The Chronically Infertile Mare

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1. Introduction

Infertility or more appropriately, subfertility is frustrating to manage because many factors impact fertility. Seasonality, aging, systemic problems such as laminitis or Cushing's disease, nutrition, obesity, group dynamics, and environmental conditions such as temperature or rainfall can all interfere with reproduction. In addition, the estrous cycle changes dynamically from the transitional phase in the spring to recurring estrous cycles and from estrus itself to diestrus or pregnancy. Hormonal signaling must be exquisitely synchronized with physical changes of the reproductive tract and deposition of fertile semen in the uterus for pregnancy to occur. Asynchrony of these events, infection, inflammation, previous trauma to the reproductive tract, or "stress" can interfere with conception and maintenance of pregnancy. Most cases can be solved with a methodical, thorough physical and reproductive examination and appropriate diagnostic laboratory aids; however, there certainly are infertility cases that reproductive specialists would rather not see walk into the clinic. Repeated examinations may be required to identify subtle abnormalities that may occur only during certain stages of the estrous cycle. Treatment protocols should be designed around the diagnosis, and antibiotics, ecbolics, or steroids should not be used indiscriminately. The causes of chronic infertility are many (Table 1), but clients present mares for only a few reasons or "complaints." Because most of us deal with "complaints," the causes for infertility are organized under four common client complaints. They are (1) mare exhibits abnormal or no estrous cycles, (2) mare accumulates fluid in her uterus during or immediately after estrus, (3) mare has repeated uterine infections, and (4) mare looses the pregnancy after the 14-day examination. It is assumed that the mare was bred by a fertile stallion and that the dose of cooled or frozen semen used for insemination was adequate. However, this should never be taken for granted when evaluating a mare for infertility, because a subfertile stallion bred to a subfertile mare greatly decreases the likelihood of pregnancy.

2. Examination

A physical examination and evaluation of the musculoskeletal system should always be performed in chronically infertile mares because systemic abnormalities can contribute to infertility. Chronic pain, lameness, poor body condition, and obesity have been associated with abnormal or no estrous cycles, anovulatory follicles, and intra-uterine fluid accumulation. Farm management and nutritional programs should be reviewed, because group dynamics may adversely affect body condition of young or old mares, especially if mares are timid, fed in pens,

NOTES

Table 1. Chronic Infertility: Client Complaints and Their Causes

Mare not exhibiting normal estrous cycles

- Anovulatory follicles
  - Cause unknown (aging)
  - Insulin resistance, Cushings disease
  - Endometritis
  - Laminitis
  - Persistent endometrial cups
- Delayed follicular development secondary to aging
- Infection or inflammation
- Lactational anestrus
- Metabolic (insulin resistance, Cushings disease)
- Ovarian tumors
- Pain
  - Laminitis, musculoskeletal, colic
- Poor body condition
- Retention of endometrial cups

Mare has intra-uterine fluid during estrus OR immediately after breeding

- Abnormalities of the caudal reproductive tract
  - Pneumovagina
  - Recto-vaginal fistulas or tears
  - Vestibulovaginal reflux of urine
- Cervical abnormalities
  - Incompetence (does not open properly; "old maiden" mare disease)
  - Fibrosis (inhibits drainage)
  - Trauma-tears or adhesions (does not close properly)
- Uterine abnormalities
  - Adhesions
  - Delay in uterine clearance
    - Post-mating induced endometritis
      - Abnormality in uterine contractions
      - Pendulous uterus
      - Fibrotic cervix
  - Endometritis-bacterial or yeast (may be a secondary problem)
    - Diffuse or focal
  - Inadequate blood flow
    - Angeosis
      - Lymphatic lacunae
  - Foreign bodies (mummified fetus, retention of marble)
  - Persistent endometrial irritation
    - Excessive glandular secretions
    - Mucus production by epithelial cells
  - Uterine degeneration
    - Endometrial fibrosis
    - Glandular ectasia
    - Angeosis
    - Lymphatic lacunae

Mare is infected repeatedly OR infection does not resolve after repeated treatments

- List is the same as for fluid retention. Emphasis on:
  - Anatomical problems
    - Caudal reproductive tract
    - Cervical trauma or incompetence
    - Pendulous uterus
  - Chronic infectious endometritis
    - Gram negative infections
    - Yeast or fungal infections
  - Inadequate uterine perfusion

Mare looses pregnancy after 14 days and before 45 days

- Cervical tears (may be associated with endometritis)
- Chromosomal abnormalities, "old eggs"
- Endometritis (see above)
- Failure of endometrial glands - periglandular fibrosis
- Luteal insufficiency
  - Endotoxemia, laminitis, colic
- Lymphatic lacunae (may be associated with endometritis, inadequate blood flow)
- Nutritional/colic/toxins

Fig. 1. Poor perineal conformation.

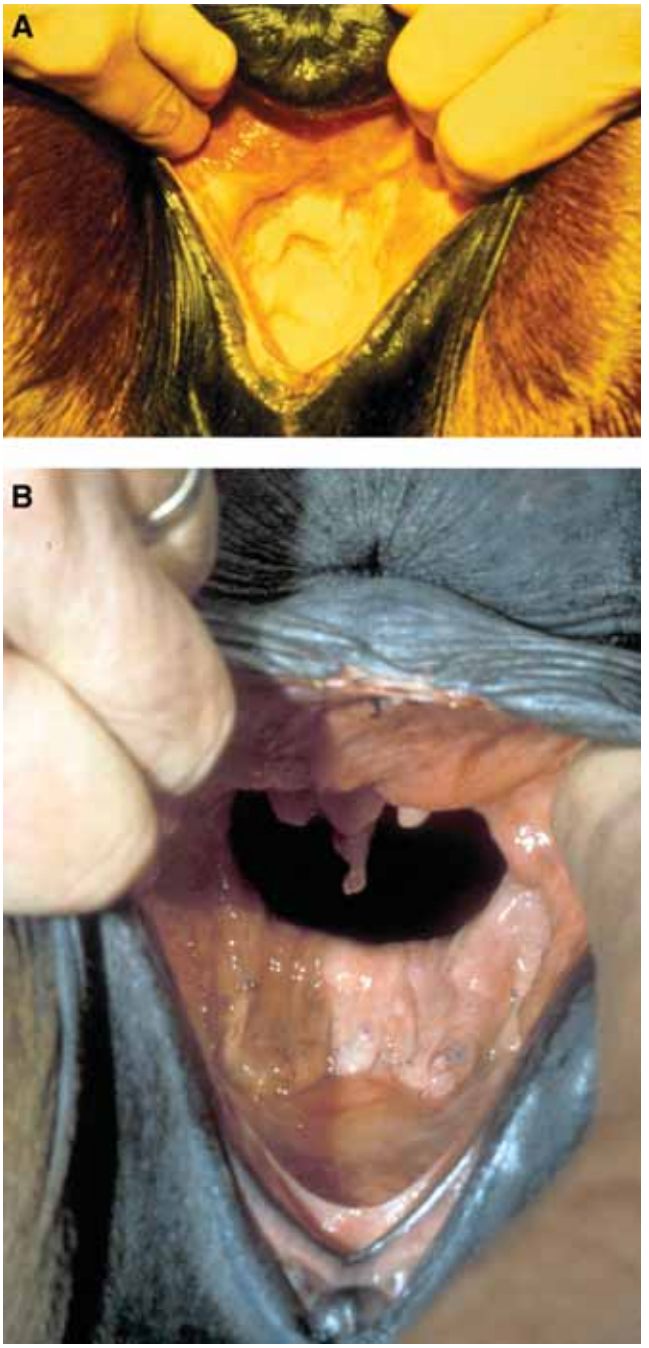

Fig. 2. (A and B) The vestibule-vaginal fold is the second barrier to contamination. Loss of the fold may result in chronic uterine inflammation.

crowded during feeding, or not given shelter from cold or wet conditions. As complete of a history as possible should be recorded because it provides important information on the possible causes of the infertility and the diagnostics that should be performed. Medications given during the mare's performance career, age when first bred, past foaling history, treatments administered around breeding, type of semen used (fresh, cooled, or frozen), and manner by which the mare was bred (artificial insemination or natural mating) should be recorded. Because semen quality may contribute to the mare's fertility problem, pregnancy rate/cycle or the stallion's past reproductive performance should be obtained.

The reproductive examination should consist of an evaluation of the mare's conformation, an examination of the reproductive tract through rectal palpation and ultrasonography, a vaginal speculum, and a manual cervical examination.[1] Laboratory diagnostics are usually determined from the examination findings and include uterine culture and cytology, endometrial biopsy, and/or endoscopic evaluation of the uterine lumen. Findings need to be recorded to formulate a proper management plan and to allow for subsequent review.

The anatomy of the pelvic region and perineum of the mare must be evaluated critically, because most uterine infections are caused by bacteria that normally inhabit the caudal reproductive tract ascending into the anterior vagina. The perineum is best evaluated during estrus when relaxation and elongation of the vulvar lips are at their greatest point. Mares with >2.5 cm of vulva open dorsal to the pelvis and those with a cranial ventral tilt of $>20°$ should receive a vulvoplasty (Caslick) to prevent pneumovagina and/or aspiration of feces into the vagina (Fig. 1). The vulvar lips should be parted, and the vestibulovaginal fold should be evaluated. This fold of tissue is the second barrier to uterine contamination (Fig. 2). It may be stretched or torn in mares that have had a large foal, in pluriparous mares after delivering many foals, or in a mare bred by an aggressive stallion. If air enters the anterior vagina when the vulvar lips are parted in a mare that experiences repeated bouts of chronic bacterial endometritis, a vestibuloplasty (also referred to as a deep Caslick or a Gad operation) should be considered.

The rectal examination provides information as to when a mare may ovulate, where she is in her es-

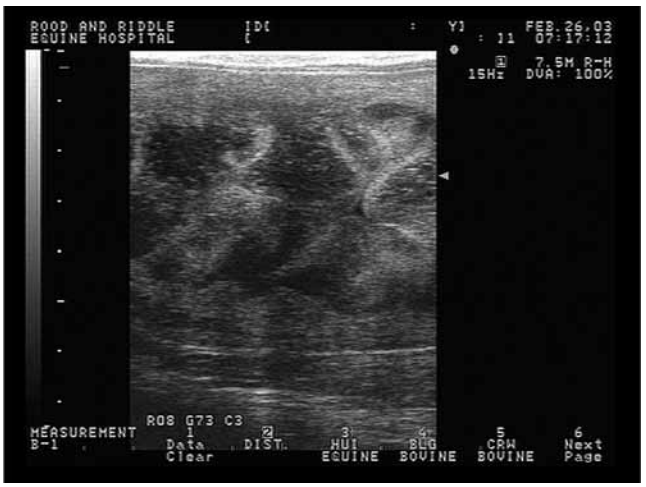

Fig. 3. Excessive endometrial edema after ovulation.

trous cycle, if she is pregnant, if there is cervical, uterine, or ovarian pathology, and if there is an inconsistency between cervical findings, uterine tone, and follicular development. When evaluating the reproductive tract through rectal palpation, the length and width of the cervix (recorded in inches or centimeters) and the location of the uterus in relationship to the pelvis should be recorded. Mares with cervical fibrosis will have an elongated, narrow cervix during estrus (length > 5–6 cm, width = 1–2 cm), whereas mares with a torn cervical muscle will have a short (<3–4 cm), wide (>4–5 cm) cervix that is not well defined, even during diestrus. Mares with a uterus located >15 cm below the pelvis may accumulate intra-uterine fluid after breeding, because they are unable to clear inflammatory debris. Uterine tone that is thick, doughy, edematous, and flaccid is associated with physiological changes in the estrous cycle, seasonality, and pathological processes. Size, shape, and turgidity of follicles on the ovary are used to predict ovulation and identify abnormal structures such as suspected anovulatory follicles or tumors.

Repeated ultrasonographic evaluations of the reproductive tract may be needed to identify the cause of infertility because the reproductive tract changes dynamically during the estrous cycle; therefore, subtle abnormalities may be noted only during a specific time of the cycle. Inconsistencies in the relationships between follicular growth, ovulation, uterine edema pattern, and cervical relaxation give clues as to why the mare may be infertile. Mares with excessive uterine edema on day 1 or 2 of estrus or mares that accumulate a small amount of fluid cranial to the cervix may not dilate their cervix properly or may have chronic inflammation, whereas mares that retain uterine edema after ovulation may have lymphatic lacunae, endometritis, angeosis, or all three abnormalities[2] (Fig. 3).

A vaginal speculum examination should always be performed in infertile mares, because the integrity of the vestibulo-vaginal sphincter, the presence of fluid such as pus or urine in the vaginal vault, and any discrepancies between cervical relaxation and stage of estrous cycle and the color and moisture of the vaginal mucosa are best identified visually. The external os of the cervix may be adhered to the vaginal fornix or the cervix may be inflamed, closed, and located off the vaginal floor during estrus in a mare with endometritis.

Manual examination of the cervix is often overlooked, although cervical abnormalities are a common cause of chronic infertility. Incomplete dilation of the cervical canal during estrus results in fluid accumulation both before and after breeding in old, maiden mares. Cervical trauma at foaling may result in a cervical tear, cervical fibrosis, or adhesions between the external os and vaginal fornix or within the canal itself. Mares that present repeatedly with uterine infections, intra-uterine fluid, a history of early embryonic death, or abortion should be considered suspect of cervical malfunction. If a cervical tear or adhesion is suspected during estrus, a digital examination of the cervix should be conducted when the mare is under the influence of progesterone. Fibrosis of the cervical canal is best identified during estrus, because the cervical canal will be tubular, narrow, lack elasticity, and elongated. The canal may also deviate laterally.

3. Laboratory Diagnostics

Uterine cytology and culture are the laboratory diagnostics most commonly performed on barren mares. In some cases, endometrial biopsy and/or uterine endoscopy are needed to identify the cause of the infertility. Uterine culture samples are commonly obtained with a guarded culture swab, whereas cytological specimens can be obtained by passing a second swab into the uterus, collecting cells on the cap of a guarded swab, or using a cytology brush.

Uterine cytology may be the most accurate method of diagnosing infertility associated with endometritis. Riddle et al.[3] identified twice as many mares with endometritis by endometrial cytology than by culture swab. They also found that the degree of inflammation (number of neutrophils/high power field) adversely affected 28-day pregnancy rates more than the mere presence of neutrophils in a cytology smear. The 28-day pregnancy rate of mares with >5 neutrophils/field was only 23% compared with a 36% rate in mares with 2–5 neutrophils/field and a 60% pregnancy rate in mares with 0–2 neutrophils/field. In addition, the correlation between cytological findings and isolation of bacteria differed between micro-organisms. Recovery of beta-hemolytic *Streptococcus*, *Staphlococcus*, or *Klebsiella* were more likely to be associated with a positive cytology (defined as having >2 neutrophils/field) than recovery of *E. coli*, *Pseudomonas*, or *Enterobacter cloaca*.[3] Isolation of micro-organisms was associated with reduced pregnancy rates, even

in the apparent absence of inflammation. Additionally, false positives and false negatives occur, and correlation between cytology and culture results varies between organism recovered. Therefore, these findings stress the importance of interpreting laboratory data in relationship to clinical signs.

Obtaining a uterine culture with a guarded cotton swab may not always identify mares with bacterial endometritis. Neilsen[4] recently reported that only 38 of 84 mares (45%) with bacteria isolated from the surface of an endometrial biopsy had bacteria isolated from a uterine culture swab. Possible causes for this poor correlation is that only a small area of endometrium is cultured with a guarded swab, focal infections are missed, or the mare has a pendulous uterus and the affected area is not sampled. We have recently reevaluated the small-volume uterine-flush technique as a method for culturing the uterus of chronically infertile mares.[5] The technique was first described by Ball et al.[6] in the 1980s, and the study found it to be more sensitive in identifying mares with chronic endometritis than a culture swab. Results from our evaluation of 309 chronically infertile mares indicated that flush culture was twice as sensitive as swab culture previously estimated by Neilsen[4] (0.71 versus 0.34, respectively) in identifying endometritis when the same "best standard" (presence of neutrophils within the endometrium) was used.[4,5] Therefore, flush culture doubled the ability to detect chronic endometritis in mares based on culture alone. The improved sensitivity resulted from improved detection of gram-negative organisms, because recovery of β-hemolytic *Streptococcus* from uterine flush was similar to that reported previously when samples were collected by culture swab. The false-positive rate of the technique was 11% if a false positive was defined as a positive culture result in the presence of a clear efflux and a cytological smear that contained no debris, neutrophils, or bacteria.

A small-volume flush can be performed during estrus or diestrus. If the sample is collected during diestrus, it is recommended that the mare be given prostaglandin so that she returns to estrus. Before the procedure, the perineum should be scrubbed, rinsed, and dried. A sterile uterine catheter is passed per vaginum into the uterus by an examiner whose arm is covered by a sterile sleeve; 10–12 cm of catheter should be inserted into the uterus. Sterile saline is then infused into the uterus by either attaching a 60-ml catheter-tip syringe containing 60 ml of saline onto the catheter or attaching a 150-ml bag of sterile saline to the catheter. The uterus is then manipulated by transrectal palpation to distribute the fluid throughout the uterine lumen. The uterine horn containing the catheter tip is then cradled by the veterinarian's hand, and the saline is either drained into a sterile 50-ml conical tube by gravity flow or is collected into the 150-ml bag of sterile saline. Clarity of the efflux is recorded. Recovery of a cloudy flush or a flush with mucoid particles is highly associated with isolation of bacteria, and therefore, treatment of the uterus can begin immediately if the sample is not clear. After collection, the precipitant is allowed to settle, or the sample can be centrifuged at 400 *g* for 10 min. The supernatant is poured off to leave ~5 ml of fluid. Two sterile cotton-tipped swabs are placed into the pellet in the bottom of the tube. One sterile cotton swab is used for uterine culture, whereas the second is rolled over a glass slide for cytology. Cytological smears should be evaluated for debris, neutrophils, and bacteria. The amount of debris covering the slide (0–25%, 25–75%, or >75% of the slide contains debris [i.e., takes up stain]; Fig. 4) is associated with recovery of bacteria.[5] The small-volume flush technique is not as sensitive as a dry swab or brush cytology in identifying neutrophils because the fluid dilutes out the number of inflammatory cells per high power field. However, if there are neutrophils or bacteria on the slide, there is a ≥80% chance of recovering bacteria from the culture.

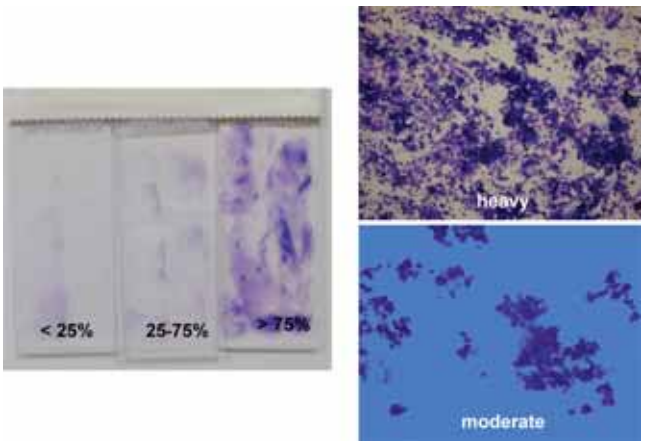

Fig. 4. The amount of debris in a cytological smear is estimated by the percentage of the slide that takes up Diff quik stain.

Endometrial biopsy is a valuable tool for identifying the cause of infertility. However, many practitioners have avoided using the technique because the histological interpretation they receive does not change mare management. If endometrial biopsy specimens are interpreted by a veterinary pathologist or theriogenologist with expertise in equine reproductive pathology, the specimen provides valuable information on the severity of uterine degeneration, lymphatic drainage, uterine irritation and inflammation, and glandular function (Fig. 5).[7–9] Findings are used to formulate management protocols and are helpful in determining if previous treatments were successful. Endometrial biopsies are most useful in identifying pathology in mares that are presented without an adequate history, mares with palpable abnormalities in uterine tone on rectal examination, and mares with chronic inflammation or repeated early embryonic death; they also help to identify if treatment was successful in

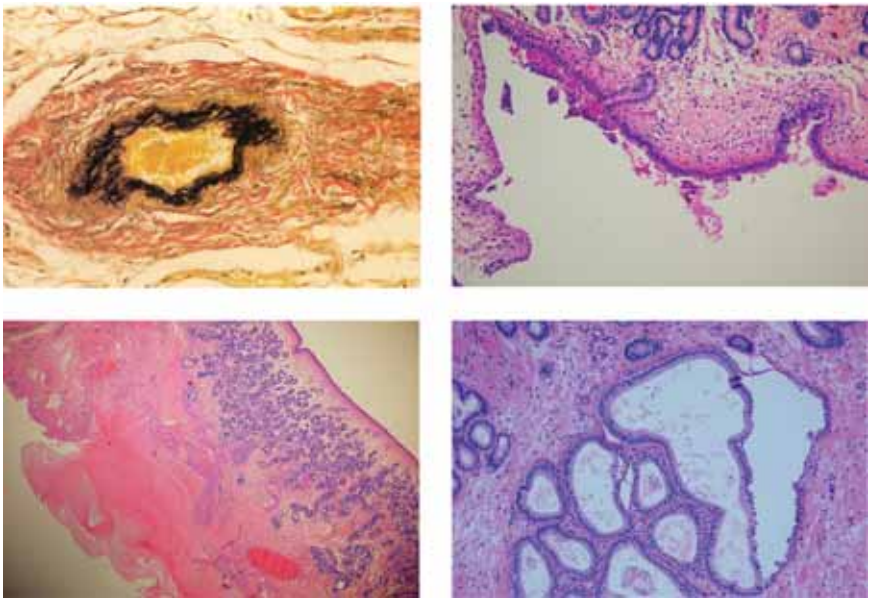

Fig 5. Various pathologies observed in endometrial biopsies: angeosis (top left), mucus and inflammation (top right), severe endometrial edema (bottom left) and glandular ectasia, inspissated material and mild fibrosis (bottom right).

clearing inflammation.[7–9] Endometrial biopsies can be obtained during estrus or diestrus. Samples should be placed in Bouin's solution (pitric acid) for 8–24 h after which time the sample is placed in 70–90% alcohol. Signalment and history should always be provided.

Uterine endoscopy is limited to mares that may have focal infections, uterine adhesions, retention of endometrial cups, or foreign bodies. It is also useful in identifying mares with endometritis that are presented for a history of not exhibiting estrous cycles. The procedure can be performed during diestrus or in early estrus. The uterine lumen may be filled with either air or saline. Visibility is best with air insufflation; however, air can be irritating to the endometrium. On completion of the examination, the infused air should be removed through a catheter or pump, the uterus should be lavaged with saline, and in some cases, antibiotics should be infused if the mare has a history of endometritis.

4. Management of Mares Not Exhibiting Normal Estrous Cycles

Abnormal estrous cycle length or lack of estrous cycles during the cyclic season may be associated with uterine infection, aging, poor body condition score (<4 of 9), pain, ovarian tumors, anovulatory follicles, systemic condition such as Cushing's disease or insulin resistance, and ovarian senility. Conditions that affect the mare systemically such as Cushing's disease or laminitis can adversely affect hormonal signaling or be associated with secondary endometritis. Aging affects cyclicity in that old mares (>18 yr of age) have shortened cyclic seasons and prolonged follicular phases; additionally, they may ovulate small follicles before they are bred. Old mares may require additional care, special diets, blanketing in cold weather, or treatment of arthritic conditions to advance the first ovulation of the year. Ambient temperatures consistently below freezing in the spring may not only affect the timing of first ovulation of the year, but it may also interfere with cyclic patterns in mares that have begun to exhibit estrous cycles, especially if the mares are thin, shipped from a warm to cold climate, or are lacking a winter hair coat. Diagnostics for mares that are not cycling properly are determined from the mare's history, body condition score, physical examination, and reproductive examination. Endocrine profiles, uterine cytology, uterine culture, or endoscopic evaluation of the uterus may be needed. Hormonal therapy may be required, because the neuroendocrine signals from the brain may not be synchronized or reaching the reproductive tract.

Anovulatory Follicles

A frustrating but common cause of abnormal estrous cycles is anovulatory follicles. They are more common in aged mares than in young mares (<10 yr of age)[10] and tend to occur more frequently later in the breeding season, possibly as a result of chronic inflammation, infection, or repeated use of prostaglandin. Anovulatory follicles have also been associated with laminitis and insulin resistance. Theories for their development include an inability of the follicle to respond to luteinizing hormone (LH), prostaglandin release from chronic endometritis, and a high plasma cortisol and/or insulin concentrations associated with pain or obesity. Mares

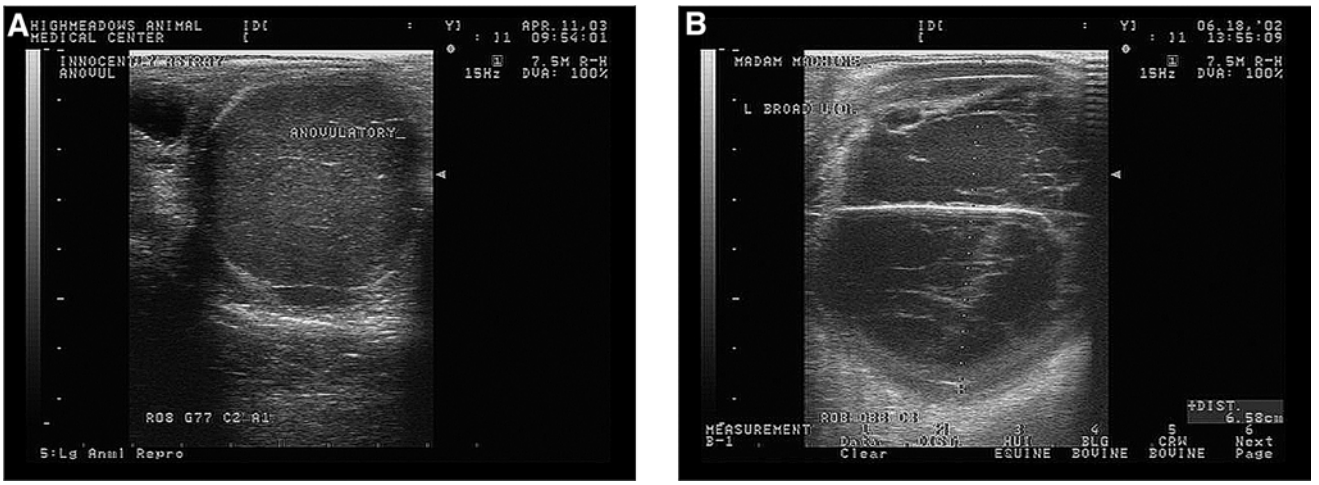

Fig. 6. (A and B) Anovulatory follicles can be filled with a hyperechoic fluid or can contain fibrous strains.

that develop an anovulatory follicle exhibit normal estrous behavior; however, estrus is prolonged, and the dominant follicle continues to grow even after it is expected to ovulate. On ultrasonographic evaluation, anovulatory follicles are usually >45 mm in diameter and have hyperechoic particles within a fluid-filled lumen, fibrous strains within the lumen, and/or a thick white rim along the perimeter of the follicle (Fig. 6, A and B). Treatment with either human chorionic gonadotrophin (hCG) or deslorelin (GnRH agonist) is futile, because anovulatory follicles do not respond to ovulatory agents; ~65% of anovulatory follicles will eventually produce progesterone, whereas the remaining 35% do not.[10] Anovulatory follicles that produce progesterone respond to exogenous prostaglandin, and mares given prostaglandin will return to estrus. Unfortunately, many will form another anovulatory follicle during the next estrus.

Infection or Inflammation

Uterine infection should always be included on a list of differentials in mares that are not exhibiting normal estrous cycles or are anestrus during the cyclic season (see section 6).

Metabolic Problems

Insulin resistance or pituitary tumors may result in abnormal estrous cycles, anestrus, development of anovulatory follicles, and/or endometritis. Mares with chronic foot pain, obese mares, mares exhibiting hirsutism, polydipsia, or polyuria, and mares with repeated bacterial or yeast infections should be tested for insulin resistance and pituitary tumors. Diagnostic tests include dexamethasone suppression, measurement of adrenocorticotropic hormone (ACTH), cortisol, and insulin in plasma samples. Mares treated with pergolide need to be monitored closely, because doses >2 mg daily may result in anestrus. Pergolide is a dopamine agonist and may inhibit secretion of GnRH.

Lactational Anestrus

This condition occurs most commonly in mares that foal before April 1 in the Northern Hemisphere. It has been associated with the seasonal effects of light, chronic pain, subclinical endometritis, and metritis. Mares typically exhibit a normal foal heat and ovulate a follicle, but they do not return to estrus at the 30-day heat or do not respond to injections of prostaglandin. Ovaries of afflicted mares may become small and hard, or they may contain multiple follicles that are <15–20 mm in diameter. Diagnostics are directed at determining if factors other than seasonality are contributing to the condition. Mares should be examined for musculo-skeletal pain, especially if they have experienced a difficult foaling. Treatment with non-steroidal anti-inflammatory drugs, chiropractic manipulations, or acupuncture may be helpful. The uterus should be cultured, preferably by a small volume uterine flush, to determine if bacteria, mucus, or white blood cells are present. Appropriate uterine treatments should be instituted (see endometritis) if cytology or culture results indicate inflammation or infection. Uterine infection may be secondary to pain because mares tend to limit their mobility, which results in intra-uterine fluid retention. Treatment of the primary problem (i.e., pain or endometritis) frequently results in the formation of follicles >15 mm. If follicular development does not proceed normally, mares may be treated with equine follicle stimulating hormone (eFSH), sulpiride, or domperidone after follicles reach a size of 18–20 mm. To induce follicular development, eFSH is given at a dose of 6.5 mg two times a day in the muscle until the largest follicle is 30 mm in diameter, usually 5–10 days. The mare is given hCG (2000–3000 IU, IV) when the largest follicle is 32–35 mm in diameter.[11] Unfortunately, the availability of eFSH is limited at this time. Sulpiride has been shown to induce cyclicity in mares in transition,[12] and it seems to be effective in inducing cyclicity in mares during lactational anestrus. Best results

are obtained if the mare is subjected to 14.5 h of light for 2 wk before the first injection of sulpiride (400 mg, IM, one time daily). The drug is given for 10–16 days. If there is no response after 16 days, the drug is discontinued. Reserpine and domperidone, both dopamine antagonists, have also been administered to mares in lactational anestrus and have had some success.

If seasonality is the cause of the anestrus condition, afflicted mares usually exhibit estrous cycles in late April or early May. To prevent lactational anestrus from reoccurring in subsequent pregnancies, pregnant mares sensitive to day length should be subjected to 14.5 h of light daily beginning on the 15th of December.

Retention of Endometrial Cups

Although an uncommon problem, retained endometrial cups have been reported in mares that had aborted or foaled in a previous year.[13] Affected mares have ovaries that contain many luteinized follicles, clusters of large (>35 mm) follicles, and corpora lutea. Mares with retained cups may exhibit no estrus or irregular or prolonged estrus behavior. A diagnosis of retained endometrial cups can be made by ultrasonography or by measuring equine chorionic gonadotrophin. Retained endometrial cups will eventually slough, although they may take months. Endometrial cups have been removed by laser surgery; however, complications including endometritis, uterine hemorrhage, and inability to remove all cups at one session because of limited visibility from smoke have been reported.

5. Accumulation of Intra-Uterine Fluid by the Mare During Estrus or After Breeding

Intra-uterine fluid is a clinical sign and not a diagnosis. Causes include impaired cervical function, post—mating-induced endometritis, a dependent, pendulous uterus, impaired lymphatic drainage, angeosis, infection, or a combination of the above.[8,14–19] The location of fluid accumulation within the uterine lumen, if it is visualized in the uterine body, near the cervix, or within the uterine horns, provides a clue as to the cause. Mares with primary cervical incompetence tend to retain fluid just cranial to the cervix, because myometrial contractions push the fluid towards the cervix; however, the cervix impedes flow into the vagina. The condition most commonly affects young or old nulliparous mares or mares with a history of a traumatic foaling. Mares susceptible to post–mating-induced endometritis accumulate fluid in the uterine horns and body as a result of impaired myometrial contractions, poor lymphatic drainage, excessive glandular secretions, angeosis, a dependent uterus, and possibly, abnormalities in hormonal and neurological signaling.[14–18] They tend to be pluriparous and >12 yr of age with a pendulous uterus. Some mares with post-mating endometritis may also have cervical adhesions or cervical fibrosis that will impair uterine clearance.

Mares that retain intra-uterine fluid for >18 h after breeding experience decreased pregnancy rates.[20,21] If repeatedly bred, these mares may develop bacterial or yeast infections. Chronic endometrial irritation may result in excessive mucus production, which further contributes to fluid accumulation.[20,22,23] Susceptibility to post—mating-induced endometritis can occur gradually because of degenerative uterine changes, acutely as a consequence of a traumatic foaling, or secondary to repeated manipulations from sequential embryo flushes.

Unfortunately, identifying mares susceptible to post-mating endometritis may be difficult, because some mares show no signs of inflammation before mating only to accumulate fluid after mating. Afflicted mares are diagnosed using a combination of diagnostic techniques including rectal palpation, ultrasonographic examination of the reproductive tract, cervical and vaginal examinations, and evaluation of the mare's conformation. The position of the uterus in relationship to the pelvis and the length and width of the cervix should be noted because mares with a pendulous uterus or those with an elongated, fibrotic cervix or a narrow, long cervix are prime candidates for post—mating-induced endometritis. A small amount of intra-uterine fluid during estrus and before breeding may not adversely affect pregnancy rates. However, fluid volumes >2 cm have been associated with decreased pregnancy rates.[24] The presence of intraluminal fluid ≥18 h after breeding or retention of uterine edema after ovulation is diagnostic of defective uterine clearance. An ultrasonographic examination of the reproductive tract conducted 12–24 h after breeding will determine if additional therapy is needed.

Treatment for post-mating endometritis is aimed at improving physical clearance of inflammatory byproducts associated with insemination, reduction of the length of the inflammatory response, and prevention of bacterial infection. Physical clearance of inflammatory debris has been improved with uterine irrigation and administration of oxytocin or cloprostenol.[20,21,25–31] The inflammatory response has been modulated with dexamethasone and immunomodulators, whereas intra-uterine antibiotics have been used to prevent bacterial infections.[27,28,32–38] Uterine irrigation combined with an ecbolic drug, oxytocin (10–20 IU, IV or IM), or cloprostenol (250 μg, IM) given between 4 and 8 h post-breeding has increased pregnancy rates in highly susceptible, barren mares.[38,39] Uterine irrigation is most commonly performed post-mating if mares have >2 cm of intra-uterine fluid.[24] If fluid accumulations are smaller or there is residual endometrial edema after ovulation, treatment with only oxytocin or cloprostenol may be needed. The two ecbolics differ in the amplitude and length of uterine

contractions. Oxytocin induces high-amplitude uterine contractions for ~30 min during estrus and in the 48-h period after ovulation,[26] whereas cloprostenol, a prostaglandin analog, is associated with contractions of lower amplitude that last for ~5 h.[29,40] Cloprostenol induces premature luteal regression, and therefore, its use should be limited to estrus and within 12 h of ovulation.[41–43]

Administration of cloprostenol has been advocated for lymphatic stasis because of its prolonged effect on uterine contractility. Lymphatic stasis is associated with angeosis, a pendulous uterus, aging, and in some cases, cervical incompetence.[8,19] A recommended treatment is to irrigate the uterus between 4 and 8 h after mating and then give 10–20 IU of IV oxytocin. Between 12 and 18 h after mating, the mare is given 250 μg of cloprostenol IM. The rationale for using both ecbolics is that oxytocin induces strong uterine contractions for ~30 min, which results in rapid clearance of intra-uterine fluids[26]; however, prostaglandin $F_{2\alpha}$ produces low-amplitude contractions that persist for 4–5 h.[29,38,39] Prolonged low-amplitude contractions may assist in lymphatic drainage, because lymphatic vessels do not contain smooth muscle and must rely on uterine contractions for emptying.

Exercise

Exercise is helpful in draining the uterus. Increased intra-abdominal pressure associated with movement helps to evacuate uterine contents. Broodmares restricted to a stall because of an ill neonate or injury and mares with laminitis or foot pain accumulate fluid in their uterus after breeding. If exercise must be limited, small injections of oxytocin given every 4–6 h may assist in fluid clearance.

Corticosteroids

The length and severity of the inflammatory response may be diminished by administration of corticosteroids given before or after breeding.[32,33] A single injection of dexamethasone administered at breeding (50 mg, IV) has resulted in decreased uterine fluid accumulation and increased pregnancy rates in mares with abnormal reproductive parameters such as pneumovagina, pendulous uterus, and cervical incompetence.[33] Oral administration of prednisolone (0.1 mg/kg) given every 12 h for 4 days beginning 48 h before breeding has resulted in decreased neutrophil numbers in uterine fluids of barren mares, decreased uterine fluid, increased fluid clarity, and increased pregnancy rates.[32]

Intra-uterine antibiotics have been infused into the uterus after natural mating since the mid 1970s to prevent bacterial endometritis. Clinical studies and field experience have shown effectiveness of a single post-mating antibiotic treatment in combating endometritis.[27,28] Pregnancy rates were similar in mares treated with a single dose of oxytocin or a single dose of broad-spectrum antibiotics, but they were highest in mares who received both, which indicates that the two treatments may result in an additive benefit.[27,28] Others have shown that treatment of mares susceptible to endometritis with intra-uterine saline lavage, prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), or penicillin, IV, 12 h after an intra-uterine infusion of *Streptococcus equi* spp. *zooepidemicus* were equally effective in eliminating bacteria from the uterus.[25]

6. Repeated Infection or Infection That Does Not Resolve After Treatment in the Mare

Chronic Infectious Endometritis

Chronic infectious endometritis is most commonly seen in older (>12 yr of age), pluriparous mares with defects to their perineal conformation. It may also occur in mares that are susceptible to post—mating-induced endometritis that are not treated after mating, in embryo-donor mares that are repeatedly manipulated for embryo recovery, and in mares with cervical incompetence. Treatment for chronic infectious endometritis should include removal of the offending organism through uterine lavage, ecbolics, and antimicrobial therapy in addition to repair of anatomical defects. It is recommended that mares with bacterial endometritis be treated with intra-uterine antibiotics for 3–7 days; however, there is limited scientific data to support various treatment protocols. Treatment length depends on the chronicity of the infection, the bacteria isolated, the mare's ability to clear uterine fluid, and her history. It is recommended that the uterus be irrigated for the first 2–3 days of treatment before infusion of antibiotics to remove inflammatory debris because some antibiotics are chemically altered when the pH is excessively high or low or if there is gross debris. For example, aminoglycosides are most active in an alkaline environment.[44] Increased local acidity secondary to tissue damage or bacterial destruction may account for failure of aminoglycosides to kill usually susceptible micro-organisms. In addition, purulent debris binds to aminoglycosides and inactivates them.[44]

Intra-uterine antibiotic therapy for endometritis should be performed during estrus because its use during the progesterone phase has resulted in fungal infections and infections with resistant bacteria.[45,46] The status of the reproductive tract should be reassessed after the last intra-uterine treatment to determine if there is residual intra-uterine fluid, excessive uterine edema, and ovulation. A thorough reproductive examination should be conducted at the next estrus to determine if treatment was successful. If bacteria are isolated, systemic therapy combined with local therapy should be considered. Tables 2 and 3 include dosages of intra-uterine and systemic antibiotics for treatment of bacterial endometritis. Treatment failure is most common when dealing with fungal or chronic gram-negative bacterial infections. Failure may be asso-

Table 2. Guidelines for Administration of Intra-uterine Antibiotics

Drug	Dose per Infusion	Comments
Amikacin	2 g*	Buffer with bicarbonate or large volume of saline (200 ml); Excellent gram negative coverage
Ampicillin	2 g†	Use only the soluble product; susceptible gram-positive and *E. coli*
Ceftiofur sodium	1 g	Resistant to many beta-lactamases; broad spectrum; save for resistant organisms
Gentamicin	1–2 g*	Buffer with bicarbonate or large volume of saline (200 ml); Effective against some *S. zooepidemicus; Enterobacter* spp., *E. coli, Klebsiella* spp., *Proteus* spp., *Serratia* spp, *P. aeruginosa, S. aureus*
Penicillin (potassium)	5 million U	*S. zooepidemicus*
Neomycin	4 g	Gram-negative organisms; Some *E. coli* and some *Klebsiella* spp.
Ticarcillin	6 g	Gram-positive organisms; effective against some *Pseudomonas* spp. Infuse with a minimum of 200 ml of saline
Ticarcillin—clavulanic acid	3–6 g	Beta-lactamase inhibitor confers greater activity against *Enterobacter; S. aureus, B. fragilis;* Infuse with a minimum of 200 ml of saline

*Buffered with equal volume of 7.5% bicarbonate and diluted in saline.
†Use at high dilutions because it can be irritating.

ciated with continual contamination of the uterus because of loss of anatomical barriers, inappropriate dosage regimen, drug resistance, inappropriate microenvironment (biofilms, mucus, and breakdown of intra-uterine antibiotics), or impaired host's defense mechanisms. When response to treatment is not as expected, the mare should be reevaluated in an attempt to determine cause.

Factors to be considered in the selection of antimicrobials for treating reproductive conditions include susceptibility of the microorganism, local versus systemic treatment, concentration of drug attainable at site of infection, and effects of the drug on immediate and future fertility.[1] Antimicrobial selection should be based on culture and sensitivity results. Unfortunately, in vitro and in vivo efficacy may not always be equivalent. For example, most uterine cultures of *Pseudomonas* spp. show in vitro sensitivity to polymyxin B, yet few mares with endometritis caused by *Pseudomonas* spp. respond well to polymyxin-B therapy. *Streptococcus equi* spp. *zooepidemicus* shows in vitro sensitivity to trimethoprim-sulfonamide combinations but rarely does the drug effect a clinical cure.

Drug compatibility is an important principle of therapy. The polypharmacy approach of multiple antibiotics and antiseptics mixed together may be of no therapeutic value and could be harmful. Penicillin, gentamicin, ampicillan, or ticarcillin should not be mixed, because physical and chemical incompatibilities exist.[44] Penicillin and ampicillan are inactivated when the pH of a solution is <5.5 or >8, and therefore, they should not be mixed with sodium bicarbonate, gentamicin, or sulfonamides. Gentamicin is physically incompatible with beta-lactams, and they should never be mixed in the same syringe.

Complications may arise with intra-uterine antibiotic treatment. Mares may develop secondary bacterial or fungal infections, exhibit severe endometrial irritation, or fail to resolve an infection. Treatment for one pathogen may result in the proliferation of another pathogen that is often more difficult to manage than the original. An example is treatment of a streptococcal infection that, after treatment with the appropriate antibiotic, is followed by development of a yeast or *Pseudomonas* infection. In these cases, a mixed infection may have existed initially, and antibiotic use merely allowed proliferation of the other organism. In other cases, a second organism may be introduced accidentally during the course of treatment for the primary infection.

Administration of systemic antibiotics should be considered in chronic infectious endometritis and in mares with defects in the caudal reproductive tract. Systemic administration of antibiotics results in higher minimal inhibitory concentrations (MIC) throughout the genital tract compared with intrauterine therapy. In addition, there is less likelihood of super infections secondary to changes in vaginal flora, the antibiotics are not degraded by

Table 3. Antibiotics for Systemic Treatment of Bacterial Endometritis

Drug	Dosage	Route	Interval	Comments
Amikacin sulfate	10 mg/kg	IV or IM	24 h	Excellent gram-negative coverage
Ampicillin sodium	29 mg/kg	IV or IM	12–24 h	Susceptible gram positive organisms and *E. coli*
Ceftiofur	2.5 mg/kg	IM	12–24 h	Broad spectrum gram-positive and some gram-negative organisms
Gentamicin	6.6 mg/kg	IV	24 h	Slow intravenous infusion; *Enterobacter* spp., *E. coli, Klebsiella* spp., *Proteus* spp., *Serratia* spp., *P. aeruginosa, S. aureus*
Enrofloxacin*	5.5 mg/kg	IV	24 h	Slow intravenous infusion; gram-negative infections caused by susceptible bacteria resistant to alternative, first-choice drugs; seminal vesiculitis; epididymitis
	7.5 mg/kg	PO		
Penicillin G (potassium)	25,000 IU/kg	IV	6 h	Synergistic with aminoglycosides; Do not store mixed in syringe for >12 h; do not mix in syringe with gentamicin; *S. zooepidemicus, Leptospirosis*
Penicillin (procaine)	25,000 IU/kg	IM	12 h	As for above
Trimethoprim-sulfonamide	30 mg/kg (combined)	PO	12 h	*S. aureus, E. coli, Klebsiella* spp. *Proteus;* some *Nocardia* spp.

*Should not be used in pregnant mares or in young growing horses because of the risk of arthropathy.

conditions in the uterine lumen, and parental therapy does not irritate the endometrium. The length of systemic treatment is not dictated by the estrous cycle, and antibiotics may be given for 7–10 days if deemed necessary. Systemic therapy eliminates the need to invade the vestibule, vaginal canal, and cervix. The vestibule and clitoral fossa harbor a vast array of bacteria, even in reproductively normal mares. These organisms serve as a source of uterine inoculation when the hand or an instrument is passed through the vulva to cannulate the cervix during intra-uterine infusion.[47] Care should be taken when treating uterine infections with systemic antibiotics, because gut flora may be adversely affected.

Immunomodulators

Immunomodulators induce a non-specific cell-mediated response predominantly by activation of macrophages and release of cytokines that elicit a general increase in immune-system activity.[48] Two immunostimulants are currently labeled and marketed for use in horses. A cell-wall extract of *Mycobacterium phlei*[a] has been approved as an adjunctive treatment in mares with uterine infection caused by *Streptococcus equi* and *Propionibacterium acnes,*[b] and they have been used as an adjunct treatment for horses with equine respiratory disease complex. The effect of *Propionibacterium acnes* on live foal rates was recently evaluated in a controlled field study.[34] Pregnancy and live foal rates were improved after *P. acnes* was used as an adjunct to conventional treatments in mares with a cytologic diagnosis of persistent endometritis. The optimal effect was detected in mares bred during the interval extending from 2 days before to 8 days after the first treatment with *P. acnes*.

The effects of *Mycobacterium phlei* cell-wall extract in modulating the immune response of mares susceptible to endometritis has been compared with reproductively normal mares.[35–37] These studies showed that (1) mares susceptible to endometritis have higher levels of pro-inflammatory cytokines and lower levels of IL-10, a cytokine that inhibits production of pro-inflammatory cytokines, in their endometrium before breeding than reproductively normal mares, (2) after insemination, susceptible mares expressed significantly higher levels of pro-inflammatory cytokines in their endometrium 7 days after ovulation than normal mares, and (3) susceptible mares treated with *Mycobacterium phlei* at the time of artificial insemination had an endometrial immune environment similar to that found in reproductively normal mares.

These studies indicate that immunomodulation may help restore homeostatic local inflammatory

mechanisms. This may assist in the prophylaxis of post—mating-induced endometritis or chronic infectious endometritis.[36,37]

Intra-Uterine Antiseptics and Chelating Agents

A variety of preparations have been infused into the uterus of mares in attempts to treat uterine infection, including vinegar, betadine, iodine, dimethylsulfoxide (DMSO), kerosene, chlorhexidine-gluconate, magnesium sulphate, and hypertonic saline.[49] Care should be taken when using these preparations, because they may cause inflammation, necrosis, and reproductive sterility. Some antiseptics can be administered safely if given at appropriate concentrations. A dilute povidone-iodine solution (0.05% in 5 ml of 10% povidone-iodine in 1 l of saline) 4 h post-mating had no adverse effects on pregnancy rates in reproductively sound mares compared with saline and did not cause inflammation on endometrial biopsy. Because antimicrobial activity is maintained to concentrations as low as 0.01%, this lavage at 0.05% may have a role in management of some bacterial uterine infections.[31]

Bacterial Resistance and Biofilms

Bacterial resistance to antimicrobial agents occurs in mares with chronic endometritis. This resistance may be conferred through the production of biofilms, which are communities of microorganisms attached to a surface that can express an increased resistance to antimicrobial agents.[50] There are many theories on how and why biofilms form.[50–53] Treatments also vary widely and include incorporation of enzymes and solvents in protocols in an attempt to break up the mucoid layer. An agent that has shown efficacy against biofilm production in human medicine and against chronic otitis externa, chronic dermatitis, and recurrent cystitis in dogs is EDTA-tris (ethylene-diaminetetraacetic acid-tromethamine).[54,55] In the mare, bacteria that may be associated with antibiotic resistant endometritis include *P. aeruginosa*, *E. coli, Klebsiella* species, *Enterobacter* species, and yeast. In the horse, EDTA-tris has been evaluated for its efficacy against clinical isolates of pseudomonas species. Pseudomonas isolates that were exposed to EDTA-tris solution exhibited decreased viability.[56] The treatment seems to be a safe adjunct therapy for equine genital infections when used as an infusion of 250 ml of 3.5 mM EDTA (0.05 M tris; pH 8) into the uterus; in fact, it induced an inflammatory response that was no greater than saline. We have added EDTA-tris (120 ml) to amikacin (1.5 g) for the treatment of chronic persistent endometritis cases that have been caused by *Pseudomonas spp.*, *Enterobacter spp., Klebsiella* species, and *E. coli*. Pregnancies have resulted in mares after treatment.

Mares with chronic endometritis have a thicker mucoid layer overlying the epithelium of the endometrium than reproductively normal mares, regardless of stage of cycle.[22] The mucoid layer may represent increased output by luminal goblet cells or may represent a biofilm, because both consist of cross-linked carbohydrates that take up specific carbohydrate stains. Solvents and mucolytic agents have been added to uterine irrigation fluids in an attempt to clear mucus or biofilm from the uterus of mares with persistent gram-negative bacterial or yeast infections. Agents used include DMSO, kerosene, and a 20% solution of acetylcysteine. Of the three compounds, only DMSO has been investigated in a clinical trial.[57] When a 30% concentration of DMSO was used as an intra-uterine infusion in 16 barren mares, there was a trend toward a higher pregnancy rate compared with mares irrigated with saline. In addition, 30% intra-uterine DMSO therapy resulted in a significant improvement in endometrial biopsy classification in 18 of 27 mares, whereas only 2 of 18 barren mares improved after intra-uterine saline treatment. Addition of these compounds should be used with care, because mares may have adverse reactions.

Fungal Infections

Treatment of fungal endometritis is frustrating because affected mares tend to be old and pluriparous with either poor perineal conformation or cervical incompetence. Oftentimes, they have been treated repeatedly with intra-uterine antibiotics. In addition, success rates are poor, and relapses are common. Mares with chronic fungal infections may be immunosuppressed or have an endocrine dysfunction such as equine pituitary disorders or insulin resistance. Prolonged progesterone therapy may also predispose mares to fungal endometritis; this is caused by a decrease in cervical drainage and an alteration in uterine muscular activity and neutrophil function.

Both yeasts and molds have been recovered from the uterus of mares.[58] Yeast appear as small, round, single-cell, brown to black spores on cytological smears obtained from infected mares, whereas molds have long, filamentous hyphae (Figs. 7 and 8). In chronic infections, hyphae may form large, tangled masses called mycelia. Mycelia may by visualized ultrasonographically within the uterine lumen as hyperechoic, irregularly shaped white structures surrounded by fluid. Mycelia recovered in vaginal or uterine fluids may have a “cotton ball” appearance or appear as yellow-tinged mucoid strains. *Candida* spp. and *Aspergillus* spp. are the most commonly isolated fungi from the equine uterus.[58] *Candida* spp. is a normal commensal of the gastrointestinal tract and vagina. Cytological smears obtained from mares with *Candida* endometritis usually contain spores, but hyphae may be seen in chronic infections. *Aspergillus* is a mold that produces hyphae. Other fungi isolated from the uterus include *Actinomyces* spp., *Fusarium* spp. (filamentous fungi), *Mucor* (filamentous fungi), *Pae-*

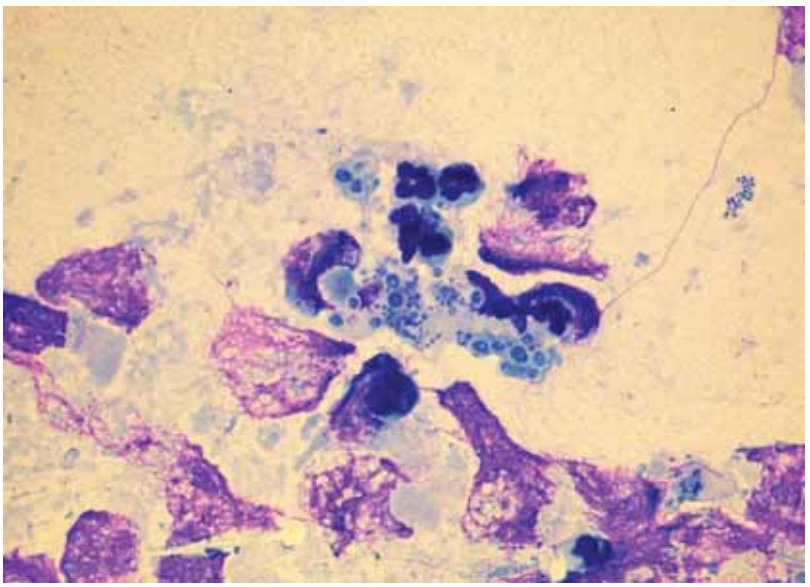

Fig 7. Yeast spores (small, light blue, circular structures of varying sizes), neutrophils (circular structures with dark purple nuclei and mucus on a cytological smear).

cilomyces spp., *Rhizopus* (filamentous fungi), *Rhodotorula* spp., and *Trichosporon* spp.

Yeasts tend to proliferate in fluid, whereas hyphae are better adapted for penetrating tissue.[58] These characteristics will affect treatment regimens and success. Yeast such as *Candida albicans* may be treated successfully with uterine irrigation and intra-uterine infusion of antifungal agents. Fungal infections caused by molds such as *Aspergillus*, *Mucor*, or *Rhizopus* may be deep seeded, requiring both systemic and local therapy.[59]

Development of fungal endometritis is a frequent consequence of repeated intra-uterine antibiotic treatments. Many of these cases resolve spontaneously if the mare has normal perineal anatomy and adequate uterine clearance. If the infection does not resolve in 1–2 estrous cycles, the uterus should be irrigated with a disinfectant for 5–7 days. Disinfectant solutions used for fungal infections include 3% (v/v) hydrogen-peroxide solution (30 ml hydrogen peroxide in 1 l of 0.9% saline), 2% (v/v) acetic acid (20 ml of white vinegar in 1 l of 0.9% saline), 0.1–0.2% (v/v) povidone-iodine solution, and 20% DMSO.[58–60]

Fig. 8. Hyphae on cytological smears appear as gray to black cylinders

If fungi are isolated after uterine irrigation with a disinfectant, a uterine culture should be submitted to a microbiology laboratory for an anti-mycotic sensitivity pattern. Recalcitrant fungal infections require prolonged therapy that is costly, and relapses are common. Both local and systemic treatments have been advocated, and in some cases, mares are treated by both routes. Drug dosages are not well defined in the horse, and minimum inhibitory concentrations for these drugs have not been established for equine uterine infections. Most of the data are extrapolated from humans, dogs, cats, or animal models. Breaches in anatomical barriers must be repaired. The most difficult fungal infections to resolve, in the author's opinion, are those in mares with a fibrotic cervix and inadequate uterine drainage and in mares with insulin resistance or Cushing's disease. In human medicine, a minimum of 10–14 days of treatment is recommended for fungal or yeast infections, whereas in the mare, intra-uterine treatment is usually limited to the duration of estrus or 5–7 days. To resolve an infection, mares may require two to three treatment sessions conducted during consecutive estrus periods. The interval between treatments can be shortened by administration of prostaglandin. The uterus should be recultured after the second treatment session to determine if the infection has cleared. If fungi are isolated, the reproductive tract should be reevaluated for anatomical defects, cervical incompetence, and/or testing for Cushing's disease or insulin resistance.

Table 4. Systemic and Topical Antifungal Agents Used for Fungal Endometritis

Drug	Dose	Route	Interval (h)	Spectrum
Systemic				
Amphotericin B	0.3–0.9 mg/kg	IV*	24–48	Broad spectrum
Ketoconazole	20 mg/kg (in 0.2 N HCl)	NGT†	12	Yeast‡
Fluconazole	Loading dose 14 mg/kg 5 mg/kg	PO, IV	24	Yeast
Itraconazole	5 mg/kg	PO,§ IV	12–24	Broad spectrum¶
Topical				
Clotrimazole	400–700 mg	IU	24 h × 7 days	Broad spectrum
Miconazole	500–700 mg	IU	24 h × 7 days	Broad spectrum
Nystatin	0.5–2.5 million units**	IU	24 h × 7 days	Yeast
Amphotericin B	100–200 mg	IU	24 h × 7 days	Broad spectrum
Fluconazole	100 mg	IU	24 h × 7 days	Yeast

*Diluted to 1 mg/ml in 5% dextrose and administered over 1–2 h.
†Nasogastric intubation is needed to avoid the irritant effect of HCl on the oral cavity and throat.
‡Yeasts: *Candida* spp.
§The bioavailability of the oral suspension is superior to that of the capsules.
¶Broad spectrum: yeasts, *Aspergillus,* dimorphic fungi
**Must be diluted in sterile water (100–200 ml) because it precipitates in saline.

Systemic therapy for a minimum of 21 days should be considered. Table 4 contains antifungal drugs and dosages for systemic and local infusion.

Inadequate Uterine Perfusion, Angeosis, Lymphatic Lacunae, and Cysts

Adequate blood flow to the uterus is needed for normal function. Hormonal distribution, signaling, uterine contraction, placentation, and early and late gestation are vascular-dependent events. Age-related changes and "inflammatory vascular alterations" in the walls of uterine arteries have been reported.[8,61] These include hyperplasia of smooth muscle cells in the tunica media and an increase in elastic fibers. The incidence as well as the severity of vascular lesions increased with the number of pregnancies and with aging.

Angiosis seems to indirectly reduce fertility through a reduction in endometrial perfusion and through disturbances in uterine drainage caused by reduced function of veins.[8] The most obvious clinical finding in mares with angeosis is the persistence of endometrial edema after ovulation. Endometrial lymphangectasia develops physiologically during estrus, which results in the typical estrous edema of the uterine wall. The edema disappears rapidly after ovulation, provided that the drainage mechanisms are functionally intact. If they are not, the result is a pathological endometrial edema, morphologically characterized by persistent lymphangectasia. The ventrally, dependent uterus in older multiparous mares seems to contribute to the problem.

Blood perfusion of the equine uterus has recently been studied by infusion of microspheres into the heart. Mares were grouped by degree of vascular degeneration and endometriosis on endometrial biopsy. Uterine blood perfusion during diestrus was greater in reproductively normal mares than in mares with vascular degeneration. Blood perfusion differed by 40% between the two groups, and maximal difference was in the distal uterine body.[62] Transrectal color and power Doppler ultrasonography have been used to study uterine blood flow and perfusion in mares with and without cysts. Findings showed a positive association between uterine cysts and disturbed uterine hemodyamics. A larger lymphatic cyst causes a greater perturbation of blood flow.[63]

Acupuncture has been used for many years in central Kentucky for mares that accumulate fluid in their uterus around breeding, for mares that reflux urine in their uterus, and for mares with pendulous uteri to improve their uterine tone. Clinical impression is that treatment decreases uterine size, improves uterine tone, provides a more rapid clearance of uterine fluid, and increases pregnancy rates. These findings may be caused by the effect of acupuncture on blood flow. It is used in human medicine for infertility and chronic pain.

7. Loss of Pregnancy by the Mare After the 14-Day and Before the 45-Day Examination

Pregnancy loss before 35 days may be associated with defective eggs, poor-quality embryos, or infection. The equine embryo is dependent on the yolk sac for nourishment until ~25 days of gestation. After that time, nourishment is attained primarily through endometrial secretions. If inflammation persists in the first weeks of gestation, it may result in death of the embryo. Mares carrying embryos that are small for gestational age or embryos with an irregular heartbeat at the 28-day pregnancy exam should be evaluated for uterine inflammation or infection.

Failure of the Endometrial Glands

Kenney[7] first suggested that advanced periglandular fibrosis is highly correlated with pregnancy loss be-

tween 35 and 80 days. The specific mechanism for interference of glandular secretion has not been defined, but many workers support the concept that periglandular fibrosis has a significant effect on early pregnancy survival. A presumptive diagnosis of fibrosis is made from a repeated history of early embryonic death between 35 and 80 days. Definitive diagnosis is made by histological examination of the endometrium. Endometrial samples will contain widespread moderate and/or severe periglandular fibrosis. There is no known treatment. However, many of these mares are placed on supplemental progesterone therapy, and there are anecdotal reports that the mare carried her fetus until term.

Progesterone Insufficiency

If corpora lutea do not produce progesterone or the ovaries are removed before 80 days of gestation, mares will abort.[64] The effect of rapid decline in progesterone levels on the tubular tract is loss of uterine and cervical tone and loss of the conceptus. Therefore, it has been hypothesized that partial failure of the corpus luteum (CL) could reduce the absolute amount of progesterone needed to maintain pregnancy. As a result, many schemes of administering exogenous progestins have been used to prevent early pregnancy loss. The value of these schemes is debatable because no critical evidence supports the concept. Nevertheless, we are often obligated to supplement this gonadal hormone because of client pressure and convincing histories. Progesterone supplementation in mares with prior histories of endometritis or lymphatic lacunae may be contraindicated, because progesterone may aggravate the problem.

Supplemental progestins does seem to be warranted in pregnant mares that are experiencing endotoxemia. Prostaglandins are released from the gastrointestinal tract during colic. The prostaglandins will induce lysis of the CL in mares that are <90 days pregnant, which results in abortion. After 90 days, the placenta takes over and begins to produce progestins that maintain pregnancy for the remainder of gestation. Progesterone therapy may also be beneficial in pregnant mares during the second and third trimesters if they have placentitis, colic, or severe systemic disease because it may inhibit premature uterine contractions.

Historical data are used to make the determination of progesterone supplementation. Reasons for placing mares <30 days pregnant on progestins include (1) history of early pregnancy loss, (2) low-plasma progesterone concentrations on days 5, 8, and 12 of gestation (the definition of low varies and ranges from 1 to 6 ng/ml), (3) visualization of uterine edema at pregnancy exams conducted between days 14 and 28, and (4) colic, laminitis, and vaginal discharge in late gestation. Treatment of mares experiencing endotoxemia in early pregnancy is administration of progestins (altrenogest dosed at 0.044 mg/kg, q 24 h, orally) to prevent pregnancy loss. Mares may also be given 150–200 mg of progesterone in oil daily or a long-acting progesterone[c] one time per week to maintain progesterone concentrations. Mares can be weaned off the progestin after 90 days of gestation as the fetoplacental unit begins to produce a number of progestins. If mares late in gestation are to be supplemented with progestins because of systemic illness or placentitis, the dose of altrenogest should be doubled (0.088 mg/kg, q 24 h, orally), because the recommended dose for suppression of estrus behavior did not prevent abortion in an experimental model.[65]

8. Conclusion

Our ability to diagnose the cause of a specific chronic infertility and manage it has improved greatly in the last 10–15 yr. Diagnostics, our understanding of reproductive physiology, new and old therapies, and the proper use of these treatments have resulted in increased pregnancy rates. However, the proper management of a chronically infertile mare is determined after a thorough reproductive examination is performed. If results are not as anticipated, one should always start again and reexamine the mare for anatomical defects or lack of synchrony between the ovary, uterus, and cervix during the estrous cycle. Many defects are subtle and only identified during certain phases of the estrous cycle. If no defects can be found, one may request to change stallions or obtain information on the stallion's recent fertility.

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