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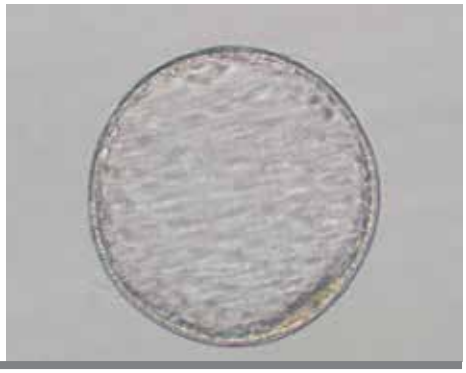

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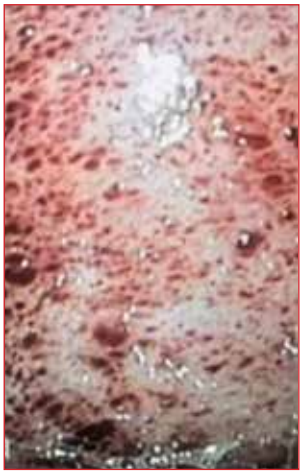

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Vitrifying LARGER EMBRYOS Without Puncturing

By Marie Rosenthal MS

On left, 460 micron embryo prior to vitrification. On right, the embryo after 4 minutes in equilibration solution.

Courtesy of Dr. Sandra Wilsher

Embryos that are between 300 µm and 480 µm may be vitrified without puncturing or aspirating them, and still achieve a fairly high pregnancy rate, according to **Sandra Wilsher**, PhD, who discussed the new technique at the British Equine Veterinary Association Annual Congress, recently held in Liverpool, England.

Embryos smaller than 300 µm tend to be easier to cryopreserve without additional manipulation, but the larger they become the more challenging it is to vitrify them, explained Dr. Wilsher, who practices reproductive medicine at Sharjah Equine Hospital in the United Arab Emirates.

"Equine embryos have always been a little bit challenging to vitrify, particularly if they're over 300

microns," Dr. Wilsher said. "This is chiefly due to the large blastocoele cavity filled with fluid, but also these embyros have a relatively small inner cell mass compared with their size."

Also once they've started to blastulate, they are developing an endoderm layer and a capsule, which increases the number of layers through which any cryoprotectants must pass, she explained.

As the embryo increases in size, it increases the risk for the developing ice crystals.

"However, we can get very good pregnancy rates when we puncture or aspirate this fluid. And what traditionally we've always thought is you must remove between 90% and 95% of this blastocoele fluid prior to vitrifying the embryo," she said.

The amount of fluid as the embryo increases in size is the primary concern because it increases the risk for the development of ice crystals, which can damage the embryo. The most common technique is to puncture the embryo, remove some of the blastocoele fluid and run it through a vitrification protocol.

Many reproduction specialists tend to use a micro manipulator to hold the embryo during aspiration, and they do get excellent results with embryos between 300 µm and 560 µm. However, the manipulator can be expensive. Manual puncture uses fine needles which are easily damaged.

"However, once you get skilled at using the micromanipulator, you can get really good results vitrifying embryos up to about 800 microns," she said.

Dr. Wilsher routinely uses the manual method for embryos less than 560 microns. In this case, one does not need to remove fluid, just make a hole, before vitrifying.

Both techniques require a steep learning curve, which can be challenging in a busy practice because not many clients "want you to practice kababbing their embryos," she joked.

"However, you can get really good results with this if you get good at it, but you can only really do it on embryos that are up to about 560 microns. But you don't need to aspirate the fluid, you just need to make the hole. And then we run through the vitrification protocols with them."

Once the embryo is manipulated, Dr. Wilsher uses an equilibration medium, which has ethylene glycol (EG) and dimethyl sulfoxide (DMSO) in it, and the embryos are exposed to around 5 to 6 minutes. And then they move them into the actual vitrification solution, which has higher amounts of both EG and DMSO, for about 60 to 90 seconds before they are loaded onto the minimal vitrification device and plunged into liquid nitrogen.

Skipping a Step

However, her group has come up with a method that appears successful in embryos up to about 480 µm, which enables them to skip the first step of puncturing and/or removing fluid.

They use commercial vitrification kits from Kitazato. The equilibration medium contains 7.5% DMSO and 7.5% EG; and the vitrification kit contains 15% DMSO and 15% EG.

"The only difference we have made in the protocol is we don't puncture them, and we leave them in the equilibration solution for 15 minutes," she said.

They drop the intact embryo into 2 separate 50 µL droplets for about 7.5 minutes each, although she said the total time, rather than the length in each droplet, is the most important aspect of the timing. Then, the embryo is moved into 2 separate droplets of the vitrification solution, which has the higher amounts of DMSO and EG for up to 90 seconds total time, which includes the time for loading onto the vitrification device, which in Dr. Wilsher's facility, is Cryolock. Then it is capped quickly and plunged into the liquid nitrogen. She caps it before she puts it into the nitrogen.

"We normally average about 1 minute 20 seconds for the time before it's dropped into the vitrification. But we have gone up to 1 minute 30. The thing to do is not to panic, which most people do. So, make sure it does stay in the vitrification solution for long enough. Don't, because you've only got a minute and a half start to panic and load it too quickly. So, it's only been in the solution for 1 minute or so. It's better, if it has been [in the droplet] a little bit longer," she advised.

Thawing Embryos

They use the same protocol as they do for the aspirated or punctured embryos when thawing them. They warm the 1M sucrose solution and the microscope plate to 42° C uncapping the Cryolock and plunging it straight into the thawing media, for 1 minute.

Then they move it through the dilution solution that is at room temperature, which has a slightly lower level of sucrose to make sure it does not expand

suddenly. Again, just as they did with the freezing protocol, they use 2 droplets and move the embryo from 1 to the other—leaving it in each droplet for about 2 minutes each, but a total of 4 minutes.

Then they move it to 2 droplets of the hold solution again for a total of 4 minutes at about 2 minutes a drop.

She admitted that most people do not warm the plate and solution up to 42° C. "The reason I started doing this is I read something about vitrification of some semen, and they were doing it at 42° C and got better results. We have compared doing it at 37° C versus 42° C, I haven't really seen any big differences. So, it may be just that there isn't really a benefit to doing it at this slightly higher temperature. I don't have enough data to be able to say yay or nay," Dr. Wilsher explained.

Most people do not warm the plate and solution to 42° C before thawing the embryos. Most thaw at 37° C.

Dr. Sandra Wilsher

So, far—and she admitted it was early days—they have seen an 82% pregnancy rate with this new protocol, which was done in 11 embryos that were smaller than 300 µm.

"These have never posed a problem to us anyway—vitrifying them before without puncture. They've always been very successful," she said.

When they used the protocol for larger embryos—300 µm to 500 µm—16 out of 20 resulted in pregnancies, so an 80% pregnancy rate without puncturing or aspirating the embryos.

However, the best results were among those that were no larger than 480 µm, she said. "The largest embryo that survived was 480 µm.

"If you are over 480 µm, basically you're going to have to puncture it because It's not possible to do it with this method at the moment," she said, although they are still tinkering with the protocol in the hope that they can improve the odds for larger embryos. **MeV**

Standing Fracture Repair Viable Option for P1, MC/MTIII Fractures

Standing fracture repair appears to be a worthwhile option for treating sagittal proximal phalanx (P1) and parasagittal metacarpal/metatarsal III (MC/MTIII) fractures because it can restore the horse's presurgical athletic abilities and enable it to return to the track within a reasonable time, according to a recent study.

Repairing P1 and MC/MTIII fractures used to be done under general anesthesia, but now is done under standing sedation. However, the success of the standing fracture repair has not been studied in a large cohort, so the researchers set out to determine the short- and long-term outcomes of horses undergoing these procedures.

They did a retrospective clinical record review of cases undergoing standing repair of P1 or MC/MTIII fractures from Jan. 1, 2007 to June 30, 2021, collecting data about signalment, fracture configuration and complications. They also reviewed the full race records of the horses and compared their pre- and post-surgical outcomes on the track.

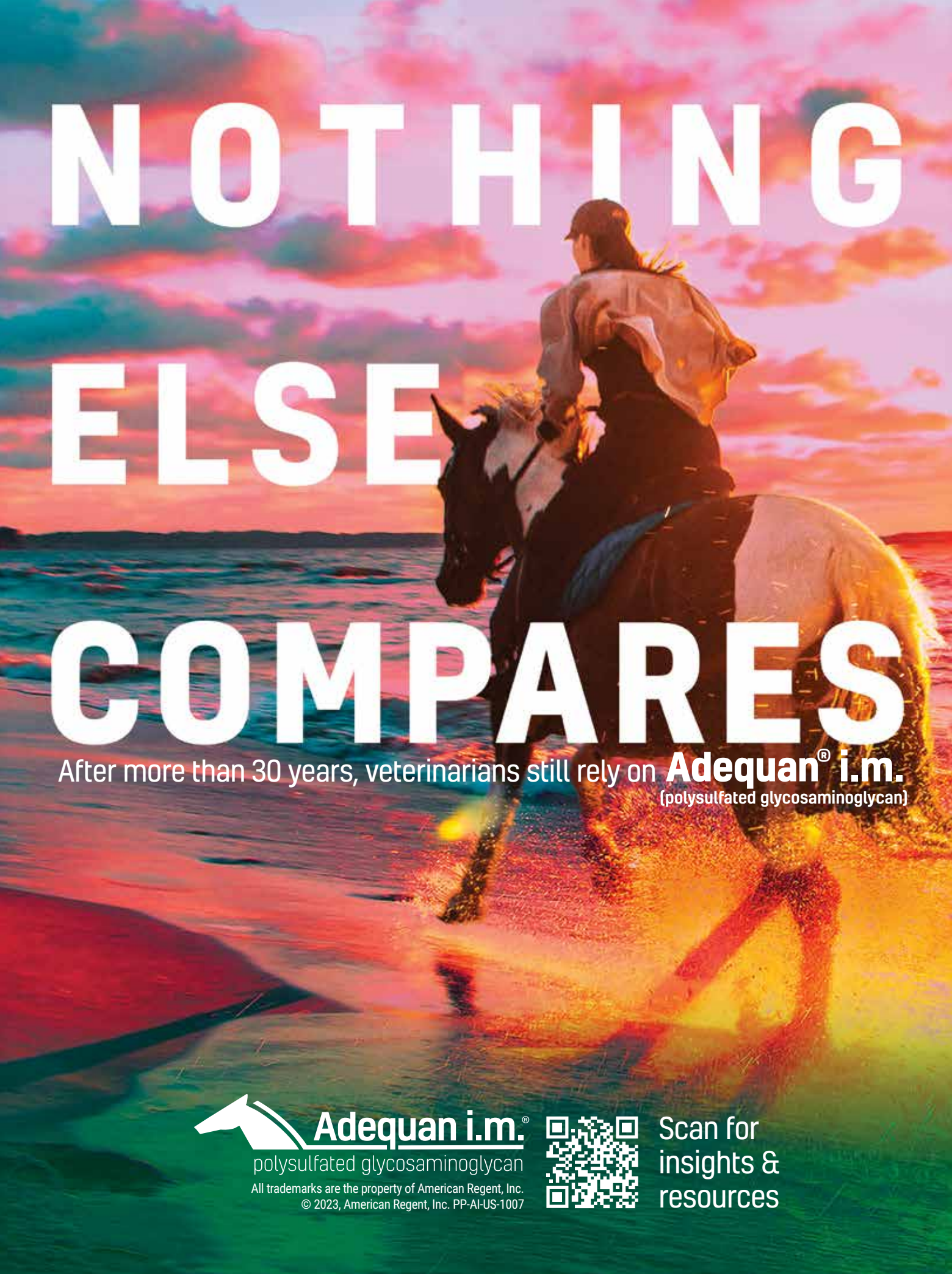
Of the 245 cases reviewed, 101 or 41.2% fractured the proximal phalanx and 144 (58.8%) had a condylar fracture. Of the repaired fractures, 64.9% (159/245) were bicortical and 35.1% (86/245) unicortical fissure fractures.

Almost all the horses—98%—survived to discharge, and just more than 75% raced a median of 241 days after surgery. The horses that were unable to race after surgery were more likely to experience complications (36.5% vs. 17.3%).

The researchers found no significant differences between the horse's pre- and post-operative racing performance and earnings per start (median £628.00, interquartile range 115.90–1934.80 vs. £653.20, 51.00–1886.40, P=0.7) or placing first–third (77% [95% CI: 68%–85%] vs. 71% [95% CI: 62%–80%, P=0.5] in at least one race. **MeV**

For more information:

Colgate VA, Robinson N, Barnett TP, et al. Outcome and racing performance following standing fracture repair in 245 horses. *Equine Vet J.* 2023 Oct. 6. **https://beva.onlinelibrary.wiley.com/doi/10.1111/evj.14016**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Ask the Financial Expert

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How can I help my clients be financially prepared for equine care?

BY MIKE POWNALL, DVM, MBA

How many times have you found yourself in the following scenario? A horse needs emergency care, but the owner balks at the cost. They ask if your practice can extend credit. While you discuss options, the horse is suffering and your frustration grows. Why don't they have insurance or an emergency fund? Why are they so unprepared? Don't they know horses are accidents waiting to happen?

As equine practitioners we deal with clients' financial challenges on a regular basis. And these challenges aren't limited to emergencies: horse owners are often just as unprepared to pay for routine care or chronic issues such as ongoing lameness. The solution is to offer clients financial care just as you provide patient care. Here are four strategies that can help you do this.

1 Build a healthy financial relationship from day one.

Be proactive about establishing a financial relationship with your clients early on. This means helping them figure out a way to pay before the crisis moment. Be open and transparent about the cost of equine care, and discuss the payment options you offer to help them be ready for the unexpected (and the expected) throughout their horse's lifetime.

2 Be aware of the financial realities horse owners face.

Depending on the type of horse they own (competitive, recreational or backyard), your clients may spend anywhere from nearly $300,000 to $900,000-plus taking care of their horse over its life.[1] What's more, owners tend to vastly underestimate these costs, spending three or four times more on their horses than they realize—no wonder 85% of them feel some kind of stress about horse-related expenditures.[1] Your understanding of these realities can lead to empathy and a foundation of trust between you and your clients.

3 Have a clear financial policy that sets the stage for lifelong care.

An ideal veterinary service agreement establishes your expectation that payment will be rendered at the time of service, lists the types of payment you accept, and highlights the value of your expertise in keeping horses healthy. In addition to sharing your policy at the first client visit, make sure to post it on your website and include it in brochures, treatment plans, invoices, client newsletters and so on.

4 Offer payment options that give clients a way to manage costs.

Insurance, wellness plans and financing solutions such as the CareCredit health and animal care credit card are all excellent ways to help clients manage lifelong costs. Everyone on your team—CSRs, technicians, practice managers, billing coordinators and veterinarians—should be comfortable talking about these options. Consider staff training focused on cost-of-care discussions (CareCredit offers a number of resources at carecredit.com/equineinsights), and note that financing is not just for emergencies—many horse owners use it for preventive care too. To make it easy for them to pay, provide convenient online and mobile payment options accessible through their phones or computers.

In my experience, the biggest benefit of these financial strategies is healthier horses getting the care they need. Whether you're dealing with an emergency colic or routine vaccination series, wouldn't you prefer to get right to providing care? Clients who are prepared with an established way to pay are much more likely to help you do just that.

1. CareCredit Equine Lifetime of Care Study, 2023, equinelifetimeofcare.com. CareCredit is a Synchrony solution.

To discover more about how veterinary financing can help you, your patients, your clients and your practice, visit CareCredit's Equine Insights page. Learn how you can provide the contactless digital financing clients want at carecredit.com/mycustomlink.

Dr. Mike Pownall, co-owner of McKee-Pownall Equine Services near Toronto, is a practicing veterinarian and veterinary business management consultant. He writes and speaks frequently on business topics for the equine veterinary profession.

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Why Are Some Mares More Reactive Than Others?

By Tom Rosenthal

Older Thoroughbred broodmares were more likely to have fearful responses to the stress of encountering unfamiliar objects than younger broodmares who recently retired from their careers, according to research presented at the BEVA Congress, held in Liverpool, England.

And while the foal is at foot, it appears to mimic the mothers' startled reactions to the novel objects, said **Alexandra Moss**, DVM, an internal medicine specialist currently working on her PhD in clinical genomics at the University College Dublin. Whether this similarity in response is maintained into adulthood is soon to be investigated.

Dr. Moss hypothesized older broodmares, who have had more opportunity for environmental exposures would have a lower startle response than younger mares, but found the opposite to be true, perhaps, because the younger mares were more used to unfamiliar situations due to their recent careers, while an older mare had spent more time away from these stressful situations.

"What was interesting for us was that we actually found with increasing age, we were getting a higher startle response time and a lower behavior score (indicative of increased stress-associated behaviors), which was actually counter to what we would expect when you think older horses are more likely to have experienced more in their lives," Dr. Moss said.

She posited if this was because the longer the broodmares have been out of racing, the less exposure they have to unknown objects so, were more startled when encountering novel objects.

The study is part of an overall effort to examine the concept of behavioral plasticity—the ability of animals to adapt to environmental stimuli with a reduced stress response, Dr. Moss explained. "Some animals cope better with stress than others. We don't necessarily know why. But we're interested in that, and our group in particular is interested in if there is an epigenomic contribution to that difference in stress response."

Dr. Moss said that much of the research conducted in this field has been done in the study of people.

"What we know is that early life stress leads to significant deleterious neurobiological effects, specifically on the hippocampus," she said. "And the hippocampus is crucial in life for regulating emotional responses, but also in forming learning pathways. And so that can lead to poor stress adaptation later in life."

Dr. Moss said that when it comes to horses, "obviously that's a welfare concern, but they're also performance animals, so that has significant economic consequences as well."

She added, "Our aims were to test the hypotheses that fearfulness could be established in Thoroughbred broodmares using both subjective and objective responses to a novel object. And we were particularly interested in age, parity and the time since they were last raced and whether these could be used as predictors for the level of fearfulness in these horses."

The study involved 25 Thoroughbred broodmares ranging from 5 to 16 years old that were outfitted with an ECG heart rate monitor system and let loose in a 5-by-5-meter box. Seventeen were multiparous, having had 2 or more foals, previously, and 8 were maiden or had only 1 foal previously.

Their responses to the introduction to a novel object—a self-opening bright red umbrella—were videotaped for 5 minutes. The ECG recorded the horses' responses during the 5 minutes and a 30-minute recovery period. And the tapes were reviewed by blinded observers. All the mares were introduced to the umbrella before it opened.

The behavior score and the startle response time, which averaged about 13 seconds, were proxies for stress response, and the positive linear relationship found with heart rate variability data obtained during the exposure validated their use, according to Dr. Moss. The older mares tended to be more reactive and took longer to settle down than the younger ones.

The mean stress response was 12.8 plus or minus 11.9 sec, mean peak heart rate was 110 plus or minus 34.3 beats per minute. Both the stress response and behavior were moderately correlated to age ($r=0.49$, $P=0.01$) and ($r= 0.53$, $P=0.007$).

"Ultimately what we can conclude from that is that subjective and objective behavioral responses in our cohort at least can be used to classify the Thoroughbred's response for fearfulness response to a novel object," she said.

Dr. Moss noted that the foals emulated their mothers' behaviors in responding to an unknown object but that by the time the yearlings had reached 18 months, anecdotally, they appeared to be moving away from that behavior. **MeV**

The study was funded by the Science Foundation, Ireland, the Frontiers for the Future Program, and a University College Dublin Veterinary School Summer Research Grant.

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Should you REFER that foal?

Look for the flags and trust your instincts

By Paul Basilio

A neonatal foal can take a turn for the worst quickly in the field, and knowing when to refer them to a fully outfitted clinic can make all the difference.

"You cannot monitor a foal too closely," said **Emily Floyd**, BVSc, DACVIM, MRCVS, at the 2023 BEVA Congress in Liverpool. "If you're not sure, examine them again, and then examine them again. But don't be afraid to ask for advice."

But how can a veterinarian be sure when the foal needs more treatment options than what's available on the truck? As with everything else in life, just make a note of the red flags.

Images courtesy of Dr. Emily Floyd

Sorting the flags

To begin the decision process, it's nice to rule out the low-hanging fruit. Conditions like moderate-to-severe sepsis/SIRS, severe colic or diarrhea, or orthopedic abnormalities that prevent the foal from standing are most likely going to require a facility with more resources.

However, foals with early or mild signs of those conditions could be successfully managed in the field, provided the signs don't become too serious.

"When we think about identifying the need for referral, I think it helps to be logical," said Dr. Floyd, clinical director at Rossdales Equine Hospital in Newmarket, UK. "Going through a list in a logical fashion will help you pick up the red flags when they're there."

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2. CAN I PROVIDE nutritional support?
3. CAN I TREAT or prevent sepsis?
4. CAN I TREAT the primary disease?

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5. CAN I PROVIDE adequate nursing care?

If the answer to any of these questions is "No," then a referral to a clinic is probably a good idea.

While it's nearly impossible to compile an exhaustive list of conditions that will require referral, Dr. Floyd ran through some examples where her 'Referral Alarm Bells' would immediately start to ring.

Sepsis is 1 of the leading causes of death in foals, and it's often a complicating factor in other conditions.

RED FLAG #1: Sepsis and/or organ dysfunction

Sepsis is 1 of the leading causes of death in foals, and it's often a complicating factor of other conditions, Dr. Floyd said.

"If there's a foal with mild neonatal maladjustment but the client says that the foal has been getting up and nursing, the client is likely not appreciating the fact that the foal could also be developing early sepsis," she explained. "This is a confounding factor, and you'll go from having a foal that is sort of doing ok to having a foal that is definitely not doing ok."

One of the big red flags to look out for with sepsis is an obvious sign of cardiovascular compromise, but often the first sign of sepsis is as simple as a foal that's just lying around. For example, if a foal can stand and nurse from the mare but flops into a heap when left unattended, that's a red flag.

"Don't dismiss these early signs," Dr. Floyd said. "If you're picking those up early, you're more likely to have success."

Entropion is another one of the early signs that a foal is not nursing well and may be becoming dehydrated.

One of the more profound signs of sepsis is inappropriate vasodilation, which can manifest in coronary band hyperemia. It's a sign of sepsis, but it can also indicate systemic inflammation. Some foals that experienced dystocia can have a hypoxic event that can trigger a marked vasodilatory process.

"If you see the coronary band hyperemia even before the foal collapses, that's a good sign that the foal has inappropriate vasodilation, and it could start to have cardiovascular dysfunction," she added.

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Animal Safety Warnings and Precautions:
Zycosan has been shown to prolong coagulation parameters up to 24 hours after injection, therefore caution should be used when administering this drug before or after strenuous activities (see Target Animal Safety). Due to the anticoagulant effects, this drug may exacerbate Exercise Induced Pulmonary Hemorrhage (EIPH).
The concurrent use of NSAIDs with Zycosan has not been evaluated. Due to the anticoagulant effects of Zycosan and known anticoagulant effects of some NSAIDs, caution should be used if NSAIDs are concurrently administered. Horses concurrently treated with Zycosan and NSAIDs should be monitored for hemorrhage or other clinical signs of abnormal bleeding (e.g., petechiae, ecchymosis, or epistaxis). The safety of long-term repeat use of Zycosan has not been evaluated. Pigmentary changes in the retina (pigmentary maculopathy) have been reported in human patients following long-term oral use of pentosan polysulfate sodium. It is not known if a similar finding occurs in horses. The safe use of Zycosan has not been evaluated in breeding, pregnant, or lactating horses.
Other Warnings:
Do not use in horses intended for human consumption.
ADVERSE REACTIONS:
Injection site reactions were the most frequently reported adverse reactions in the field study. Injection site reactions were associated with clinicopathology changes in some cases. Other adverse reactions reported in more than one horse were prolongation of coagulation parameters (activated partial thromboplastin time (aPTT) and prothrombin time (PT)), lethargy, behavior changes, and colic.
To report suspected adverse events, for technical assistance or to obtain a copy of the Safety Data Sheet (SDS), contact Dechra at (866) 933-2472 . For additional information about adverse drug experience reporting for animal drugs, contact FDA at 1-888-FDA-VETS or online at http://www.fda.gov/reportanimalae.
STORAGE CONDITIONS: Store at room temperature 68-77ºF (20-25ºC), with excursions to 59-86ºF (15-30ºC).

MANUFACTURED FOR:
Dechra Veterinary Products
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RED FLAG #2: Neurologic signs

An early sign of neurologic dysfunction is increased muscle tension, including rigidity of the neck—something Dr. Floyd calls "the motorbike."

"It's when you go to get them up and they lean on you," she said. "You can't step away from them because they'll fall."

Sometimes, a foal can equilibrate once you get them standing for a few minutes. But even still, these early neurologic signs—particularly in a foal that experienced dystocia—could indicate that the foal's health could easily plummet if no intervention is provided.

If these neurologic signs persist, seizures could result. Seizing foals can be difficult to treat on the farm.

RED FLAG #3: GI signs

"Generally, I think we tend to be quite complacent about what GI issues we can treat," Dr. Floyd said. "We look at a foal and think, 'Oh they've just got meconium impaction or a little diarrhea. I can manage this at home.'"

However, she added, it's important to think of clues that may indicate that the GI signs could be outside of the normal spectrum. The presence of abdominal distension, for example, could indicate that the foal is beyond a simple meconium impaction.

"If you see that, don't just keep persevering," Dr. Floyd said. "Either do some more diagnostics or think about other help that you can provide for the foal. [Veterinarians] can get tricked into thinking that the foal is just a little dysmature or has a mild maladjustment in addition to the meconium impaction, but the foal also may have something like a small bladder tear. If you're not being critical about re-assessing the foal, there may be all sorts of things that you can miss that indicate it's not a simple issue."

RED FLAG #4: Recumbency

If a foal is persistently recumbent, field management may not be the best option. Recumbency can be a sign of sepsis, or the foal could have an orthopedic issue such as a muscle rupture or congenital defect.

"If you conduct a thorough history and physical exam and you think you can provide treatment yourself, then you want to be critical of what's happening during treatment," Dr. Floyd added. "If the foal [doesn't respond], then your alarm bells should be ringing quite loudly. Be critical. Don't just keep going. Think about referral or think about whether there's other diagnostics you can perform."

RED FLAG #5: Lab abnormalities

Clinical assessment of foals is extremely important, but some laboratory abnormalities can indicate whether this foal should be treated at a facility.

Caring for a sick foal.

- Hemoconcentration or anemia
- Leukopenia/leukocytosis or increased inflammatory markers
- Evidence of organ dysfunction or multisystemic disease
- Electrolyte abnormalities

"Some basic bloodwork can be an accentuation of your physical examination," Dr. Floyd said. "If you have a foal that looks hemolytic or jaundiced, then sometimes you can underestimate that. But then you get the lab work back and it turns out that the foal has a PCV of 12%, and then you've possibly got more on your hands than you expected."

For measuring lactate, Dr. Floyd doesn't necessarily have a hard cut-off for what constitutes a referral red flag.

"I wouldn't say that a lactate of 3 is fine and a lactate of 4 needs to go to the hospital," she explained. "But if you're thinking about whether you can manage the foal [in the field] and you have a POC lactate monitor that is showing a 4 or 5, then that's just another indication of severe disease."

One of the biggest laboratory red flags is an elevated creatinine level with a high urea concentration. Many of these foals will have several signs of perfusion abnormalities, and they will be predisposed to renal problems or acute kidney issues.

As with all red flags, Dr. Floyd has simple advice.

"Trust your instincts," she said, "If it doesn't seem right, then it probably isn't. If the foal isn't doing quite what you expect, then don't dismiss that. You're probably right. Just don't be afraid to ask for advice." **MeV**

Case Study: Dribbling Urine in a Warmblood Mare

By Megan Born, MS, LVT, and Nimet Browne, DVM, MPH, DACVIM

An adult Warm-blood mare weighing approximately 600 kg presented for a history of partial urinary incontinence. The mare had been dribbling urine for a few days prior to admission. The mare is used as a riding horse and was open when seen at the hospital. An ultrasound was performed in the field prior to referral, which demonstrated a plethoric bladder and hyperechoic sediment.

She was started on trimethoprim-sulfadiazine (TMP-SMZ) of unknown dosage prior to admission. Laboratory analysis done 24 hours before admission of a complete blood count, fibrinogen, and chemistry had reportedly normal values. A urinalysis done in the facility's laboratory found hazy urine with trace leukocytes and 3+ calcium carbonate crystals.

The mare was bright, alert and responsive on admission. On physical examination her heart rate was 40 beats per minute (BPM), her respiratory rate was 12 breaths per minute (BrPM), and her temperature was 99.6° F. The capillary refill time was 2 seconds, and her mucous membranes were pink and moist.

Her digital pulses were normal in all 4 limbs with the rear limbs having stronger pulses than the front. The hindlimbs had significant swelling below the tarsus. The semimembranosus area had some skin scalding from the dripping urine.

A neurologic exam was performed, and her mentation was normal, and cranial nerve responses were intact. No ataxia was noted when walking in a straight line and while circling. On clinical examination of this mare, she had normal systemic and neurologic parameters. The urinary tract needed more diagnostics to help determine the cause of the urinary incontinence.

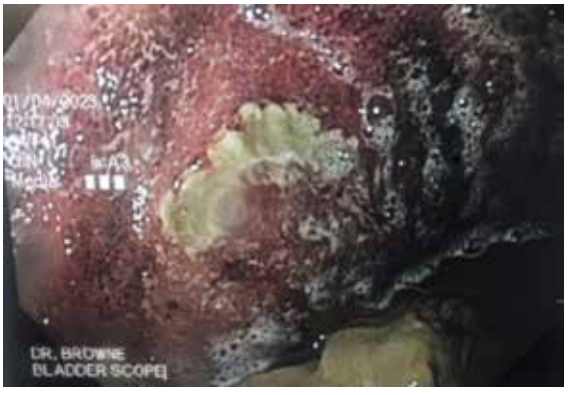
A urinary catheter was placed vaginally into the urethra. Urine was siphoned and was straw colored. Approximately 16 L of urine was retrieved. Toward the end of the fluid retrieval the urine became thick with sediment. A cystoscopy was performed to visually examine the bladder. No cystoliths were observed. A lot of sediment was present in the ventral bladder. The bladder wall had crystals adhering to it, and portions of the bladder urothelium were ulcerated.

The mare was bright, alert and responsive on admission. A neurologic exam found her mentation was normal.

The veterinarians made an initial diagnosis of severe hemorrhagic cystitis. It was observed that this cystitis had a sabulous component, which means that the accumulation of sediment was likely due to incomplete emptying of the bladder due to a possible nervous system component to the cystitis. The reason for the bladder atony could be neurologic or idiopathic. The clinician gave a

Images courtesy of Megan Born

Ulcerated and hemorrhagic bladder urothelium



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guarded prognosis to the client. The client asked about potentially using this mare as a broodmare, and the veterinarian was unable to give an affirmative until the cystitis resolved.

Differentials

The mare was tested for equine protozoal myeloencephalitis (EPM) prior to arrival to assess whether EPM was the cause of the bladder nerve function component. The results were not available on admission. Other causes of urinary incontinence include equine herpesvirus 1, sacral/coccygeal trauma, congenital causes, neoplasia, idiopathic bladder paralysis, equine motor neuron disease and polyneuritis equi, according to Mair, et al.

The geographical region where the mare lives has a high prevalence of EMP infection making this disease the likely cause of her condition.

This mare did not exhibit a fever or other systemic indicators of herpesvirus. No evidence on clinical examination or prior history of predisposition to any of these other causes of bladder atony was found. Ectopic ureters can also be the cause of urinary incontinence, but the cystoscopy showed normal functioning ureters. Mares after foaling can have some nerve damage that could cause bladder atony, but the mare was a maiden.

The geographical location of the region the mare lives has a high prevalence of EPM infection making that disease the most likely cause of the sabulous cystitis if not of idiopathic etiology.

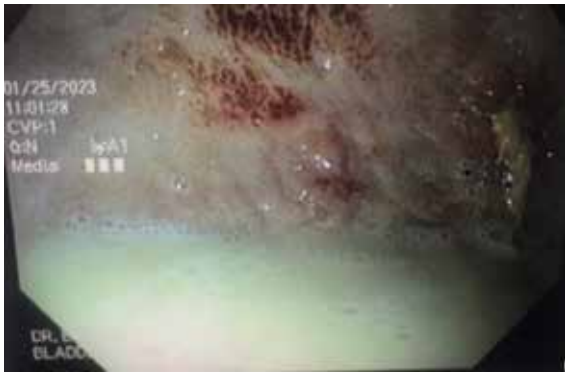
An EPM serum titer of *Sarcocystitis neurona* (causative agent of EPM) types 2, 3 and 5 was performed, and the titer level was 1:500. This titer level only confirms exposure. A cerebrospinal fluid (CSF) tap would have been the gold standard for the definitive rule out of this disease as the cause of the sabulous cystitis. A CSF tap also cannot completely determine pathogenicity of EPM in the individual horse as a positive serum CSF tap is not always indicative of the organism causing systemic disruptions. A CSF tap that is negative for the organism, however, is always an indication that *S. neurona* is not implicated in the clinical signs. The owners of this mare were financially constrained, so a CSF tap was not feasible for them at this time.

Sabulous cystitis can be a secondary sequela to the presence of bacteria, as well as can be predisposed to bacterial proliferation due to the static urine retention. A urine culture was done after admittance to determine presence and classification of bacteria. Two isolates were found, the first was a light alpha *Streptococcus* species and the other was a scant *Pseudomonas aeruginosa*. A sensitivity was done as well to determine if the TMP-SMZ regimen the mare presented with was adequate for continued therapy. The sensitivity showed that *Streptococcus* had intermediate sensitivity, but the *Pseudomonas* was resistant to TMP-SMZ. The owners had some financial constraints that prevented a change in antibiotic therapy. However, fortunately *Pseudomonas* can be a contaminant, and TMP-SMZ tends to accumulate in the urinary tract potentially making both bacteria susceptible to higher doses.

The mare was managed during the hospitalization period with the following treatments and procedures:

- Complete physical examination with vital parameters every 6 hours;

Large amount of sediment on bladder floor



Cystoscope 3 Weeks after Initial Admission Showing Improvement in Hemorrhagic Cystitis and Sediment Accumulation

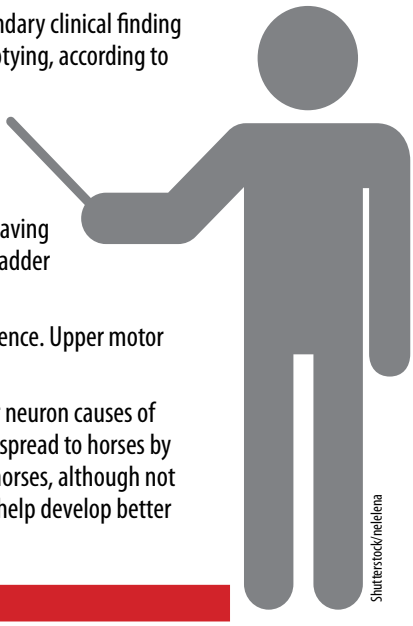
Teaching Points

Sabulous cystitis is the accumulation of sediment in the bladder of the horse. It is generally a secondary clinical finding due to paralysis of the bladder or other neurologic problem that results in incomplete bladder emptying, according to Schott. The condition can be first recognized when the horse develops urinary incontinence.

Urinary incontinence can be caused by both upper and lower motor neuron deficits. In upper motor neuron disruption, the nerves that are affected are cranial to the sacral area. Clinical signs such as intermittent urine dribbling and accumulation of sabulous material in the bladder may be seen as the horse does not void urine normally. Lower motor neuron deficits are characterized by having other pelvic area symptoms such as reduced anal tone, tail paralysis, and hindlimbs deficits. The bladder continually overflows and dribbles urine. (Mair, 2022).

This mare showed clinical signs of possibly an upper motor neuron etiology of the urinary incontinence. Upper motor neuron causes of incontinence are rare in the horse but include equine herpesvirus 1 and EPM.

As stated previously EPM can be a cause of neuron disease and can be both upper and lower motor neuron causes of incontinence. EPM is caused by *Sarcocystitis neurona*, a coccidian organism that is most commonly spread to horses by the ingestion of opossum feces. Once infected, lesions can develop in the brain and spinal cord of horses, although not every horse that is exposed to EPM develops symptoms of disease. More research is being done to help develop better testing procedures as definitive diagnosis can be difficult in an area where EPM is endemic.



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- Assessment of urine output (non-quantifiable) every 2 hours;
- Assessment of manure output every 4 hours;
- Monitoring functionality and placement of urinary catheter every 2 hours;
- Flush urinary catheter every 4 hours to prevent blockage;
- Analgesics (detailed in next section); and
- Repeat cystoscopies to assess progress and response to therapy.

On the day of admission, the mare became uncomfortable later in the evening, showing signs of colic. It was not determined whether she was having bladder pain or generalized abdominal pain. A nasogastric tube was passed and electrolytes given orally. She was also muzzled overnight in case the pain signs were due to a gastrointestinal problem. She had low manure production until the morning. Flunixin meglumine was given every 12 hours. The mare was also walked to help with GI motility.

Fluids were given the first day due to possible GI issues at a rate of 10 L every 12 hours. The bladder was lavaged with 1 L of saline and 50 mLs of 99% DMSO every 12 hours. The mare's heart rate was in the 40s (bpm) on the first day of admission. Flushes were added to the chart every 4 hours after sediment was found to be building up in the urinary catheter.

Sabulous cystitis can be a secondary sequela to the presence of bacteria, as well as a predisposition to bacterial proliferation.

On the day after admission no fluids were given. The mare was given free choice hay and made comfortable. Her heart rate had dropped to the low 30s (bpm). A repeat cystoscopy was performed, and the inflammation and sediment build up weres improving.

On the third day after admission bethanecol was started to assist with the emptying of the bladder. TMP-SMZ were started every 12 hours to deter bacterial growth. The IV catheter was removed, and the mare was changed to receiving the same dose of flunixin meglumine the injectable given orally to reduce owner cost. Mare remained comfortable with normal appetite, manure and vital signs.

On the fourth day after admis-

The veterinarian reiterated to the client that although there were marked improvements in the mare's condition, the cystitis was ongoing.

sion the bladder lavages were changed to 500 mLs of tricide flush (Bottle contained 8mM USP Disodium, 0.01 mM Disodium/kg).

On day 5, the mare continued to improve with stable parameters and behavior. Flunixin meglumine was reduced to every 24 hours. Another cystoscopy was performed and showed a reduction in inflammation by 75%.

On day 6, the urinary catheter was removed, and the mare was monitored for urination. Bladder ultrasounds were also performed to assess normal micturition.

The mare was discharged to the farm on the seventh day after admission.

On discharge from the hospital the mare had improved considerably. The most recent endoscopes showed less sediment and decreased inflammation by 75%. The mare was able to posture and urinate on her own without assistance from a urinary catheter. The mare had also stopped dribbling urine.

The veterinarian reiterated to the client that although there were marked improvements, the cystitis is still ongoing. Therefore, systemic treatment was still needed once discharged to the farm to give the mare the best possible outcome. The veterinarian also reminded the clients of the fact that a cerebrospinal fluid tap is the only way to definitely rule out EPM as the cause of the sabulous cystitis.

The goals of therapy in this case were to assist with micturition until whatever inflammatory or nervous process resolved enough for the mare to be able to fully void her bladder. The cystoscopies were helpful as an objective assessment of improvement. The mare did better than expected with therapy especially due to the financial concerns of the owners. **MeV**

About the Author

Megan Born, MS., LVT is a licensed veterinary technician who has worked in the Internal Medicine and Critical Care Unit at Hagyard Equine Medical Institute since 2012. She is also a part time instructor for veterinary technicians at the University of Missouri Online. She enjoys showing reining horses and attends Clays Mill Baptist Church.

Dr. Nimet Browne studied veterinary medicine at University of Tennessee, graduating in 2010. Following veterinary school, Nimet went on to complete an internship in a private practice in Illinois, then a large animal internal medicine residency at Virginia Tech. During that time, she also obtained a Masters In Public Health with a focus on infectious disease. Following her residency, she completed a yearlong fellowship at Hagyard Equine Medical Institute. In 2016, she took a faculty position in the equine internal medicine department at North Carolina State University. Her interests include neonatal medicine, gastrointestinal disease, infectious disease and public health. She is currently working on manuscripts regarding infectious causes of diarrhea in neonates as well as the use on enrofloxacin in neonates

For more information:

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Assessing Severity in Horses With Trigeminal-Mediated Headshaking

The History, Rest and Exercise Score (HRE-S) is a reliable method for evaluating disease severity in horses with trigeminal-mediated headshaking.

The HRE-S score is a combination of 3 subscores: the history score, resting score and exercise score.

Clinical signs of trigeminal-mediated headshaking include neuropathic facial pain, but the assessment tends to be subjective, and an incorrect assessment could mean the horse is suffering severe pain, according to the researchers, who wanted to develop and validate a precise scoring system for trigeminal-mediated headshaking.

Seven blinded observers with different experience used HRE-S to score 40 video recordings taken during rest and lunging in 9 horses with trigeminal-mediated headshaking and 3 without, who acted as controls. The videos included 5 duplicates. For every video recording, the observer graded the severity of clinical signs using an intuitive global-type-scale and interobserver reliability was calculated.

Reliability for HRE-S was excellent, irrespective of observers' experience, according to the researchers. Horses with grade 3/3 had significantly higher average E-S and total scores compared with an existing score than those with grade 0/3 or 1/3 ($P < 0.001$).

Interobserver reliability for intuitive global-type-scale was fair to substantial with wide variability. **MeV**

For more information:

Kloock T, Pickles KJ, Roberts V, et al. History, rest and exercise score (HRE-S) for assessment of disease severity in horses with trigeminal-mediated headshaking. *Equine Vet J*. 2023 Aug. 22. Published online.
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