoma formation in the pars intermedia of the pituitary (Equine Cushing’s Disease, or ECD) have been reported to have abnormal estrous cycles, infertility, or both.\textsuperscript{21,22} The mechanisms by which ECD cause reproductive abnormalities have not been determined. A majority of horses diagnosed with ECD are older, with the average age being approximately 20 years. Consequently, the decrease in reproductive efficiency in mares with ECD may be partly due to advanced age. Possible causes of reproductive abnormalities in mares with ECD include an increased production of androgens from the adrenal gland and compression of the hypothalamus or anterior pituitary by the enlarged pars intermedia. Both factors may lead to a decrease in gonadotropin secretion and consequently a reduction in ovarian follicular development. In addition, mares with ECD may be predisposed to uterine infections. Documentation of the effects of ECD on reproductive performance in the mare is limited.

Clinical signs of ECD include hirsutism and abnormal hair-coat shedding patterns, polyuria, polydipsia, and hyperhidrosis (Fig. 11).\textsuperscript{23} Diagnostic tests for ECD include measurements of serum glucose, insulin, adrenocorticotropin hormone (ACTH) and cortisol levels and dexamethasone suppression, ACTH stimulation, and thyrotropin-releasing hormone response tests. The measurement of single samples for basal cortisol or ACTH concentrations are of limited value in the diagnosis of ECD.

The medical management of ECD includes the administration of pergolide mesylate, a dopamine receptor agonist, at a dosage of 0.5–2 mg q 24 h per adult horse. Cyproheptadine, a serotonin antagonist, has also been used, but it may not be as efficacious as pergolide. The dose of cyproheptadine is 0.25 mg/kg q 24 h, given once in the morning.

D. Other Ovarian Abnormalities

1. Persistent Corpus Luteum

The corpus luteum that forms after ovulation is usually functional for 14–15 days in the nonpregnant mare. Corpora lutea that fail to regress at the normal time postovulation are considered to be pathologically persistent.\textsuperscript{24} Luteolysis, or destruction of the corpus luteum, occurs as a result of prostaglandin release from the endometrium. Occasionally, however, a mare may fail to regress her corpus luteum spontaneously at the normal time. The most common causes of a persistent corpus luteum are (1) inadequate prostaglandin release at days 14–15; (2) ovulations late in diestrus, resulting in corpora lutea that are immature (<5 days old) at the time of prostaglandin release; (3) embryonic loss after the time of maternal recognition of pregnancy; and (4) chronic uterine infections, resulting in destruction of the endometrium and therefore a diminished prostaglandin release.

If untreated, the corpus luteum may persist for 2–3 months. This syndrome may be suspected clinically in mares that are not expressing normal estrous behavior during the physiologic breeding season, and it must be differentiated from the syndrome of mares with silent heat. In addition, mares that have been bred and do not return to heat and are later diagnosed as not pregnant may also have a persistent corpus luteum.

Diagnosis of a persistent corpus luteum is made by an analysis of plasma progesterone concentrations or a clinical response to prostaglandin administration. Progesterone concentrations >1.0 ng/ml are indicative of the presence of luteal activity. Mares with a persistent corpus luteum will have good tone in the cervix and uterus on palpation, and the cervix will appear tight and dry on vaginal speculum examination because of the influence of progesterone.

A persistent corpus luteum will usually be eliminated by the administration of a single intra-
muscular dose of prostaglandins (PGF$_{2\alpha}$, 10 mg; cloprostenol, 250 mg).

2. Anovulatory Follicles
Ovulation failure is a normal physiologic event for the mare during the spring and fall transition periods. The development of anovulatory follicles may occasionally occur during the physiologic breeding season. Anovulatory follicles may be quite large (5–15 cm in diameter), persist for up to 2 months, and result in abnormal estrous behavior and prolonged interovulatory intervals. The cause of ovulation failure has been suggested to be endocrine in nature, either from a lack of sufficient pituitary gonadotropin stimulation to induce ovulation or from insufficient estrogen production from the follicle itself.

Anovulatory follicles may contain blood and are consequently often termed hemorrhagic follicles. The hemorrhage can be detected ultrasonically as scattered free-floating echogenic spots within the follicular fluid. The follicular fluid may form a gelatinous, hemorrhagic mass within the follicular lumen (Fig. 12). Ultrasonographically, these structures may contain echogenic fibrous bands traversing the follicular lumen (Fig. 13). A thickening of the follicular wall may be observed in some anovulatory follicles. This thickening may be associated with luteinization of the follicular wall. In some mares, plasma progesterone concentrations may be elevated over baseline levels because of the presence of luteal tissue. The administration of prostaglandins may result in the destruction of the luteal cells in those mares.

A majority of the anovulatory follicles will spontaneously regress in 1–4 weeks. Human chorionic gonadotropin (2500 IU IV) or GnRH (2.2 mg SQ) may induce ovulation or luteinization of some persistent anovulatory follicles. Unfortunately, most persistent follicles are not affected by human chorionic gonadotropin or GnRH treatment.

Pregnancy does not usually occur if a persistent follicle eventually spontaneously ovulates or is induced to ovulate. This is likely a result of degeneration of the oocyte over time. Pregnancy obviously will not occur if the follicle becomes hemorrhagic or luteinized without ovulating.

4. Discussion
A majority of ovarian abnormalities can be diagnosed with a minimum of equipment or diagnostic tests. However, some abnormalities require a more extensive evaluation, which may include blood samples for hormone analysis or karyotyping. Disorders such as persistent anovulatory follicles, persistent corpora lutea, and ovarian hematomas usually will resolve spontaneously over time if treatment is not administered. Surgical intervention is warranted for ovarian tumors that have potential effects on the contralateral ovary, cause recurrent abdominal pain, or have potential for metastasis.

A decision to remove one or both ovaries should be made only after careful deliberation. If a clear diagnosis cannot be determined on clinical signs, palpation, ultrasonography, hormone analysis, or other diagnostic tests, it may be prudent to postpone surgery until it is certain that the ovary will not return to normal function.
Appendix A: Clinical Endocrinology or Karyotyping Services for the Horse

Endocrinology
Animal Endocrinology Laboratory
P.O. Box 30076 (U.S. Postal Service)
Lansing, MI 48909–7576
or
629 West Fee Hall B (Courier Services)
Michigan State University
E. Lansing, MI 48824–1315
(517) 353-1683

BET Reproductive Laboratories, Inc.
6174 Jacks Creek Rd.
Lexington, KY 40515
(606) 273-3036
(606) 273-0178 (Fax)

Clinical Endocrinology Laboratory
School of Veterinary Medicine
University of California at Davis
Davis, CA 95616
(916) 752-0298
(916) 752-6318 (Fax)

Diagnostic Laboratory
P.O. Box 7586 (U.S. Postal Service)
or
Upper Tower Rd. (Courier Services)
New York State College of Vet. Med.
Cornell University
Ithaca, NY 14852-5786

Endocrinology Laboratory
ARBL/Foothills Campus
Colorado State University
Fort Collins, CO 80523-1683
(970) 491-1645
(970) 491–3557 (Fax)

Equitech Laboratories, Inc.
12085 Research Dr.
Alachua, FL 32615
(904)418-1525
(904) 462-0875 (Fax)

Texas Vet. Medical Diagnostic Laboratory
P.O. Drawer 3040 (U.S. Postal Service)
or
#1 Sippel Rd. (Courier Services)
College Station, TX 77841-3040
(409) 845-3414
(409) 845–1794 (Fax)

Karyotyping
Center for Reproductive Biology
Springcreek Ranch
380 S. Collierville-Arlington Rd.
Collierville, TN 38017
(901) 853-7661

Veterinary Genetics Laboratory
School of Veterinary Medicine
University of California at Davis
Davis, CA 95616
(916) 752-2111

Vivigen, Inc.
435 St. Michaels Dr.
Santa Fe, NM 87501
(505) 988-9744

References and Footnotes


