27th Equine Health and Nutrition Conference

Applying the Science

February 5–6, 2024
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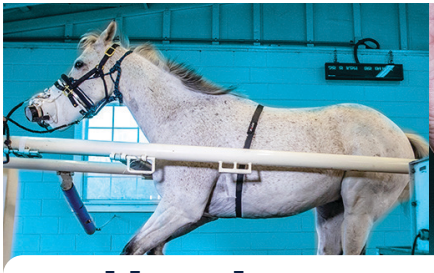

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Introduction

Welcome to the 27th Equine Health and Nutrition Conference, presented by Kentucky Equine Research. We are so glad you are here!

In keeping with the history of these conferences, we have put together an incredible roster of relevant topics and world-class speakers for you to enjoy over the next two days. The equine health and nutrition landscape has changed incredibly since Kentucky Equine Research was founded in 1988, and we have kept abreast of important changes in part through gatherings like this one.

Contained within these proceedings is a compilation of easy-to-read, informative Q&As. We posed questions to our speakers about their areas of interest, and they provided candid responses. We appreciate the willingness of the speakers to participate in this conference and in the creation of these proceedings.

About the Speakers

Warwick Bayly, BVSc, MS, PhD, DACVIM

Dr. Warwick Bayly is a professor of equine medicine in the Department of Veterinary Clinical Sciences at Washington State University. He graduated from the veterinary faculty at the University of Melbourne and went on to get his master's degree at Ohio State University and doctorate from the University of Liège in Belgium. His principal academic interests are equine exercise science and exercise-associated diseases, with emphasis on the respiratory system. His current focus is on the pathogenesis and management of exercise-induced pulmonary hemorrhage. He has written or cowritten over 150 scientific papers and textbook chapters related to equine exercise science and medicine and has co-edited all four editions of the well-known textbook, *Equine Internal Medicine*. He also served as the dean of the College of Veterinary Medicine at Washington State University, past president of the World Equine Veterinary Association, and a former director of the American Association of Equine Practitioners.

Robin Bell, BVSc, MVSc, DipVetClinStud, DECVS, DACVSMR

Dr. Robin Bell earned his veterinary degree from the University of Sydney and then completed an internship at the University Veterinary Teaching Hospital Camden. Following his residency training in equine surgery and master's degree at Massey University in New Zealand, he worked in a large referral lameness clinic in the United Kingdom. He then oversaw the lameness and imaging service at the University of California, Davis, before returning to the University of Sydney. He is an official international veterinarian with interests in jumping, dressage, and eventing. He is currently the veterinarian for the Australian jumping team. His research interests include the treatment of tendon injuries with stem cells, clinical applications of MRI and CT in equine practice, and the early diagnosis of musculoskeletal injuries in equine athletes.

Samantha Brooks, PhD

Dr. Samantha Brooks is an associate professor of equine physiology at the University of Florida. After earning a bachelor's degree in agricultural biotechnology from the University of Kentucky, she remained there to study at the Gluck Equine Research Center, where she earned her doctorate in veterinary science, specializing in equine genetics. She was awarded the Paul Mellon Postdoctoral Fellowship to study the expression of inflammatory genes in horses affected with laminitis. She taught at Cornell University before moving to the University of Florida. Her research program explores a variety of topics relevant to horse health ranging from gene expression studies to mapping of genetic disorders in the horse. Her research group discovered genetic mutations and markers for coat colors, height, sarcoid tumors, and two neurological conditions. Ongoing work targets variation in gait, susceptibility to infectious disease, metabolic syndrome, and skeletal defects using genome-wide association, genome resequencing, and transcriptomics.

Hilary Clayton, BVMS, PhD, DACVSMR, FRCVS

Dr. Hilary Clayton is a veterinarian and scientist who, for more than 40 years, has researched the areas of locomotor biomechanics, lameness, rehabilitation, and conditioning programs for equine athletes, and the interaction between rider, tack, and horse. She served as the Mary Anne McPhail Dressage Chair in Equine Sports Medicine at Michigan State University's College of Veterinary Medicine from 1997 until she retired from academia in 2014. She has published seven books and more than 200 scientific articles on these topics. She continues to perform collaborative research with colleagues in universities around the world. Clayton is a charter diplomate and past president of the American College of Veterinary Sports Medicine and Rehabilitation. She is an Honorary Fellow of the International Society for Equitation Science and has been inducted into the International Equine Veterinarian Hall of Fame. She is a lifelong rider and has competed in many equestrian sports, most recently focusing on dressage.

Mike Davis, DVM, PhD, DECEIM

Dr. Mike Davis is a professor, the Oxley Endowed Chair in Equine Sports Medicine, and the director of the Comparative Exercise Physiology Laboratory at the Oklahoma State University College of Veterinary Medicine. He graduated with a veterinary degree from Texas A&M University and then went on to receive a master's degree in veterinary science from Virginia Tech and a doctorate in physiology from Johns Hopkins University. He has received millions of dollars in research funding to study the effects of exercise stress in animal models, particularly racing sled dogs. This work has resulted in detailed metabolic studies of the effects of stress. He is the senior author of over 30 publications related to stress physiology in companion animals.

Sally DeNotta, DVM, PhD, DACVIM

Dr. Sally DeNotta is a clinical assistant professor at the College of Veterinary Medicine at the University of Florida. She earned a veterinary degree in 2008 from Oregon State University and her doctorate of philosophy in 2018 from Cornell University. She teaches several courses at the university, and has authored numerous papers in peer-reviewed journals. Her clinical and research interests include neonatology, neurology, infectious disease, hemostatic therapies, and colic.

Joe Pagan, MS, PhD

Dr. Joe Pagan is the founder and president of Kentucky Equine Research, an international equine nutrition and exercise physiology company. He earned his master's and doctorate degrees from Cornell University in equine nutrition and exercise physiology. Not long after graduation, he formed Kentucky Equine Research and has been conducting innovative research for 35 years at the flagship facility in Versailles, Kentucky, and at the Kentucky Equine Research Performance Center in Ocala, Florida.

Kentucky Equine Research served as equine nutrition consultants for the last six Olympic Games and several World Equestrian Games. He received the American Feed Industry Association (AFIA) Award in Equine Nutrition Research in 2005. This award recognizes excellence in equine nutrition research and the contributions of an individual to equine feeding management practices and the equine feed industry.

Wendy Pearson, MS, PhD

Dr. Wendy Pearson is an associate professor of equine physiology at the University of Guelph. She earned her master's in nutritional toxicology and her doctorate in biomedical toxicology from the University of Guelph. She studies nutraceuticals and nondrug veterinary pharmaceuticals. Her research focuses on clinical nutrition and dietary modifications to treat inflammatory conditions in horses, such as arthritis. She aims to develop nutritional and dietary approaches to improve horse health and has worked on clinical trials with arthritic horses. With more than 60 research papers to her credit, she is a prolific writer. Part of her work at the University of Guelph involves mentoring graduate students.

Ben Sykes, BSc, BVMS, MS, DAVCIM, DECEIM, MBA, PhD

Dr. Ben Sykes is an associate professor in equine internal medicine at Massey University and a professor of curriculum development at Southern Cross University. He graduated with a veterinary degree from Murdoch University in 1997, completed an internship at Randwick Equine Centre in 1998, and held a residency in equine internal medicine in Virginia. He earned a master's in business administration from the University of Liverpool, and a doctorate in veterinary pharmacology from the University of Queensland. For his doctorate, he investigated the pharmacokinetics and pharmacodynamics of omeprazole in horses. Throughout his career, he has worked with a wide range of horses, focusing on high-performance horses. He maintains a strong interest in clinical research as well as product development and commercialization. His primary research focus is gastrointestinal disease with a specific interest in equine gastric ulcer syndrome.

Stephanie Valberg, DVM, PhD, DACVIM, DACVSMR

Dr. Stephanie Valberg is a pioneer in understanding and managing equine neuromuscular disorders in horses. Her research has transformed equine clinical practice and has led to the discovery of previously unknown muscle disorders, the identification of their genetic basis, and the development of nutritional strategies to minimize muscle pain. She worked with Kentucky Equine Research to develop the first feed used to reduce the incidence of tying-up and was a member of the team that sequenced the equine genome. She was named the Mary Anne McPhail Dressage Chair in Equine Sports Medicine at Michigan State University's College of Veterinary Medicine in 2015 and remained in that position until she retired in 2022. She mentored more than 60 graduate students, interns, residents, and postdoctoral students. She is widely published and is a recipient of numerous awards for teaching and mentorship. In 2012, she became the first woman to be inducted into the University of Kentucky Equine Research Hall of Fame and twice received the Pfizer Research Excellence Award.

Emmanuelle Van Erck, DVM, PhD, DECEIM

Dr. Emmanuelle Van Erck practices equine sports medicine in France and Belgium. She graduated from the Ecole Nationale Vétérinaire d'Alfort and later received her doctorate on respiratory functional tests in horses at the Veterinary Faculty of the University of Liege (Belgium). Her work led her to participate to research projects in equine sports medicine at Cornell University and at Uppsala University. These experiences triggered her passion for equine sports medicine. She then became a senior consultant at the Centre for Equine Sports Medicine at the University of Liège, where she dealt with referred cases in sports medicine, participated in numerous research projects, and collaborated with the training of veterinary students. She is the author of more than 40 scientific articles and regularly lectures at international scientific meetings. She was appointed team veterinarian for Belgium and has served as president of the Belgian Equine Practitioner Society.

Exercise Physiology Research: Past, Present, and Future

Warwick Bayly, BVSc, MS, PhD, DACVIM
Mike Davis, DVM, PhD, DACVIM

First, provide an of overview. Can you define exercise physiology? What is it about this field of study that interests and inspires you?

Davis: Physiology is defined classically as the study of the function of living organisms. I tend to tell my students that physiology deals with movement. Obviously, there is no shortage of moving things to study in the context of exercise, but it is important to point out the range of scales: in the context of exercise physiology, someone can study the movement of electrons through the skeletal muscle mitochondria or the movement of horses racing down a track.

I am drawn to the study of exercise physiology because exercise defines the limitations of different physiological processes. In many instances, success at exercise is a function of an athlete's limitations (or lack thereof, relative to the competition). A physiologist can impact an athlete's exercise performance by understanding not only the limitations but how to address them either through managing performance or conditioning to change the limitations. And seeing that impact is satisfying—you know you have made a difference.

Bayly: Fundamentally, exercise physiology deals with the conversion of chemical energy into mechanical energy. By definition, the latter therefore involves movement, as mentioned above by Mike. In terms of actual exercise, this movement is equated with the sequential contraction and relaxation of muscle. This is a "basic" aspect to the scientific discipline of exercise physiology that relates to studying the intricacies of how chemical energy is converted to mechanical energy and is rooted in biochemistry and respiratory and cardiovascular physiology.

"Applied" physiology pertains to: (1) the application of basic exercise physiological principles to subjects like conditioning and training and the assessment, prediction, and improvement of competitive performance; and (2) understanding the causes of, prevention, and treatment of exercise-associated maladies/injuries. More recently this area of applied physiology has been bundled under the designation of "sports medicine."

As well as being drawn to the spectacle of elite athletic performance, I have always been fascinated by the complexities of the physiological and biochemical bases of exercise; i.e., the "how" part. This led to applying this basic information to the "why" of certain exercise-associated medical problems, the thinking being that if you understand the "why," perhaps you can avoid or mitigate them, and/or treat them more effectively.

Is the study of exercise physiology intended to improve performance or better understand poor performance?

Davis: Improving performance versus understanding poor performance is a good example of a distinction without a difference. For elite athletes, any shortfall of performance relative to the competition is inevitably viewed as some degree of "poor" performance, highlighting a need to improve next time. Sometimes poor performance is due to a simple lack of capacity secondary to conditioning, and sometimes it is acquired secondary to disease. Exercise physiology per se does not make that distinction, but it is no accident that many exercise physiologists are also veterinary or medical clinicians who are trained to address both possibilities.

Bayly: Your question really relates to the "applied" aspects of the study of exercise physiology, as per part of my answer to your first question. Most people, including those who own and/or deal with competitive animals, tend to equate exercise physiology with its applications of the more basic study of the production of mechanical energy.

What are the different areas of exercise physiology (cardiovascular, respiratory, etc.)?

Davis: The discipline of exercise physiology probably can be subdivided into infinite areas; it all depends on how closely one wants to examine the phenomenon of exercise and the pre-existing level of knowledge.

An excellent example of this evolving paradigm can be found in the session titles of scientific meetings related to exercise physiology: as long as it has been scientifically studied, scientists have known that heart and lungs play a major role. Early on, they were combined as "cardiopulmonary," but more recently they tend to be separated into "cardiovascular" and "respiratory." And those are increasingly subdivided into "cardiac" and "vascular" and "airway" and "gas exchange." And if the meeting is large enough, you can find "airway" divided into upper and lower disciplines.

At a practical level, there remains overlap due to logistical and financial constraints—one may not have the ability to focus narrowly if there is not the funding stream to support that level of focus, and a facility that can effectively study cardiac aspects of exercise is at least partially equipped (both in equipment and knowledge) to also study pulmonary effects. Currently, in equine exercise physiology, the general areas are cardiopulmonary, muscle, biomechanics, metabolism, fluids and electrolytes, and nutrition.

Not all exercise is the same, obviously. Does exercise physiology overlap among disciplines? What are the bottlenecks of performance for different disciplines?

Davis: To the degree that nearly all organ systems are involved in exercise, there is the potential for considerable overlap across equine athletic disciplines. All exercise and all athletic performance will require a horse to increase activity and stresses on its body to some degree, so some reserve must be present over basal requirements for any type of exercise to occur. But different disciplines require different activities and produce different stresses, just as in human athletes.

A defensive tackle in the National Football League can probably perform a pirouette, but probably not at the level to gain entrance to the Bolshoi. And a ballerina can at least attempt to stop a running back carrying the ball up the middle but is unlikely to be successful (unless he is exceedingly polite). Similar distinctions of form and function are seen in equine athletes—draft horses can produce an amazing amount of raw power but will be ill-suited for a 50-mile endurance competition.

The list of specific limitations for specific disciplines is as long as the list of specific disciplines, and a complete list is beyond the scope of this presentation. But some of the general principles include the capacity for ventilation as a limitation for Thoroughbred racehorses, the availability of fast-twitch muscle fibers for short-duration activities such as rodeo and barrel racing, and heat dissipation for endurance horses. However, these are just broad generalizations, and it is relatively easy to identify exceptions within those disciplines.

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Considering the early days of exercise physiology research, who were some of the key players as far as researchers and veterinarians? What were the breakthroughs in the early days of exercise physiology research?

Davis and Bayly: Individuals that come to mind include Sune Persson, Warwick Bayly, David Snow, Arne Lindholm, Dennis Milne, Gary Carlson, David Hodgson, David Evans, Rueben Rose, and Birgitta Essén-Gustavsson.

Some of the breakthroughs include:

- Use of a treadmill to facilitate the study of exercise physiology;
- Development/standardization of muscle biopsy techniques;
- Development of the biochemistry of equine exercise as a subdiscipline;
- Advent of heart rate meters;
- Emergence of automated analytical methodologies that could be used on equine blood and plasma/serum;
- Factors that led to the determination that exercise-induced pulmonary hemorrhage was due to overpressurization of pulmonary capillaries (i.e., cardiovascular studies characterizing pulmonary hypertension, endoscopy, and bronchoalveolar lavage); and
- The "crossing over" of human exercise physiologists like Bengt Saltin and Phillip Gollnick into the world of equine exercise.

Moving on to the present, what is the state of exercise physiology research?

Bayly: There has been a gradual shift from studies of basic physiology to those pertaining to sports medicine and physiotherapy. There is now a lot more emphasis on biomechanics and locomotion and less on the cardiovascular, respiratory, and muscle responses to exercise.

What areas of exercise physiology study do you feel we completely understand? What areas are less well understood?

Davis: We cannot think of a single area that we could confidently state that we completely understand. There are some areas, such as cardiac performance and upper airway physiology, that have been very well studied. There are others, such as mechanisms and early indications of cyclic fatigue of skeletal structures, that we are just starting to explore at the level that they demand.

Bayly: Some of the more specialized equine sports have not been as well studied as classical activities like racing over all distances; i.e., very short events like barrel racing or Quarter Horse racing to endurance events of more than 100 miles over several days.

Are you currently engaged in exercise physiology research? If so, would you like to describe it?

Davis: My current area of research is in skeletal muscle mitochondrial function, particularly the changes that occur as a result of exercise and conditioning. Skeletal muscle undergoes the greatest metabolic stress during exercise as a result of the increased demand for ATP in this tissue, and as a result, mitochondria must be able to function effectively under a wide range of temperatures and chemical conditions. Often, they do not, and the failure of mitochondrial production of ATP leads to fatigue and possibly (in some instances) muscle damage. Improved mitochondrial function can improve exercise capacity, but we are just now developing the analytical tools and techniques to study changes in mitochondrial physiology at the level necessary to understand their role in exercise.

Bayly: I am mainly focused on:

1. Studies related to the pathophysiology of exercise-induced pulmonary hemorrhage and the effect of putative treatments on its prevalence and severity; and
2. Problems responsible for career- or life-ending injuries in racehorses, and the identification of factors responsible for this attrition to reduce this wastage.

As far as the future, what pressing questions must be asked? What questions are you able to ask now that you were not able to ask five, ten, or twenty years ago?

Bayly: The development of technologies that lend themselves to use in the field rather than on treadmills has the potential to open up new research possibilities in terms of conducting larger studies with greater statistical power and collecting data from horses exercising in their natural environments.

In a nutshell, the availability of digital/Bluetooth technology and batteries that can power onboard data recorders will enable some previously asked questions to be re-evaluated and new ones to be addressed. Good examples relate to the availability of overground endoscopy, ergospirometers, high sensitivity inertial sensors capable of recording >1,000 data points/sec, and ultimately, the application of artificial intelligence programs specifically developed for application in the world of exercising/competing horses, regardless of the discipline.

Are there any new technologies that will help you better understand exercise physiology?

Davis: As previously mentioned, the development of tools that facilitated the detailed laboratory study of exercising horses resulted in major leaps in our understanding of how athletic horses work. Now, we need to move those studies into the field so that we are studying those equine athletes in their "natural" environment, whether that is a show ring, mountain trail, or racetrack.

Preserving the level of analytical precision and detail in the field that is the hallmark of a well-conducted laboratory study is the single greatest challenge that we currently face, and that challenge will be met by improved technology matched with an understanding of what we must quantify and measure.

In today's heightened world of social license, how can exercise physiology research contribute to the well-being of equine sports?

Davis: The movement of high-quality exercise physiology studies into field settings is not just a scientific mandate, it is also mandated by the greater attention paid to social license. By performing the studies on the actual athletes, the public sees that the equine sports community is not just paying lip service to the need to advance scientific knowledge but actually participating in the effort.

Bayly: There also has to be a greater emphasis on generating truly verifiable evidence that stands the test of time with respect to guiding decisions related to conditioning/training and prevention/treatment of various exercise-associated maladies. As it stands, there are still too many supposed "facts" that are too heavily based on opinions rather than actual evidence.

What do you want people to know when they leave your presentation?

Bayly: To have a clearer idea of what is and is not known regarding equine exercise science, and especially, a heightened sense of what we do not know, but need to know as soon as possible.

Can you recommend three to five seminal references on exercise physiology written by you or someone you admire in the field?

1. Franklin, S.H., E. Van Erck-Westergren, and W.M. Bayly. 2012. Respiratory responses to exercise in the horse. Equine Veterinary Journal 44:726-732.
2. Van Erck-Westergren, E., S.H. Franklin, and W.M. Bayly. 2013. Respiratory diseases and their effects on respiratory function and exercise capacity. Equine Veterinary Journal 45:376-387.

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Equine Athletes: Field Performance Testing

Emmanuelle Van Erck, DVM, PhD, DECEIM

Let us begin with a bit of background. What is field performance testing?

Field performance testing is a way to monitor a horse's response to exercise through connected on-board devices in real field conditions.

How did your interest in performance testing arise?

I visited the Equine Sports Medicine Lab at Cornell University as a veterinary student. The sight of a Thoroughbred running full speed on a treadmill sparked my passion for performance testing!

In the past, how did riders assess fitness in horses? How well do people understand fitness in horses? Does it depend on the level of involvement in the discipline?

Until recently, fitness assessments were seldom conducted, and trainers and riders relied on subjective "feelings" on the horse's performances in competition. Conditioning programs have remained empirical. There is still so much to be done and every discipline is different!

What technology do you use when you perform this testing?

We use the Equimetre (Arioneo) to measure heart rate, ECG, speed, and several locomotion parameters and complete with lactate measurements with a handheld meter. But your eyes are also valuable: you get to see the horses ridden and can get information from their behavior, the rider's equitation, the tack, and other factors.

In what disciplines are you currently using field performance testing? Does the testing change among disciplines?

We conduct tests in all disciplines, from racehorses to endurance horses, from jumpers to dressage horses. Of course, each test is tailored to the horse's discipline.

How is field performance testing used, as a regular part of a training program or to evaluate poor performance?

We use field exercise tests to follow a horse's fitness and progress over a competitive season, but we also use exercise tests as a diagnostic tool to assess horses with unexplained poor performance. It allows us to evaluate which problems affect the horse's response to exercise, how and to what level.

Can the testing foretell problems? If so, can you give examples?

Yes, and that is why we need to implement them in our routine assessment of our equine athletes! Racehorses that feel pain have a progressive increase in heart rate, more frequent arrhythmias, and a degradation of their stride parameters that can be detected weeks before an injury.

How can performance testing be used by veterinarians if they suspect a problem?

The tests are so easy to implement once you have the tools. These are commercially available and affordable. It just requires educating horse owners about a new service.

Explain your work with the Belgian Equestrian Federation. Are you using this technology to better understand how these elite athletes prepare for the rigors of international competition?

Yes, we follow both individual riders and national teams. With the Olympics coming up this year, we want to ensure our horses are healthy and fit to bring back medals! This is an excellent prophylactic strategy to ensure we prepare well, not push horses too far, and detect subclinical problems before the horse gets injured.

Based on your knowledge, do you believe that most show jumpers on the international playing field are fit enough to do their job?

No, there is a lot of room for improvement. You cannot expect a horse to perform at his best when he works on the flat an hour a day and goes to the walker. It is important to introduce some cross-training, dressage, and hacking.

How fit are elite performance horses compared to those competing at a level just below that?

They never cease to amaze me by the combination of their physical and mental capacity. Training is also working to boost the horse's mindset.

What would people be surprised to learn about exercise monitoring?

Horses speak loudly about how they feel, and you can tune into that by monitoring them during exercise.

What do you want people to know when they leave your presentation?

Field exercise testing is easy, rewarding, fun, enlightening and, above all, extremely valuable.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on fitness or performance testing written by you or someone you admire in the field?

I would encourage horse owners and trainers to review the chapter on field exercise testing that will be published in the new edition of *Equine Sports Medicine and Surgery* (K.W. Hinchcliff, A.J. Kenaps, R.J. Geor, and E. Van Erck-Westergren, editors), 3rd ed.

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Dressage Training and Conditioning

Hilary Clayton, BVMS, PhD, DACVSMR, FRCVS

How did your interest in dressage come about? Is it a lifelong passion?

Horses are a lifelong passion in both my personal and professional lives. I chose to become a veterinarian at a time when few women were entering the profession. After graduating, I worked in mixed practice for a few years, which was what I had always wanted to do, but then I was curious to try research. It did not take long to realize that was my true calling. Around the time I finished my doctorate, a small group of veterinarians, most of whom were involved with endurance racing, formed the Association for Equine Sports Medicine (AESM). I became a member, and later president, of AESM and found my niche in applying sports science, more specifically biomechanics, to equine athletes.

I believe the last stop on your academic career was Michigan State University, where you were the Mary Anne McPhail Dressage Chair in Equine Sports Medicine. How did this opportunity arise? How did it change your research goals?

I met Mary Anne McPhail in 1980 when I was on sabbatical at Michigan State University. She was a longtime Michigan resident and a very good dressage rider. Michigan was at the forefront of U.S. dressage at that time. Mary Anne and I met at dressage events and sometimes I would be making a presentation.

Fast forward 20 years to the late 1990s when Mary Anne made a donation to MSU (her alma mater) to endow a professorial chair in equine sports medicine with specific reference to dressage horses. I applied for the position and was honored to become the first Mary Anne McPhail Dressage Chair in Equine Sports Medicine.

Being an accomplished rider has undoubtedly helped you with your academic pursuits. Can you give an example of how "living the research," even in your personal time, affected your scientific curiosity?

I have ridden since I was 10 years old and am fortunate to have competed in a wide range of equestrian sports. When I was younger and still living in the U.K., I loved hunting and eventing but after I moved to Saskatchewan in 1983, eventing opportunities were few and far between. I have always enjoyed dressage, and I became increasingly interested in dressage when I lived in Canada.

How do dressage horses differ physiologically from other sport horses?

To be successful in international dressage competitions a horse must be very strong in a highly specific way. Good balance is also important.

Can you explain the differences between "training" and "conditioning"?

I regard *training* as the preparation of the horse to perform the technical demands of the sport. A show jumper must learn good jumping technique, and a working equitation horse learns to negotiate the obstacles on the course. A dressage horse learns to move in self-carriage, which implies an advanced level of balance that facilitates the performance of the most difficult exercises.

Conditioning is the physiological preparation of the horse's body—cardiovascular, respiratory, and muscular systems are trained to fulfill the sport-specific requirements for cardiovascular fitness and muscular strength. Good trainers will find ways to integrate conditioning exercises into the training program.

You have been known to say, "The best way to train and condition a dressage horse is to do dressage." Can you explain what you mean by that?

When I became interested in the science of conditioning, I thought the best way forward would be to understand the physiology and biomechanics of what horses had to do in different sports, and then find exercises to simulate the way the muscles were used in the sport. If you think about a human athlete, they do not just practice the sport, they go to the gym and use exercises that selectively strengthen their muscles to improve their sporting performance. I wanted to develop similar exercises for horses but in dressage the movements performed at the highest levels of the sport cannot be simulated. Dressage horses go through a long training process with each successive level of performance building on what has gone before. It is a stair-step progression; horses learn a new movement, and with practice, their technique improves and their muscles develop accordingly. One of the areas I focus on is using the movements of dressage in an interval training format to build strength.

What are appropriate conditioning programs for dressage horses?

For all sports, I think of conditioning in three phases:

1. The introductory phase is the first 6–12 months of training when the horse is trained in a basic, nonspecific way to carry a rider, to understand the rider's aids, and to develop a baseline level of fitness. Basic core strength is established in this stage.
2. The middle stage of training becomes directed towards a specific type of sport. The horse learns to perform the technical skills and the conditioning work becomes oriented toward the requirements of the sport in terms of sprinting, endurance, agility, and strength.
3. In the final stage of training, the horse is competing at the higher levels of competition and using the movements of the sport to build muscular strength and endurance.

What is the primary factor, in your opinion, that inhibits dressage horses from progressing? Is it training theory, submission, or conditioning factors? If you have a horse with innate athletic ability, what is the best way to maximize that potential?

I would say the primary factor that limits a horse's progression is injury. Most of the injuries we see are repetitive strain injuries, which implies that they arise because of performing the same movements repetitively, day after day. A strenuous workout leads to microscopic damage to the tissues of the locomotor system. The natural course of events is for the damage to heal over the following 24–48 hours. However, if the horse performs the same workout every day the damage tends to accumulate and may eventually become a clinical injury. To keep horses sound, it is important to vary the type of exercise from day to day, to work on different types of footing and terrain—in other words, to do cross-training.

From a physiological perspective, is there a common limiting system among dressage horses? If you can, please explain.

Strength, strength, and more strength! However, it must be specific to the muscles that are used in the sport. Muscles are heavy, and we do not want horses to be carrying around big, heavy muscles that are not needed for the sport.

Excessive muscle development also causes overheating and interferes with heat dissipation. We need dressage-specific muscle strength.

What is it that people misunderstand about the training and conditioning of horses engaged in different disciplines—for example, an upper-level dressage horse and an international show jumper?

In both dressage and jumping, muscle strength is paramount but strength training is very different for the two sports. To maximize the benefits, it needs to be specific, not only to the muscles that are used in the sport but also to the range of motion of the joints and the speed of muscle contraction. Jumpers need an explosive burst of energy from the muscles to propel them over large fences.

Dressage horses use highly controlled contractions in specific muscles to perform the unique movements of dressage. Muscle coordination patterns are important.

Dressage is one phase of three-day eventing. How would you recommend training an event horse for dressage in comparison to a horse involved in solely dressage?

Dressage has become increasingly important as a determinant of the final placing in eventing competitions but the movements performed, even at the highest levels of eventing, do not require the same degree of strength and self-carriage as the specialist dressage horse. In eventing, cardiovascular fitness is the single most important physiological determinant of success. Fitness for dressage in an event horse is not a limiting factor. Dressage training for eventers should focus on the technical aspects of the test.

What do you want people to know about dressage training and conditioning when they leave your presentation?

To maximize the benefits of training and to avoid too many repetitions of the same exercise with the risk of repetitive strain injuries, remember this saying: *"Practice does not make perfect. Perfect practice makes perfect."*

When training the movements, always strive to have the horse perform to the best of his ability, avoid repeating the same movements too often, and use cross-training to vary the workouts. *Training teaches technical excellence. Cross-training keeps horses sound.*

At the risk of being repetitive, I would like the audience to go away understanding how the technical demands of dressage, as espoused in the pyramid of training, require great strength that comes from practicing the movements correctly but excessive repetitions lead to repetitive strain injuries. Horses need to get out of the sandbox and do other types of exercise to reduce the risk of injury.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on training and conditioning—generally or dressage specifically—written by you or someone you admire in the field?

Book recommendations:

1. *Conditioning Sport Horses* by Hilary M. Clayton. Published in 1991 by Sport Horse Publications, Mason, Michigan. Available from SportHorsePublications.com or Amazon.com.
2. *Activate Your Horse's Core* by Narelle C. Stubbs and Hilary M. Clayton. Published in 2008 by Sport Horse Publications, Mason, Michigan. Available from SportHorsePublications.com or Amazon.com.

Paper recommendation:

1. Clayton, H.M. 2016. Core training and rehabilitation. Veterinary Clinics of North America: Equine Practice 32:49–71.

Comparative Nutrition of the Perfomance Horse: An Energetic Perspective

Joe Pagan, MS, PhD

Horsemen tend to lump all performance horses into a single group. Is this an oversimplification of the various exercise demands we place on them?

Yes, from a nutritional perspective, this is an oversimplification. For most performance horses, there is usually a specific type of exercise they are asked to do, and that exercise ultimately influences how we feed them. All performance horses have locomotion in common. Locomotion requires repeated muscle contraction. Given this, "performance" becomes a measure of which muscles are used, how often those muscles contract, and how forcefully those muscles contract.

Muscle contraction is a commonality among all performance horses, as is the fuel muscles use to make this happen. Muscles use a universal fuel, adenosine triphosphate (ATP). Despite its involvement in all muscle contraction, ATP is not stored in large amounts in muscle tissue. For the horse to have muscle contractions, metabolic pathways must be in place that allow potential energy sources to be converted to ATP quickly, sometimes replenished as quickly as it is used.

What fuels are available to generate ATP?

If we look at the fuels available to muscle to create ATP, the horse has choices. The first is fat. Horses can burn fat, breaking it down in the presence of oxygen (aerobically) to create ATP in the mitochondria of cells. Fat is a useful fuel because it is plentiful, as it is stored throughout the body. Despite its availability, fat is a slow fuel. If the horse needs to generate ATP more quickly than burning fat allows, glucose becomes the fuel of choice.

In the presence of oxygen, glucose can be converted to ATP. This conversion is efficient and twice as fast as burning fat. However, the amount of glucose, stored in muscle tissue and liver as glycogen, is limited. Running out of glycogen is an issue for many performance horses. For horses doing intense work, they can use glucose in the absence of oxygen (anaerobically) to make ATP. One end product of this process is lactic acid, so the anaerobic generation of ATP is self-limiting. The more frequently muscle contraction occurs (with speed, for example), the more glucose is used for ATP production. When the horse must produce ATP anaerobically, glycogen utilization increases exponentially. Those are the fuels used by horses to create ATP, but that is not what we feed horses.

What energy sources are fed to horses?

In terms of energy sources, we have a choice of four: plant fiber, nonstructural carbohydrates (NSC; sugar and starch), fat, and protein. In the United States, we speak about energy in terms of digestible energy (DE). DE requirements are calculated based on the horse's maintenance DE requirement plus the additional energy expended during exercise. DE is the measure of how much energy is contained in a feed (the difference in how much is digested and how much remains in the feces, both of which can be measured).

For this presentation, which focuses on a comparative approach, I am going to use a different way of talking about energy sources in feed. Normally we talk about the percentage of different energy sources in feeds (e.g., 8% fat or 30% NSC), but I am going to use the relative energy provided for each energy source as a percentage of the total DE the horse consumes. To do that, you have to know three things about an energy source: (1) the gross energy content, (2) the quantity of the energy source in the feed, and (3) the digestibility of that energy source.

When talking about energy sources, plant fiber from forage makes a huge contribution to total DE. For the sake of this presentation, though, I am going to zero in on the energy sources contained in concentrates.

Can you describe in greater detail the sources of DE?

When considering plant fiber in feed, we generally talk about total dietary fiber, composed of two components: cell wall (neutral detergent fiber or NDF) and soluble fiber (indigestible by mammalian enzymes but without any structural components; pectin is an example). We differentiate these two components because of their digestibility.

NDF has lower digestibility than soluble fiber. Regardless, both fiber sources produce volatile fatty acids (VFAs), which are the end products of their fermentation. Horses can absorb VFAs and make some energy substrates, such as fatty acids and glucose. The two most popular fiber sources used in concentrates are beet pulp and soy hulls. The fiber in beet pulp tends to be very fermentable because a high proportion of it is considered soluble. In contrast, soy hulls contain more NDF than soluble fiber, though all of it is quite digestible.

Starches and sugars are considered nonstructural carbohydrates. Starch is the main source of energy in cereal grains. Horses break down sugars and starches into glucose units in the small intestine, where they are absorbed into the blood. Glucose can be oxidized directly to produce ATP or can be changed to glycogen and stored in the muscle or liver. As I've mentioned, muscle glycogen and liver glycogen are important fuels for energy generation during exercise.

Fat is another source of DE, and it is more energy-dense than cereal grains. Horses can digest fat well, and it can have some performance benefits. The total amount of fat fed and the fatty acid profile of the fats are important considerations. Fat typically comes as vegetable oils or fat contained inside cereal grains themselves.

Although protein is considered an energy source, we typically do not manipulate it as a source of energy in feeds.

How do performance horses use these different sources of DE?

All horses can use any of those energy sources to fulfill maintenance needs and to fuel locomotion, but not all horses need the same ratio of those energy sources in their feed. The factors that affect which energy sources are appropriate for different performance horses include discipline, breed, temperament, desired body condition, and energy level of the horses (a combination of exercise intensity and duration).

If we place all performance horses on a spectrum in terms of exercise intensity and duration, an Arabian endurance horse would be on one end of the spectrum and a Thoroughbred racehorse would be on the opposite end. These two horses have different muscle compositions. Arabians have been bred to go long distances at low to moderate speeds, while Thoroughbreds have been bred to go short distances at high speeds.

We did an interesting study 20 years ago in which we took a set of Arabians and a set of Thoroughbreds, and we fed and trained them in the same way. The exercise was done on the treadmill, so it could be carefully controlled. When we exercised those horses at long, slow distances, the Arabians burned more fat than the Thoroughbreds. The Arabians preferentially chose fat as the substrate they wanted to use. In contrast, during the faster exercise, the Thoroughbreds generated more lactic acid, which is a signal of anaerobic metabolism. That showed the Thoroughbreds recruited muscle cells that were more dependent on glucose use.

Endurance horses perform at low- to moderate-intensity, so fat utilization is high, but they still burn a lot of muscle glycogen because of the long duration of exercise. Let us look at a typical endurance feed. Such a feed will likely call on all the energy sources. There will be added fat (22-25% DE as fat), so it would be considered a relatively high-fat feed, but there should also be plenty of NSC (~30% DE) and fiber (~30% DE).

Thoroughbreds engage in exercise characterized as high intensity and short duration. During training and racing, they generate energy anaerobically, burning glycogen and producing lactic acid. They typically require a mixture of energy sources that would skew more toward more calories coming from NSC and less coming from fat compared to an endurance horse. Racehorse feeds typically derive 45–50% DE from NSC. Interestingly, if you compare the NSC content from popular racehorse feeds in the United States and Europe, they tend to have a similar concentration of NSC, meaning they are reasonably high in NSC.

If we anchor those two performance horses as the two extremes, where endurance horses have more dependency on fat and fiber and less dependency on NSC compared to racehorses with a high dependency on NSC, what about the horses in between?

Many performance horses fall into the "in-between" category. One interesting example is the three-day event horse. Three-day eventing combines a range of exercise intensities, from fairly low to quite high. Exercise duration is similarly mixed, from a dressage ride of a few minutes to a much longer cross-country phase. At times, it can be difficult to know how to best fuel the event horse because of the types of exercise involved. Beyond that, not all event horses compete at the highest echelons of the sport, so the energy requirements should not be considered monolithic. This holds for other disciplines as well, i.e., a range of energy requirements based on the level of sport can be found.

We did a study where we followed the training and competition programs of event horses at several different levels, from Novice to Advanced, which would be approaching the elite tier of the sport. In this study, we followed the horses' day-to-day training by monitoring their heart rate during exercise, exercise duration, and diet. The purpose of this was to better understand energy requirements.

We converted the data collected (heart rate, exercise duration) into DE requirements. We could do that because heart rate gives a good indication of oxygen consumption. Oxygen consumption can then be used to calculate caloric consumption.

We found that when expressed as a percentage of maintenance the DE requirement increased as the level of competition increased. In other words, horses training for Novice competition had a lower DE requirement than those at the higher levels of competition. We could predict how much energy horses needed based on two different parameters: how many kilometers they trained per week or how many hours they trained per week. That speaks to the duration of exercise and how much you must feed. When compared to the NRC (*Nutrient Requirements of Horses*), those horses would have spanned from light exercise to heavy exercise in a single discipline that covered the entire spectrum of predicted DE requirements.

We also followed a lot of these horses at different levels of competition. When I mention competition, I am referring to the cross-country phase, which is the most strenuous phase of eventing. We wanted to understand how much of the energy needed was generated from muscle glycogen utilization, either aerobically or anaerobically. Muscle glycogen utilization is difficult to measure in real life because it involves muscle biopsies. We can, however, look at lactic acid in the blood to gauge how much anaerobic glycolysis occurs. Blood samples are easy to obtain. With this in mind, we looked at how much lactic acid was produced during the cross-country phase, but we also know from treadmill studies that heart rate can predict lactic acid concentrations. Using this noninvasive tool gives us an idea of how much muscle glycogen is used during different types of exercise.

We also wanted to understand how that related to different levels of exercise within one sport—in this case, eventing. We found that horses competing at the two highest levels (Intermediate and Advanced) had heart rates greater than 200 beats per minute (bpm) for much of the time, indicative of producing a lot of lactic acid. Horses engaged in lower levels did not have heart rates over 200 bpm often but they did have heart rates between 175 and 200 bpm, so they too were producing lactic acid at that point.

The bottom line with these studies is this: event horses at lower levels do not necessarily need a lot of carbohydrates in their feed but, as exercise increases based on level, higher levels of NSC to replace the glycogen used during more strenuous exercise is essential.

Many of the horses in this study were owned and managed almost exclusively by amateurs. What about the elite horses, such as Olympic-caliber horses?

Our involvement in the Olympics and the World Equestrian Games provided us insight into what high-end sport horses consume. Because we have arranged to ship and deliver feeds to the teams at many international competitions, we were able to see which feeds were the most popular. The compositions of those feeds were determined by chemical analysis, and we calculated the DE distribution of those feeds.

At the 2018 World Equestrian Games, we identified the 10 most popular feeds in five disciplines: reining, show jumping, dressage, three-day eventing, and endurance. We found the average fat content of those feeds to be 5-6% fat. Reining horses had diets lowest in fat, while endurance and eventing tended to be higher, with show jumping and dressage intermediate. Expressed as a percentage of DE, the feeds ranged from an average of 18% for endurance to 13% for reining.

When we looked at the percentage of NSC, we observed the opposite. Reining horses consumed feed highest in NSC, and endurance and eventing horses consumed feed with the lowest. The range was narrow between low and high (eventing 26% NSC; reining, 32% NSC) which equaled 32-42% expressed as a percentage of DE.

The 10 most popular feeds were all manufactured in Europe, as most horses competing were European. This data does not speak to what the American horses were fed, but the American horses were only a fraction of the total number of horses. Over 50 different national federations were represented, and these feeds were fed at home and then provided at the competition.

To the American audience, those sport horse feeds may seem fairly low in fat and high in NSC, and that is because there has been a real trend in American sport horse feeds to be higher in fat and fiber and lower in carbohydrates.

When did this trend toward high-fat, low-starch feeds start?

This trend began in the 1990s around the time we developed RE-LEVE®. At that point, most performance horses were fed low-fat feeds with traditional amounts of carbohydrates. In formulating RE-LEVE, we aimed to address a specific myopathy in Thoroughbreds, a disease called recurrent exertional rhabdomyolysis (RER). We wanted to see if feeding a low-starch feed that derives most calories from fat and fiber could help those horses.

The feed worked, but the final formulation ended up being quite low in carbohydrates, with only 15% of calories from NSC, but high in fat (30% of calories). This was a real departure from how racehorses were traditionally fed but, remember, RE-LEVE was made specifically for horses with RER.

One side effect of feeding a low-starch, high-fat concentrate involved temperament. The feed tended to have a calming effect on those horses susceptible to RER. Since that time, low-starch feeds have become popular for sport horses. Without question, when it comes to American sport horse feeds, there has been a tendency to feed more and more low-starch, high-fat concentrates since that time.

Today, it is not unusual to find sport horse feeds in the United States with under 20% of calories from NSC and 25-30% from fat. These are popular feeds and they seem to work well in many situations. In research studies, however, we found if horses do enough exercise to significantly deplete muscle glycogen, they cannot replete muscle glycogen quickly when fed low-starch, high-fat feeds.

Today, we have lots of different feeds with different "octanes" depending on NSC and fat content. In terms of the amount of energy coming from NSC, at one extreme we have racing feeds with the highest percentage. Elite European sport horse feeds fall in the middle. At the other end, we have American feeds rich in fat and fiber, such as RE-LEVE.

We have a much broader understanding of energy sources than we had 20 years ago. At the onset, this can be confusing to horse owners. If, though, they look at what exercise they are asking of their horse—is it low-intensity, long-duration exercise or is it high-intensity, short-duration exercise?—it becomes easier to see what is appropriate. Low-intensity exercise should skew toward low-starch, high-fat feeds; high-intensity, short-duration work should tip toward more NSC and lower fat.

Where do you see future research headed in this field?

There have been major advancements in our ability to measure exercise intensity and duration in all types of performance horses under field conditions. Using these wearable technologies to measure activity and heart rate in individual horses will allow us to fine-tune rations to suit a specific horse's energetic requirements.

Exercise-Induced Pulmonary Hemorrhage (EIPH): Not Just a Racehorse Disorder

Warwick Bayly, BVSc, MS, PhD, DACVIM
Emmanuelle Van Erck, DVM, PhD, DECEIM

What is exercise-induced pulmonary hemorrhage (EIPH)?

Exercise-induced pulmonary hemorrhage (EIPH) is a condition in horses where bleeding occurs in the lungs during intense exercise. It is not a disease. It most likely occurs because of the physiologic responses or adaptations that allow horses to exercise as strenuously as they do. This bleeding is typically associated with breaks that develop in the pulmonary capillaries, which are the smallest blood vessels in the lungs.

What horses are most affected by it, those competing in what disciplines? What is the prevalence of EIPH in different disciplines?

The horses most affected by EIPH are those engaged in strenuous athletic activities, particularly racehorses and performance horses in disciplines such as barrel racing, polo, show jumping, and eventing. You could say that it is an occupational hazard for them.

Based on a single endoscopic examination, the prevalence of EIPH is estimated to be between 44%–75% in Thoroughbred racehorses, 26% in Standardbreds, 62% in racing Quarter Horses, 40% in three-day event horses, 45% in show jumpers, 40% in elite endurance horses, 11% in polo ponies, and 26% in draft pulling horses. However, if horses are scoped multiple times, the prevalence increases markedly and, if the diagnosis is based on the results of bronchoalveolar lavage (BAL), the prevalence approaches 100% in racehorses and barrel racers.

Can you speak in depth about any difference observed in horses engaged in different disciplines and EIPH? Is the etiology for EIPH the same from discipline to discipline?

The fundamental physiologic mechanism responsible for EIPH is referred to as stress failure of the pulmonary capillaries and is likely the same across disciplines. Namely, anything that causes an increase in the blood pressure in the capillaries and/or the inspiratory pressure in the lungs could ultimately lead to EIPH if the magnitude of the increases in one or both pressures is great enough.

While the exercise alone can cause EIPH if it is intense enough, other factors like respiratory tract and/or pulmonary inflammation, airway obstruction, cardiac conditions, genetic predisposition, and a horse's athletic history in terms of the number of races or competitions it has been in may increase the severity and likelihood of EIPH occurring. Different disciplines may expose horses to varying levels of these factors. Sport horses involved in disciplines such as eventing, dressage, and show jumping are usually older when they reach higher levels of competition and are more prone to chronic lower airway conditions such as asthma.

The prevalence of EIPH in racehorses appears to be similar in horses two- to four-years-old after which it progressively increases. The severity of EIPH also gradually increases as the cumulative number of race starts (and breezes and timed workouts) rises.

There are many medications, dietary supplements or manipulations, and management protocols that have been promoted as preventing or reducing EIPH.

However, the administration of furosemide (Lasix) four hours before racing and the wearing of the equine nasal strip (FLAIR®) are the only treatments that have been scientifically proven to reduce EIPH. There is nothing that has been shown to prevent EIPH in all racehorses.

Management of sport horses and treatments authorized around the time of or at the competition have not been investigated, but could also play a significant role in the varying prevalence of the condition from country to country or across disciplines.

What are the performance implications for horses with EIPH?

EIPH has been associated with reduced performance capacity in racehorses when it is categorized as "severe." Severe EIPH is relatively uncommon (<15%), although its prevalence increases as the number of starts increases.

However, not all horses with severe EIPH perform poorly. While they are less likely to win or place in races, plenty of horses with severe EIPH do so. The presence of blood in the airways is pro-inflammatory and repeated EIPH causes remodeling of the pulmonary tissue, vasculature, and airways, which can irreversibly compromise respiratory function during exercise.

Is EIPH preventable?

That really depends on the intensity of the exercise; i.e., the level at which the horse is competing. If the horse runs more slowly, jumps lower fences, and competes less often, it either might not experience EIPH or the EIPH that does occur will be less severe. Doing everything possible to ensure that there is no co-existing respiratory or cardiac disease that could be responsible for increased blood pressure in the pulmonary capillaries and/or inspiratory pressures in the lungs is very important, especially in situations where the administration of Lasix and/the wearing of nasal strips is banned.

What are common treatments for EIPH?

Common treatments for EIPH include rest, respiratory health support, and in some cases, medications to address the underlying pathologic causes when they are present. Furosemide is often used to control EIPH by reducing pulmonary capillary pressures. It and the use of nasal strips are the only proven means to effectively mitigate pulmonary hemorrhage. The link with inflammation is recurrent in most studies.

Explain the use of furosemide to control EIPH.

The way furosemide works is not completely understood and is probably not straightforward. While furosemide is best known as a potent loop diuretic, it exerts several other effects. The diuresis results in increased urine production and temporary loss of fluid from various tissues, including the lungs. However, much of this effect starts to diminish about two hours after its administration. After four hours, much, if not all, of normal body compartment fluid volumes have been restored.

In addition to its short-acting diuretic effect, furosemide also causes relaxation of pulmonary venous smooth muscle. This increases the compliance of these vessels enabling them to accommodate increases in blood volume without the same increase in pulmonary venous blood pressure and, by extension, pulmonary capillaries. This is reflected by decreases in pulmonary artery wedge pressure following the administration of furosemide. The duration of this effect is uncertain, but it seems to persist well after the drug's diuretic effect has dissipated. However, this does not mean that the diuresis does not play a role. The kidney releases prostaglandins (PGs) in response to the diuresis and these PGs may mediate the relaxation of smooth muscle in the pulmonary veins and other parts of the body. A role like this might help explain why treatment with NSAID drugs before the administration of furosemide dampens or blocks the effects of furosemide on pulmonary blood pressure.

The use of furosemide in horse racing has been a topic of debate and controversy for many years. While it may reduce the severity of EIPH, concerns have been raised about its potential for masking the presence of other drugs in a horse's system. This was only germane when drug testing was based on urinalysis.

Nowadays, however, the combination of blood testing and increased sensitivity of analytical tests has rendered that concern moot. Regulations regarding the use of furosemide vary between racing or sport jurisdictions, but FEI prohibits it.

Typically, furosemide is administered four hours before racing, but there has been chatter about changing the time of administration to be earlier. How would changing the time affect the efficacy of furosemide?

Just how long furosemide exerts its effects is uncertain. While the diuretic effect is relatively short-lived, there is evidence that the increased urinary excretion of ions like sodium and calcium persists for more than 24 hours following a single furosemide injection.

One study with a small number of horses (n=6) found that the combination of controlled access to water for 24 hours (3 liters every 4 hours) and a single injection of furosemide 24 hours before a simulated race was associated with decreased EIPH based on post-exercise endoscopy. However, the BAL red blood cell number was not different. A lot more research needs to be done to determine the duration of furosemide's many varied effects in horses.

Can you describe the use of nasal strips, e.g., FLAIR strips, and the effect they have on EIPH? What is the mode of action? Do you have any other thoughts on their use?

FLAIR nasal strips have been shown to reduce nasal resistance to airflow by preventing or lowering the extent to which the soft tissue that makes up a horse's nares and false nostrils tend to collapse when the horse breathes in.

As such, this likely reduces the peak negative inspiratory pressure in the lungs, thereby lowering the pulmonary capillary to alveolus transmural pressure and reducing or eliminating the number of breaks associated with pulmonary capillary stress failure, and EIPH.

Do three-day-event riders use furosemide? Is it a legal substance?

For FEI competitions, anti-doping rules are strict and tightly enforced. The use of medication such as furosemide is forbidden and cannot be a systematic or long-term option. If and when it is used during training is not well known.

Are there any new treatments on the horizon?

Phosphodiesterase type-5 inhibitors (the best known of which is sildenafil [Viagra]) have been shown to reduce pulmonary artery blood pressure during high-speed treadmill exercise. Whether it does the same for pulmonary venous and capillary pressure and reduces the severity of EIPH has not been reported.

Research on EIPH is ongoing and focuses more on when it does and does not affect performance in horses engaged in disciplines besides Thoroughbred racing, rather than treatment per se. The horse industry needs to stay updated and informed on evidence-based approaches to managing EIPH. A single miracle treatment or supplement claiming to treat EIPH is not currently available and possibly never will be.

What would surprise people about EIPH or the use of furosemide?

EIPH can be caused by musculoskeletal pain and resolved by simply addressing the underlying lameness. However, the use of certain drugs such as pentosan, heparin, or aspirin with anticoagulant effects can increase the risk of EIPH. Cardiac arrhythmia, particularly if persistent like atrial fibrillation, can also be associated with EIPH.

What do you want people to know about EIPH when they leave your presentation?

Understanding the multifaceted nature of this condition is fascinating—one size does not fit all. However, if there is no underlying respiratory or cardiac pathology, options are probably limited beyond increasing the time between competitions and decreasing the frequency and/or duration of strenuous training sessions. That is not always possible. Thoroughly assessing the status of the respiratory tract and heart, much like you would approach a lameness case, can lead to effective management strategies.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on EIPH written by you or someone you admire in the field?

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Equine Myopathies

Stephanie Valberg, DVM, PhD, DACVIM, DACVSMR

Can you provide an overview of equine myopathies?

Myopathy is a clinical term used to describe muscle disease. There are numerous causes of myopathies in horses. To determine the potential cause of myopathy, a history, physical examination, blood work, genetic testing, and muscle biopsy are all potential diagnostic tests to come to a diagnosis.

Based on history, myopathies are classified as either exercise-associated or not exercise-associated. In this proceedings, we will focus on myopathies associated with exercise. To further define the cause of exercise-associated myopathies, the role of muscle weakness, atrophy, and muscle pain are explored. Muscle weakness and atrophy can have numerous causes in horses and can affect a single muscle or many muscles. From a nutritional standpoint, caloric balance, amino acid balance, and vitamin E can all contribute to generalized muscle atrophy. Vitamin E deficiency causes either a vitamin E responsive myopathy (VEM) or equine motor neuron disease (EMND) where vitamin E deficiency damages nerves supplying the muscles.

Myopathies causing muscle pain are subdivided into those that damage the muscle cell membranes resulting in rhabdomyolysis (the degeneration of skeletal muscle fibers) and those that cause pain from internal damage to the muscle cell without disrupting the muscle cell membrane. Rhabdomyolysis is determined by measuring serum creatine kinase (CK) and aspartate transaminase (AST) activities in the bloodstream. These muscle enzymes leak out of muscle cells into the bloodstream if the cell membrane has been damaged. It is important to measure CK and AST because the approach to treating rhabdomyolysis is fundamentally different from treating exertional myopathies that do not cause rhabdomyolysis. Dietary approaches include low-starch, high-fat diets with regular exercise and therapeutic approaches include dantrolene before exercise.

Exertional rhabdomyolysis (ER) may only occur once, in which case it is termed sporadic and is likely due to a temporary imbalance in training intensity or diet or sometimes follows corticosteroid joint injections. Chronic ER can have many causes. These include type 1 polysaccharide storage myopathy (PSSM1), the form of type 2 polysaccharide storage myopathy that causes ER (PSSM2-ER), recurrent exertional rhabdomyolysis (RER), malignant hyperthermia (MH), and myofibrillar myopathy in Arabians (MFM-Arabian). MFM-Arabian is a form of oxidative muscle damage that causes ER in endurance horses toward the end of endurance races.

Some myopathies do not cause rhabdomyolysis but do cause muscle pain. Myofibrillar myopathy (MFM) is one such myopathy. Horses with MFM have normal serum CK activity but have reluctance to go forward, to engage their hindquarters and collect. They may balk at exercise or develop behavioral aversions. Many horses with normal CK previously diagnosed with PSSM2 have been shown to have MFM. It appears to be due to oxidative damage that particularly affects a cytoskeletal protein called desmin, and in Warmbloods, weakness within the myofibrils (contractile proteins) and an inadequate training response. Dietary approaches include moderate-starch, low-fat diets with days off exercise, and therapeutic approaches include amino acids and N-acetyl-cysteine found in MFM Pellet™ as well as antioxidants such as coenzyme Q10, such as the product Nano-Q10™. These products are available through Kentucky Equine Research.

How do you diagnose myopathies? When is a biopsy needed, when is a biopsy not needed?

The diagnosis of myopathy is based on the history, physical exam findings, lameness exam, neurologic exam, and blood tests for CK, AST, and vitamin E. Depending on the results of these tests and the breed of the horse, genetic testing for PSSM1 and MH may provide a diagnosis.

In chronic cases with difficult-to-manage rhabdomyolysis, a muscle biopsy may be needed to make a specific diagnosis. Muscle biopsy identifies PSSM1, PSSM2, and MFM as well as many other myopathies. Horses with RER usually do not have any specific histopathologic abnormalities.

How has the muscle biopsy process changed over the years?

Neuromuscular diagnostic laboratories engaged in research prefer to evaluate frozen sections prepared from fresh muscle biopsies. The muscle must be shipped fresh on ice overnight to the laboratory. This allows for staining of many different substrates and enzymes, and isolation of DNA from the sample for future genetic research. Formalin-fixed sections have not been helpful because hematoxylin and eosin stains are traditionally used.

Recently, specific stains that include immunohistochemical stains have been developed to diagnose known equine muscle diseases. The advantage of formalin-fixed tissue is the ease of shipping samples without chilling, and the length of time they can be maintained in formalin. The formalin should be fresh (used within 6 months of purchase) for optimal immunohistochemical staining.

Can you explain the Valberg Neuromuscular Diagnostic Laboratory, and how it is designed to help veterinarians?

Following my retirement from academia, I wanted to continue to offer a diagnostic service to veterinarians.

The Valberg Neuromuscular Diagnostic Laboratory (NMDL) (www.ker.com/nmdl/) is designed to provide information about muscle diseases through our website and to provide a muscle biopsy diagnostic service for veterinarians. This service uses formalin-fixed muscle samples. Samples are kept in formalin for 4 days and then processed weekly with results sent to veterinarians within 7–10 days. We have recently offered a special needle biopsy technique described under the "how to take a biopsy" tab on our website.

This specialized needle is the size of a pen and is used to sample the gluteal muscle in horses with exertional myopathies. The advantage of this technique over an open surgical biopsy is that horses can go back to work the next day. We have full information on ordering the needle and performing the needle biopsy, including videos, under the muscle biopsy tab.

Explaining the nuances of treating myopathies is beyond the scope of this proceedings. Can you provide a reference for veterinarians and horse owners that can set them on the right path in terms of diet and exercise?

Joe Pagan and I put together a paper for the 2020 American Association of Equine Practitioners Convention, and it is printed in the proceedings from that meeting. Here's the complete reference: Pagan, J.D., and S.J. Valberg. 2020. Feeding horses with myopathies. In: Proc. American Association of Equine Practitioners 66:66–74.

This paper provides solid recommendations on how to manage myopathies through nutrition. Of course, horse owners will want to work closely with their veterinarians to land on a plan that works for individual horses.

Access to the paper is also available through this website address: https://ker.com/published/feeding-performance-horses-with-myopathies/.

Is there any breed of horses that is truly immune from muscle disease?

No, as athletes, all breeds of horses are prone to some sort of muscle disorders from strains to chronic issues.

What frustrations have you endured over the years as you have studied myopathies?

The general lack of research funding for equine research slows progress. With the high student debt load and changes to visas, it is harder to encourage veterinary students to do graduate work, and this slows the pace of research and will impact future work.

I am very grateful for the past support of the American Quarter Horse Foundation, the Grayson Jockey Club Research Foundation, the Morris Animal Foundation, and the Mary Anne McPhail and Martha Wolfson endowments, university grants, and Kentucky Equine Research, but many great projects go unfunded. If owners want answers, they need to proactively support these organizations.

What factors continue to hinder diagnosing and treating horses with muscle disease?

Getting to the bottom of performance-limiting problems in horses can be complex and costly. Owners sometimes bypass their vets and look to the internet for answers. A commercial company offers genetic tests for PSSM2 and MFM, for example, and owners often do not realize that the genetic testing industry is unregulated. The company offering these tests has not published scientific papers validating their tests.

My research lab has published studies that compared the results of the P2, P3, and P4 commercial genetic tests with muscle biopsy results, and we did not find that the genetic tests accurately diagnose PSSM2 and MFM. Because of this, we do not recommend their use as they can lead to false positive and false negative diagnoses.

What do you want veterinarians to know about myopathies when they leave this presentation? What might surprise them?

Most exertional myopathies were previously treated with a low-starch, high-fat diet. That has changed in recent years after the discovery of myofibrillar myopathy.

Can you give veterinarians any words of wisdom when diagnosing and managing myopathies?

When a myopathy is suspected, check serum CK and AST. If the history fits with an exertional myopathy but results for CK and AST are normal, consider an exercise test with CK measured 4-6 hours after exercise.

If you establish that the horse has a form of ER, then use either genetic testing, a muscle biopsy, or a treatment trial to figure out what form of chronic ER the horse has. If you know you have ER, then dantrolene is a very valuable tool to use to prevent episodes. Dietary approaches for ER involve low-NSC and higher-fat diets. Daily exercise is also recommended for horses with ER.

If the horse has a history of reluctance to go forward and reluctance to engage, rule out other causes of exercise intolerance first (gastric ulcers, lameness, etc.). If these are negative, then a muscle biopsy would be indicated to evaluate horses 8 years of age or older for MFM. If younger, a biopsy may not show typical changes yet (desmin aggregates) and a 4-week treatment trial might be the best approach to diagnose MFM.

For MFM horses, a moderate-NSC, low-fat diet enriched with antioxidants, amino acids, and N-acetyl-cysteine is most effective. We have developed a product, MFM Pellet, for these horses. Days of rest benefit MFM horses rather than daily exercise.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on equine muscle disease written by you or someone you admire in the field?

1. Valberg, S.J. 2018. Muscle conditions affecting sport horses. Veterinary Clinics of North America: Equine Practice 34:253-276.
2. Valberg, S.J., A.M. Nicholson, S.S. Lewis, R.A. Reardon, and C.J. Finno. 2017. Clinical and histopathological features of myofibrillar myopathy in Warmblood horses. Equine Veterinary Journal 49:739-745.
3. Valberg, S.J. 2021. Diagnosis and management of myofibrillar myopathy in Warmblood performance horses. In: Proc. American Association of Equine Practitioners 67:214-218.

Squamous and Glandular Gastric Ulcers: New Treatment Options

Ben Sykes, BSc, BVMS, MS, DACVIM, DECEIM, MBA, PhD

You are one of the world's authorities on equine gastric ulcer syndrome (EGUS)? Can you tell us how you got to this point?

I have been a veterinarian for over 26 years, and I have always worked with horses. I did my residency in the United States, and during that time I worked a lot with Mike Murray, who was one of the original gastric ulcer guys. I then moved to Finland and worked with sport horses, and gastric ulcers were a major part of the caseload on a day-to-day basis there. I recognized at that time that we had some successes with certain treatments and then some limitations with others. At the time, we were just starting to realize the distinction between squamous and glandular disease and just how important that was. I returned to Australia in 2009.

Once I got settled, I was interested in exploring this further from a research perspective, so I started some work on the factors that influenced the effectiveness of omeprazole.

This led to my doctorate work on the subject, and since then I have done a range of work in other areas. In sum, my interest began when I ran into cases that were not responding the way I wanted them to and looked at ways to optimize treatment to get the best results for the money and the best results for our horses. My interest grew from there.

Can you give us an idea of the prevalence of squamous and glandular gastric ulcers in various disciplines (racehorses vs. sport horses, for example)?

If we start with squamous gastric disease, squamous disease is very much related to management and intensity of management.

When I say "intensity of management," I am thinking about things like dietary factors, such as the amount of roughage (inadequate roughage, specifically), the amount of carbohydrate (excessive carbohydrate, specifically), and the amount of exercise (cumulative exercise).

The prevalence of squamous disease relates to that. Endurance horses and racehorses that have very high exercise loads and that consume low-roughage, high-carbohydrate diets, have a very high prevalence of squamous disease, anywhere from 80-100%, depending on the specific population. When we are looking at squamous disease in sport horses, it varies a lot from region to region primarily because of management styles, though we are still in the 50-75% range. Again, the higher the risk factors, the more likely we are to be closer to 75%. When we have sport horses in competition, even with good management, a reasonably high percentage of 50% or more are still at risk of squamous disease.

Glandular gastric disease is a different beast. It is not related as directly to the intensity of management, so we see a different set of distribution with it. We do not see a strong predilection in endurance horses and racehorses, as we do with squamous disease. Glandular disease is more typically a disease of sport horses, especially Warmblood sport horses. The Warmblood breeds are particularly predisposed to glandular disease.

What does that mean in terms of numbers? When looking at sport horse populations, we do not have huge datasets here, but we are looking at a 50-75% range for horses in competition. When thinking of endurance horses and racehorses with glandular disease, we are talking more like 25-50%, so it is an interesting relationship. Those horses at most risk for squamous disease are at lower risk for glandular disease.

Ulcers gained recognition in the 1990s and the introduction of omeprazole followed not long after that. At this point, everyone seemed to be focusing on squamous disease. Why was that?

When squamous disease was first recognized as a significant disease of the horse in the 1990s, it came down to the size of the endoscopes being used. Originally, 2.5-meter endoscopes were being used, and these were taken from human medicine. With an average-sized horse, it is about 2 meters from the nose to the entryway to the stomach, so this only gave about 50 centimeters to play with in the stomach. The stomach is one chamber, with the squamous portion in the top half and the glandular portion in the bottom half.

With a 2.5-meter scope, what we can see well is the top half of the stomach. So, in the late 1990s and early 2000s, squamous gastric disease was recognized as a widespread problem. With that came a lot of understanding about elements that contributed to disease risk. We then started using 3-meter or 3.5-meter scopes, and we recognized that glandular disease was important as well.

When did glandular gastric ulcers come onto the scene?

We began to see a clear distinction between squamous and glandular disease in 2015 or so. Part of this was because it took some time to familiarize ourselves with looking at the glandular mucosa through regular examinations. With the longer scopes, it is easier to get to the glandular region, but it takes some practice to maneuver the scope. Most of the lesions are deep within the pylori antrum.

In most horses, there is no relationship between squamous and glandular disease, so you just couldn't look at a horse and say, "The stomach is clear because the top half looks good, so the bottom half must look good, too." Ultimately, it was around the 2015 mark when the European Consensus statement came out. (See a full reference for this article at the end of this Q&A). One purpose of the consensus statement was that we wanted to make a clear distinction between squamous disease and glandular disease.

H*ow are the two types of gastric ulcers different?*

Squamous gastric disease is primarily a result of management, effectively an acid burn to the squamous mucosa. The squamous mucosa has minimal protective mechanisms and is not designed in the horse to be exposed to an excessive acid load.

Because we change management factors, specifically limited access to roughage, we see increased acid exposure to the squamous mucosa, and its simple defenses cannot protect it. At its core, squamous gastric disease is easy to explain: it is a chemical burn. It is a lot more like gastroesophageal reflux disease or heartburn in people.

Glandular disease is much more like gastric ulcers in humans, but it is not the same. It is not caused by *Helicobacter pylori* and nonsteroidal drugs, the two dominant causes in humans. Nonsteroidal drugs might contribute in some cases in horses, but they are not a major cause in terms of the general population. There are lots of other causes of glandular disease in humans, including idiopathic ulcer disease, meaning we simply do not know why the disease occurs.

Likewise, glandular gastric disease in horses is poorly understood. It appears to primarily be inflammatory gastritis, but we do not know the underlying reason why this occurs in certain individuals. This makes it difficult to treat and particularly difficult to prevent.

Underlying causes for squamous disease, as I touched on earlier, include inadequate roughage and excessive carbohydrates in the diet, and the amount and timing of exercise but none of these has a direct role in glandular gastric disease, which appears to be much more related to behavioral stress and systemic-type disease in the sense that we've got evidence that horses with increased stress response are at increased risk of glandular gastric disease. Behavioral elements, such as the number of exercise days per week, are risk factors. Therefore, for glandular disease, the absence of rest days seems to be important for gastric health and other body systems as well.

Can you describe the rating system for evaluating squamous and glandular gastric ulcers?

There is a well-established grading system for squamous disease, which is a 0 to 4 scale, with 0 being normal, 1 being some mucosal thickening, 2 being a small ulcer, and 3 and 4 being larger ulcers. Important here, there is no evidence to say an ulcer with a grade of 4 is twice as bad as a 2 or a 2 is twice as bad as a 1. It seems to be more of a presence or absence situation.

We can have clinically significant and relatively small lesions, and we can have some quite large lesions that do not seem to be clinically important in some animals. Response to treatment is a large component of understanding how much the disease is affecting the individual horse.

We do not have a well-established grading system for glandular gastric disease. Initially, the 0 to 4 system was used, but then we recognized in the glandular lesions that what we saw endoscopically correlated poorly with what was going on in the tissue itself. Our visual impression and the disease process could be quite disconnected. Again, this seems to be a presence or absence dilemma, and we would use more general terms like "mild," "moderate," and "severe," without putting numbers on them. In a research setting, we have no choice but to use numbers, but we tend to use modified grading systems, recognizing grading systems do not validate well and highlight a limitation of current research.

Like squamous disease, we can have quite significant glandular lesions that appear to have no impact on the horse or we can have mild-appearing lesions that appear to be quite clinically significant. Again, response to treatment is important.

How do you assess the clinical significance of lesions?

When we gastroscope a horse and find a lesion, how do we know it is important? Assessing clinical signs is the place to start. Clinical signs can be wide and varied for both squamous and glandular gastric disease.

For squamous, the most frequent clinical signs are changes in appetite, particularly poor appetite, and unexplained weight loss. Unexplained poor performance is often noted, too. We see other clinical signs as well.

When we want to assign significance to those, the good thing about squamous gastric disease is that it responds consistently through acid suppression with omeprazole. We want to see a change in those clinical signs as treatment progresses. That is one of the ways we can assign significance to the disease.

We have more difficulty with glandular gastric disease because the most common clinical presentations are behavioral (changes in behavior and rideability, reluctance to move forward, reluctance to do the work, etc.). What is important with glandular disease in this context is that this appears to be a pain-based response. When we think about pinning ears and kicking when girthing, these are anticipatory pain-based behaviors. What we know is that horses with lameness will do the same thing, and horses with sand enteropathy will do the same thing.

Rather than assigning those behavior changes directly to ulcers, we are taking a step back now and saying, "We believe these are pain-based behaviors, so we have to find the source of pain, one of which may be glandular gastric disease." We must consider other differentials, typically lameness, sand enteropathy, or some other sources of pain, which can be challenging.

Adding another layer of challenge, the response to treatment with glandular gastric disease is less consistent than treatment for squamous gastric disease. When we start glandular gastric ulcer treatment with omeprazole, we normally pair it with other things, but we do not always get such an obvious response as we get with squamous disease. This can make working through treatment challenging. That is where the vet must be involved in the care of the horse, start to finish, and both the owner and the vet must be willing to consider a range of differentials as to why the behavioral presentation is there in the first place and not just be myopically focused on gastric disease.

This seems like a strange thing for someone who has built his research career on gastric ulcers to say—"It is not always the ulcers."—but it is important as an equine clinician to say that it is not always the ulcers when looking at pain-based behavior.

How are the two types treated?

For squamous gastric disease, the standard treatment is omeprazole. It is well-established, and numerous publications support its use. If we get good acid suppression, there is no need to add other things to the treatment regimen for squamous disease. We have a consistently high success rate with omeprazole for squamous gastric disease.

Glandular disease treatment is more complex. We know the response rate with omeprazole is low at 25-50%. If we add sucralfate, we bring those up to maybe 50-75% but there is a wide variation in published response rates when it comes to combinations of omeprazole, sucralfate, and misoprostol. There also seem to be some regional differences in how different populations of horses respond.

Some respond better to one combination, and some respond better to another combination. Again, it is important to have the veterinarian involved in the decision-making right from the start, not only from prescribing medication but also following these patients right along. This is why gastroscopy is so important with these horses.

Gastroscopy tells us whether a lesion is present and whether it is a squamous or glandular lesion. These findings inform treatment decisions. Gastroscopy also tells us what to do next. If the horse responds and the lesion heals, great. But if the lesion heals and behavior problems persist, maybe it was not the lesion after all and we must revert to the differentials.

Gastroscopy also tells us where we need to focus management changes. With glandular disease, we must focus on rest days, environment optimization, and behavioral changes.

With squamous disease, we focus on optimizing roughage intake, the timing of exercise, and figuring out what the horse does overnight in terms of eating behavior. Gastroscopy is an important part of diagnosing the disease but is also important for treatment, prevention, and long-term management.

What's on the horizon for gastric ulcer prevention?

We know that squamous gastric disease is mainly a management-associated disease, so continuing to look at management interventions that make a difference is important. I specifically say that because we have had some standard recommendations for a long time, but we have not accumulated huge amounts of evidence about how well they work in different populations. What we see is that in certain populations management can be very effective for squamous gastric disease, but as we increase the management intensity, we need to think about other strategies. To me, squamous disease is always going to be management first and then something else second.

Glandular disease is a big question now. We do not know a lot about glandular gastric disease. It is important to say that we talk about it as a singular disease, but it is quite likely that we have several diseases with a similar clinical presentation but different underlying causes.

As we learn more and more about glandular disease, we might find different management strategies for different types. We know there is a subset of horses that are refractory to treatment or very slow to respond. We are starting to shift away from long-term pharmaceutical treatment to treat glandular disease and moving back to treating it as a behavioral disease and managing clinical disease. Let us focus less on what the stomach looks like and more on how it is impacting the horse. We are looking into using appropriate behavior management strategies and other methods to improve the horse's pain threshold and to change the expression of the disease as much as we worry about treating the disease itself. In addition to a healthy stomach, we want the horse to be happy on top of that.

Is there anything about your research that might surprise veterinarians or horse owners?

I think the most surprising thing for horse owners may be how hard it is to get funding and support for a disease that is so common and important to horse health. It is a difficult process to get relatively small amounts of money, even though this research is very important to horses and horse owners.

Further, it might be surprising how little money is needed in some cases. For $10,000 or $20,000, we can go a long way in answering some specific questions. But it can be hard to even get that much money. That's probably the biggest challenge as a researcher. I can always find the time, but having a consistent supply of funding is not always readily available so that research can be carried out logically and sequentially. I do not think many people understand what a barrier that can be.

What do you want people to know about EGUS when they leave your presentation? Give two or three important take-home messages.

EGUS is not a single disease. We need to talk about squamous gastric disease and glandular gastric disease as separate entities. What causes squamous disease does not cause glandular disease. I think this is important because we have a lot of well-meaning owners who follow the prevention protocol for squamous disease, even though half of the equation is missing because glandular disease is completely different.

When we think about squamous disease, we should focus on the three big points: (1) ensuring adequate roughage in the diet, (2) ensuring that excessive carbohydrates are not in the diet, and (3) exercise, both in terms of quantity and in terms of sorting out the timing of exercise, ideally in the afternoon when horses have had access to roughage all day, and considering carefully the time of feeding before exercise, especially when feeding alfalfa to maximize buffering capacity.

Recognize that glandular disease is different than squamous gastric disease, focusing on aspects that are different from squamous disease. From a management perspective, this means ensuring adequate rest days and environmental optimization for that horse, such as thinking about cohabitation with other horses and allowing horses to interact with one another. Consider glandular gastric disease as a medical disease and a behavioral disease, including pain-intervention strategies and other therapies to improve the horse's overall well-being.

Can you recommend some seminal references (articles, book chapters, books, etc.) on EGUS written by you or someone you admire in the field?

1. Sykes, B.W., M. Hewetson, R.S. Hepburn, N. Luthersson, and Y. Tamzali. 2015. European College of Equine Internal Medicine Consensus Statement—Equine gastric ulcer syndrome in adult horses. Journal of Veterinary Internal Medicine 29(5)1288-1289.
2. Vokes, J., A. Lovett, and B. Sykes. 2023. Equine gastric ulcer syndrome: An update on current knowledge. Animals 13(7):1261.

Imaging Technologies for Diagnosing Lameness

Robin Bell, BVSc, MVSc, DipVetClinStud, DECVS, DACVSMR

Can you provide a bit of background? How did you come to specialize in lameness?

I grew up riding horses and seeing close friends have their Olympic dreams dashed when their "horse of a lifetime" was injured. This inspired me to try to help these amazing athletes reach and maintain their potential. The university where I went to vet school and did my internship had a high caseload of sport horses, as well as Thoroughbred and Standardbred racehorses, so we saw a lot of lame horses, and the diagnosis and management of these piqued my interest. I have always been passionate about sport horses, and was fortunate to work at the Sydney Olympics in the hospital on-site. Since then, I have been privileged to be an Australian team veterinarian for the showjumpers at the WEG in 2014 and 2018, and for dressage and showjumping at Herning in 2022. I was showjumping and dressage veterinarian at the Rio and Tokyo Olympics.

Can you give us a brief timeline of how lameness diagnosis has evolved over the last 50 years?

The advances in imaging technology across the board have revolutionized how we diagnose lameness in the horse. Take radiography, for example; it has progressed from old-school film to extremely fine-detail digital radiography plates, which give beautiful, high-quality images. Stretching that example a little, we can include the multiple-slice helical CT scanners, which when taken in an overly simplistic way, are cross-sectional radiographs. When I first worked with CT, the multiplanar reformats were not diagnostic as the volumes they were taken from had so few slices (4-slice scanners). Nowadays, with upwards of 150 slices, these multiplanar reformats, and indeed to some extent three-dimensional reformats, provide us valuable diagnostic information.

Can you provide us with a description of current diagnostic tests?

The basics remain plain radiography and ultrasound.

- Radiography uses X-rays to create an image.
- Ultrasound creates an image using reflected sound waves.
- Magnetic resonance imaging (MRI) uses the signal given off hydrogen ions after applying a radiofrequency pulse through the region of interest. MRI scanners can be high-field under general anesthesia and low-field under both standing and general anesthesia. Contrast studies can be performed.
- Nuclear scintigraphy uses Tc99 bound to various tracer agents most commonly MDP or HDP to primarily image bones and joints.
- Positron emission tomography (PET) uses different tracer agents and isotopes; it allows for images to be obtained in 3D and has better soft tissue sensitivity. The half-life of the isotopes is shorter, allowing for patients to be discharged the same day.
- Computed tomography uses an X-ray source that rotates around the patient to send X-rays to a detector on the opposite side of the gantry. These are compiled and a slice is formed from the combined X-rays. The acquisition of two different energy X-rays of the same slice allows for the differentiation of tissues with similar X-ray attenuation called dual-energy CT is also possible in some scanners. These CTs can be acquired both under general anesthesia and standing.

How do you determine what diagnostic tests are performed on a given lameness? Use examples, if you like.

How I approach each case is governed by the horse's use: Is it a high-performance athlete, or a pasture ornament?

I take into consideration its competition or racing schedule, the severity and duration of the lameness as well as the owner or trainer's goals. Some owners are happy to try a treatment trial; others want to know exactly what is causing the problem.

What have current diagnostic tests taught veterinarians about common injuries?

I think that the most exciting developments have come in the early detection and diagnosis of subchondral bone injuries in racing Thoroughbreds. Rather than chalking all these catastrophic injuries as freak accidents or inherent in the risks of racing, we can identify horses at risk of these often-fatal injuries prior to them occurring.

What is coming in the world of diagnostic tests?

The most exciting thing that we are working on is dual-energy CT, which I think will be a real game changer in the diagnosis of subtle subchondral bone injuries.

How does client budget affect diagnostics?

If clients are money-limited, I typically will use the more basic (and less expensive) imaging or start with treatment trials.

What frustrations have you endured over the years as you have studied lameness?

As outlined in my talk, there is a plethora of imaging equipment available to us as equine veterinarians; however, sometimes I think we are too reliant on these modalities. They are not "answer machines," and there is no substitute for a thorough clinical examination.

As equine veterinarians, we tend to be at the forefront in terms of trialing new products and treatments for clinical conditions affecting the horse, often using these treatments before there is good scientific evidence that they work. As a profession, we are too quick to believe the hype from the companies selling these products and may cave to pressure from owners who are desperate to try anything to fix their horse.

What factors continue to be a hindrance in diagnosing and treating horses with lameness?

We still lack a good model that mimics common clinical conditions, such as tendonitis, which hampers research into treatment for these conditions. Even our models for osteoarthritis do not mimic milder disease.

What do you want veterinarians and owners to know about diagnosing lameness when they leave this presentation?

There is not one modality that is suitable to diagnose all clinical conditions and regions. To get an optimal diagnosis, you might have to use a combination of two or more modalities.

What might surprise them? In my opinion, the soft tissue phase of nuclear scintigraphy is rarely diagnostic for commonly seen lameness conditions.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on lameness written by you or someone you admire in the field?

1. The new edition of *Equine Sports Medicine and Surgery* (Hinchcliff, Kaneps, Geor, and Van Erck-Westergren, published by Elsevier) which will come out this March is very exciting.
2. Aside from that, I use the most recent version of Ross and Dyson's *Diagnosis and Management of Lameness in the Horse* (published by Elsevier) and the most recent edition of Auer and Stick's *Equine Surgery* (published by Elsevier).
3. The work of Schumacher et al. on the structures desensitized by the various nerve blocks changed my practice, and in terms of the distal extremity is summarized well in this review article: Schumacher, J., M.C. Schramme, J. Schumacher, and F.J. DeGraves. 2013. Diagnostic analgesia of the equine digit. Equine Veterinary Education 25(8):408–421.

In terms of a single paper that I always come back to, I will mention one by Chris O'Sullivan and Jonathan Lumsden because it is so well-written and outlines their approach to managing stress fractures.

Here is the full reference: O'Sullivan, C.B, and J.M. Lumsden. 2003. Stress fractures of the tibia and humerus in Thoroughbred racehorses: 99 cases (1992–2000). Journal of the American Veterinary Medical Association 222(4):491–498 doi:10.2460/javma.2003.222.491.

Role of Inflammation in Adaptations to Exercise Stress

Wendy Pearson, MS, PhD

Can you provide a bit of background? How did you come to be interested in exercise and inflammation?

I was first introduced to research in inflammation in 1997 when I returned to Canada from New Zealand, freshly graduated from my undergraduate degree in animal science.

Whilst working at Woodbine Racetrack as an exercise rider, I accidentally stumbled upon a six-month contract position at the Equine Research Centre in Guelph, Ontario, to conduct an experiment evaluating the effects of an herbal supplement on equine arthritis. My knowledge in this area was woefully deficient, so I started reading—a lot! It became evident, both through my reading and my experiences at the racetrack, that administering anti-inflammatory substances to exercising horses was common practice. There seemed to be a universal acceptance that inflammation was an inevitable consequence of exercise, and must be avoided at all costs!

Even back then, this seemed illogical to me. If exercise stimulates inflammation, and the more you exercise the more easily your body can tolerate exercise bouts, surely there must be a connection between inflammation and adaptation of tissues to exercise. Thus, my insatiable curiosity about the relationship between exercise-induced inflammation and the training effect.

What is inflammation? What causes it?

Inflammation is a normal, transient immune response to trauma or infection. Its purpose is to rid the body of pathogens and/or damaged tissue and stimulate the growth of new healthy tissue. During infectious inflammation, this is achieved by creating an environment that is hostile to the propagation of the offending organism.

In general, the key features of inflammation include:

1. Production of systemic "fever" and/or local heat, the latter of which increases local blood flow, and both of which inhibit enzymatic reactions that are critical to the viability of pathogens;
2. An increase in local tissue swelling, which reduces oxygen saturation, activates mechanotransduction pathways to pain transmission, and increases the local provision of anabolic (healing) compounds; and
3. Production of cytotoxic compounds (i.e., cytokines) that destroy pathogens and coordinate critical cell-to-cell signaling.

This cell signaling is initially in service to the upregulation of inflammation, by increasing the production of pro-inflammatory catabolic compounds, and then later to the calming and eventual shutdown of inflammation by increasing the production of anti-inflammatory, anabolic compounds. Inflammation can be triggered by the presence of a non-self-antigen, such as an infectious pathogen or a noninfectious allergen, or by release of pro-inflammatory molecules from damaged self-cells, which occurs during tissue injury or during exercise.

How is inflammation measured in horses?

Clinical signs of inflammation in horses will vary depending on the cause. Inflammation resulting from intense exercise can often be measured as a transient increase in core body temperature as well as increases in local temperature of areas such as muscles, joints, and tendons.

There is a huge battery of systemic biomarkers that can also be quantified in body fluids such as blood and synovial fluid, including those that orchestrate the perception and/or tolerance of pain [e.g., prostaglandin E2 (PGE2), nitric oxide (NO), and complement cascade fragments], those which facilitate tissue breakdown (e.g., metalloproteinases and aggrecanases) and synthesis (e.g., collagen synthesis neoepitopes and CS846), as well as those which sensitively indicate general (e.g., serum amyloid A) or specific (e.g., creatine kinase from muscle or C2C from type II collagen) inflammation.

There is also a host of biomarkers for oxidative stress (e.g., reactive oxygen species [ROS] and endogenous antioxidants such as glutathione) that can be analyzed in body fluids.

Exercise-induced inflammation in joints can also be measured in horses. Use of a flexible tape to measure the joint circumference at specific anatomical landmarks around a joint can provide an accessible (if somewhat crude) measure of joint effusion. Synovial fluid samples can be obtained directly from the joint, and these can be subjected to similar biomarker analysis as described above.

When is inflammation beneficial and when is it damaging?

This is really the million-dollar question.

Because inflammation tends to be associated—rightly or wrongly—with how uncomfortable it makes us (and our horses) feel, it is tempting to treat it as something inherently "bad" that should be avoided whenever and wherever possible.

However, the transient inflammation that is realized from exercise is key to the many benefits that are well known to result from regular exercise. For example, a cytokine called interleukin (IL)-6 is a pro-inflammatory compound that is consistently upregulated following high-intensity exercise bouts. In classic inflammation, IL-6 is a major mediator of the acute phase response, promoting the release of acute phase proteins (such as C-reactive protein) from the liver. IL-6 is also associated with the induction of fever, and amplification of pro-inflammatory cytokines and the associated increase in tissue damage. There is also evidence that IL-6 is involved in the pathogenesis of neuropathic pain. In general, a consequence of persistently elevated IL-6 is frequent tissue destruction and a net negative impact on host health.

However, when IL-6 is transiently elevated (as seen in high-intensity exercise) it participates actively in the training effect. Its varied roles in this context include an upregulation of osteocalcin, thereby contributing to exercise-induced improvement in bone mineral density and an increased tolerance to very high-intensity exercise. It also promotes the uptake of glucose and free fatty acids by muscle cells, thereby promoting increased muscle mass. Similarly, transient exercise-induced increase in PGE2 is necessary for muscle protein turnover (especially in Type I muscle fibres), and ROS activate protein kinases and gene transcription that results in muscle adaptation to exercise. The evidence suggests that, in most cases, transient inflammation can (and perhaps should) be tolerated without interference, as it plays a crucial role in the adaptation of tissue to stress. Persistent inflammation, on the other hand, is more frequently associated with tissue destruction and disease and in many cases should be controlled.

When do you know when to treat inflammation or when to leave it alone?

Transient, healthy inflammation in joints resulting from exercise typically peaks in blood within the first hour after exercise cessation (depending on the outcome measure used), and in joints approximately eight hours after exercise cessation. This inflammation is typically not associated with clinical signs of pain or dysfunction and in most cases should be allowed to proceed and self-regulate. There are occasions, however, when the exercise bout results in injury.

In the cases of injury to joints, this might be observed as an unusual increase in swelling, marked local heat, and/or lameness. These are not expected outcomes of normal exercise-induced inflammation, and it is often important to limit the extent of the inflammatory response to curb its negative impact on the tissue.

What are the different treatment modalities used to reduce inflammation?

During the early stages of inflammation, the primary goal is to limit the magnitude to which the inflammation is allowed to progress. Serious injuries, including those that put either the horse or the handler at acute risk, should only be addressed by a veterinary practitioner. For less severe injuries, early application of cold (e.g., ice boots, ice blankets, cold water, etc.) is indicated to reduce aberrant fever and/or local heat. This will help minimize acute destruction of tissues.

Nonsteroidal anti-inflammatory drugs such as phenylbutazone can be useful in the acute stages of inflammation to reduce the production of PGE2. Your veterinarian may also prescribe corticosteroids (such as prednisone) or selective COX-2 inhibitors (such as Previcox) to manage longer-term pain and inflammation. Some nutraceutical products can be effective options to manage chronic inflammation and degenerative inflammatory conditions.

How important are omega-3 fatty acids in the management of exercise-induced inflammation?

There is a dearth of research available to help us answer this question, but including omega-3 fatty acids in the diets of athletic horses is widely and enthusiastically practiced.

Unlike their omega-6 counterparts, which are metabolized mainly to pro-inflammatory prostaglandins, leukotrienes, and thromboxanes, omega-3 fatty acids metabolize to less inflammatory species. Thus, if the goal is to reduce inflammation, decreasing a horse's omega-6 to omega-3 ratio may be a sound strategy. While the literature makes a strong case that inflammation is needed for tissue adaptation, I do not think anyone could sustain a viable argument that a lot of inflammation is required.

And here we bump into our knowledge gap: if we assume that increasing omega-3s dampens the inflammatory response (we have some evidence for this, but we need a lot more equine research before we can state it as fact), does it still allow sufficient inflammation to stimulate tissue adaptation whilst limiting undesirable, pathological inflammation? Or, do omega-3 metabolites have different (perhaps better?) effects on tissue adaptation than metabolites of omega-6 fatty acids? We are in great need of more research in this area.

You have an interest in nutraceuticals. How have you incorporated that into your work with inflammation?

Nutraceuticals represent my first love in science, and they still form the core competency of my research program. As a physiologist, most of my attention is on mostly healthy animals (as opposed to sick animals), usually to prolong health and/or improve performance.

This is, in my opinion, the best and most fruitful application of nutraceuticals. Horses can live healthy and productive for upwards of 25 years, and mild inflammation is an inevitable companion on their way to old age. Thus, my research program seeks to increase our understanding of the multifaceted ways in which inflammation interferes with robust health, and how we can intelligently apply nutraceuticals to mitigate its effects.

What factors continue to be a hindrance in understanding exercise-induced inflammation?

Like most topics in science, exercise-induced inflammation research can only progress as quickly as funding bodies are willing to invest in it. Funding has always been, and continues to be, the biggest challenge to moving the needle on what we think we know about the role of inflammation in the training effect.

What do you want veterinarians and owners to know about inflammation when they leave this presentation? What might surprise them?

I want them to come away from this presentation contemplating that inflammation, in some contexts, is actually a good thing.

Whilst persistent, clinical inflammation is undesirable and should be managed, the transient inflammation associated with exercise is paramount to the adaptation of tissues and the training effect.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on exercise stress or inflammation written by you or someone you admire in the field?

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Influence of Nutrition in Leaky Gut Syndrome

Wendy Pearson, MS, PhD

What is leaky gut syndrome?

Leaky gut syndrome (LGS) is a general term to describe various clinical consequences of increased permeability of the lining of the gastrointestinal tract (GIT). The GIT lining functions as a selectively permeable barrier that is essential to proper sequestering of luminal contents, which includes a complex and dynamic microbiome.

It protects the systemic circulation from potentially toxic compounds (e.g., bacterial endotoxins) which, if allowed to leave the GIT and move into the blood, can trigger low-grade, chronic inflammation. Barrier function is regulated by tight junctions, comprised of more than 40 proteins (including claudin, occludin, and zonulin) that regulate paracellular trafficking of GIT contents.

While we have almost no research in horses, there is evidence in nonequine species that compromised function of tight junction proteins allows leakage of luminal contents.

How is it diagnosed in horses and other species?

LGS is not a current medical diagnosis in horses (or in humans), and there is no definitive test for it. It is sometimes presumptively diagnosed based on the presence of risk factors and inflammation-related disorders, when other potential causative factors have been ruled out.

What is its prevalence in horses?

There are no published studies that provide insight into the prevalence of LGS in horses. Our current knowledge on its prevalence in horses is woefully deficient.

How is it diagnosed in horses and other species?

There are only two equine-specific primary research papers in the scientific literature describing leaky gut in horses, and neither attempts to reveal how the condition is detrimental to horses. We must, therefore, lean on nonequine research to help us weigh the risk of LGS in horses.

Studies in humans and small rodents have implicated LGS in such outcomes as impaired skeletal muscle and energy metabolism, reduced athletic performance, allergic respiratory, and chronic inflammation. Whether these conditions can be attributed to LGS in our athletic horses must be explored in future research.

How do you study leaky gut syndrome in horses?

Research into LGS in horses is in its infancy. In our lab, we are starting at the beginning by asking this: "Does physiological stress result in an increase in GIT permeability?" We have generated evidence for this by orally administering a marker of GIT permeability (iohexol) and then measuring the appearance of this marker in blood. The bioavailability of iohexol in horses with normal GIT permeability is essentially 0%; thus, when it appears in blood following stress (we used a combined stress of trailer transport and moderate-intensity exercise), this provides evidence for a stress-associated increase in GIT permeability.

Concurrent with an increased appearance of iohexol, this stressor also produced an increased appearance in blood of bacterial endotoxin (lipopolysaccharide) and the acute phase protein serum amyloid A. These data support the hypothesis that the combined physiological stressor of trailer transport and exercise increases GIT permeability, and also produces an increase in markers of inflammation. However, this does not, on its own, suggest that combined transport and exercise results in LGS because the hyperpermeability we observed was not associated with any concurrent clinical signs of inflammation. It does provide evidence that this type of stressor has the potential to increase GIT permeability, which may contribute to the development of LGS in horses.

Has your knowledge of nutraceuticals helped in creating potential treatments for leaky gut syndrome?

This is an area of study that is virtually nonexistent. However, we can develop hypotheses based on research in nonequine species. Studies in humans, rodents, and large ruminants provide evidence that the GIT microbiome is a key modulator of tight junction proteins. Support of a healthy microbiome may therefore protect tight junction proteins from the negative effects of stress and limit development of LGS in horses.

We have previously explored the effect of a dietary prebiotic (fermentation product of *Aspergillus oryzae*) on stress-induced GIT hyperpermeability in horses. Feeding of this supplement for 28 days before combined trailer transport and exercise stress eliminated the stress-induced hyperpermeability seen in unsupplemented control horses. To my knowledge, this is the first study to quantify the effect of any nutraceutical on GIT hyperpermeability in horses.

What speedbumps have you encountered when studying leaky gut syndrome?

Research into LGS in horses is in its infancy, which presents many challenges for researchers interested in studying it. The lack of clinical or scientific understanding about its importance in equine health and performance makes it challenging to present a solid case to funding bodies.

We must rely on research in nonequine species that, while often at a more advanced stage than equine research, is also replete with more questions than answers. This presents a huge challenge to advancing science in this area.

We need much more research on LGS in horses to better understand its clinical consequences, its causes, and its treatment.

What is on the horizon as far as LGS?

We know little about the clinical implications of GIT hyperpermeability, and we know even less about possible contributing factors such as variables as diet, fitness level, breed, age, type and intensity of exercise, and other physiological stressors. What we do know is that the physiological stress of trailer transport and exercise increases GIT permeability, and that increase can be prevented by feeding a prebiotic fermentation product of *Aspergillus oryzae*.

Future research will provide new insight into the importance of LGS in the health and performance of our horses.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on equine leaky gut syndrome written by you or someone you admire in the field?

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The Mighty Mitochondria

Mike Davis, DVM, PhD, DACVIM

What are mitochondria? What do they do?

Mitochondria are components of most mammalian cells (mitochondria is plural, the singular term is mitochondrion). They are organelles, which are little ecosystems within the cell, bound by membranes and containing specific enzymes, transporters, and chemicals that perform specific functions in support of the cell.

There are many different organelles in a typical cell; the most well-known would be the nucleus of the cell. In the case of mitochondria, their "job" is to break down small molecules such as fragments of glucose or fatty acids, and convert the energy in the bonds of those molecules into a molecule called adenosine triphosphate or ATP. The rest of the cell uses ATP to power pretty much everything else the cell does, which is why mitochondria are commonly known as the "powerhouses" of the cell.

How do you study mitochondria? Does the study differ between species?

There are lots of different ways to study mitochondria, and we are getting better at it. For a very long time, the most common way of studying mitochondria was to simply count them based on quantifying the abundance of key enzymes like citrate synthase. That is still very common because it is quick and relatively easy. Because the mitochondria are where oxygen ultimately gets burned during metabolism, quantifying oxygen consumption of tissue or cell samples (or partially ground-up cells) was the most common way of quantifying their function. Just about all living organisms have mitochondria to provide the organism with a readily and easily usable form of energy, so the basics of studying mitochondria are common between species. However, as the specific assays become more sophisticated, some differences may start to emerge.

Can you explain mitochondrial respirometry?

Respirometry is the study of the consumption of oxygen and production of carbon dioxide. This can be done on a whole animal or any part or subunit, all the way down to mitochondria isolated from cells, which would then be called mitochondrial respirometry. The specific equipment may differ based on the scale of the sample, but the basic premise is the same.

You have done a lot of comparative work with dogs and horses. What are the differences in the adaptability of mitochondria? At what point does mitochondria become an issue for energy generation?

Different tissues will have different amounts of mitochondria based on the amount of work they may be expected to do. Similarly, the skeletal muscle of different species will differ based on the aerobic capacity of the skeletal muscle.

While horses are one of the most impressive aerobic species in the world of domestic animals, an athletic dog can have double the aerobic capacity of a horse on a pound-for-pound basis. Whether it is a horse or a dog, that magnitude of athletic capacity does not happen without an ability to respond to conditioning, and horses and dogs are very similar in that regard. We have not fully explored the adaptability of mitochondria in either species, so comparisons are difficult and speculative at this point. When viewed as simply being the cell's source of ATP, the relationship between mitochondria and energy generation/utilization is straightforward: more is better. You can only burn ATP as fast as you can produce it, and the more mitochondria you have, the faster you can produce ATP.

However, dogs are a good example of the limit of that argument. In the process of producing ATP, the chemical reactions also produce a lot of heat, so more mitochondria mean more heat that needs to be dissipated. A recent study that we just published showed that as the mitochondria become hotter, they start producing more reactive oxygen species and generating more oxidative stress that can damage not only muscle but potentially other tissue.

Has your work in dogs influenced your study of horses? Or vice versa?

The dog work and horse work have complemented each other and fed each other. Some studies are easier in dogs, some are easier in horses. Mostly, we are trending towards horses currently because the specific assays require larger muscle biopsies that are much easier to obtain from horses than dogs. But the dogs, with their world-record levels of skeletal muscle mitochondria, got us rolling.

How do you affect mitochondrial function?

As previously mentioned, an effective conditioning program will increase the functional abundance of mitochondria. I use "functional abundance" to highlight the critical difference between enumeration through the analysis of selected enzymes versus enumeration through complete functional assays such as respirometry.

Increasing the abundance of a single enzyme does little good if the rest of the cascade is not similarly increased, so there have been some wrong turns over the years in the study of mitochondria. Because mitochondria are just bags of enzymes and transporters, the physiochemical conditions can affect the function of those proteins just like any other protein. That has been the focus of our studies: how mitochondria maintain function (or do they at all) in the face of the high temperatures and low pH that we know develops in the muscle during exercise.

Our recent studies have demonstrated that high temperature is particularly deleterious to mitochondrial function, with rates of ATP synthesis dropping severely when the muscle heats up. Our research strongly suggests that high temperature is the most potent cause of skeletal muscle fatigue due to loss of capacity for producing ATP.

Can you explain why horse owners and veterinarians would be interested in this research?

We are getting down to the nitty-gritty of what causes fatigue during exercise, which is of interest to anyone who hopes their horse fatigues last in the race. So far, we've found minimal evidence that conditioning might improve mitochondrial resistance to the effects of hyperthermia (even though it clearly increases the functional abundance of mitochondria), so this research may justify greater attention to thermoregulation to preserve exercise capacity.

What are you trying to learn through this research?

I am trying to nail down exactly what causes a horse to fatigue – to slow down (as more than half of the horses in any given race are doing at the end of the race). The winner is not the fastest horse, it is the horse that slows down the least so, in a way, I am trying to identify this contribution to what makes a horse a winner.

Are there specific challenges with this research that might be interesting to other researchers or veterinarians?

Unfortunately, the research can be extremely time-consuming and tedious. We conducted a study during the summer of 2021 in which we directly measured the rates of mitochondrial production of ATP, which was a first. I finished analyzing the files produced by the respirometers last week. Tedious, but worthwhile. What it does mean is that you do not want to have to repeat things, so very careful study design and execution becomes more and more important. But the results are worth it.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on this topic written by you or someone you admire in the field?

With all modesty, I think the paper that was published recently is one of the more important ones in the field: Effects of hyperthermia and acidosis on mitochondrial production of reactive oxygen species by M.S. Davis, W.M. Bayly, C.M. Hansen, M.R. Barrett, and C.A. Blake. American Journal of Physiology-Regulatory, Integrative and Comparative Physiology 2023:325(6):R725-R734. doi: 10.1152/ajpregu.00177.2023.

Additional publications that have illustrated the value of high-resolution respirometry include:

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Moreover, the inventor of the machines that we use (Erich Gnaiger, president of Oroboros Instruments, Innsbruck, Austria) produces an excellent manual that marries basic mitochondrial physiology with the operation of the machines and design of assays: Gnaiger, E. 2020. Mitochondrial pathways and respiratory control: An introduction to OXPHOS analysis. 5th ed. Bioenergetics Communication 2. https://doi.org/10.26124/bec:2020-0002.

Cutting-Edge Technologies for Studying Muscle Metabolism

Stephanie Valberg, DVM, PhD, DACVIM, DACVSMR

A continual supply of adenosine triphosphate (ATP) is the fundamental cellular process that underpins skeletal muscle contraction during exercise, and it is essential for equine performance in events lasting minutes to many hours.

Metabolic pathways must be activated to maintain the required rates of ATP resynthesis because the muscle stores of ATP only supply fuel for contraction for seconds. These metabolic pathways include anaerobic pathways of phosphocreatine and muscle glycogen breakdown and aerobic pathways that supply ATP through the metabolism of sugar, glycogen, and fatty acids via oxidative phosphorylation.

The relative contribution of these metabolic pathways to energy metabolism during exercise is primarily determined by the intensity and duration of exercise. For most events, glycogen is the primary fuel for anaerobic and aerobic metabolism with blood glucose and fatty acids becoming more important as the exercise duration is prolonged. Understanding the role of each metabolic pathway in fueling specific types of exercise in horses helps to optimize their performance through nutrition and training.

How has muscle study changed in the past 20 years?

Muscle metabolism was initially studied in horses by obtaining blood samples before and after exercise. This gave a very limited view of muscle metabolism. With the advent of high-speed treadmills, blood samples could be obtained during exercise with measurement of blood lactate concentrations used to assess anaerobic metabolism.

Oxygen uptake studies later allowed the assessment of whole-body aerobic and anaerobic metabolism during exercise and the relative role of carbohydrates and fats in fueling skeletal muscle during treadmill exercise. The percutaneous needle muscle biopsy technique perfected by Arne Lindholm in Sweden and David Snow in the U.K. opened a window to the world of muscle metabolism.

Using muscle tissue snap-frozen in liquid nitrogen, changes in muscle glycogen, ATP, lipid, and lactate concentrations were determined at varying speeds, exercise durations, and training levels. The information derived from these studies of muscle metabolism in healthy horses was integral to subsequent studies of muscle metabolism in horses with exertional rhabdomyolysis due to various causes.

Relative to human muscle, equine muscle has remarkably high glycogen concentrations, high oxidative and glycolytic capacities, and a remarkable ability to produce and withstand high lactate and low ATP concentrations with intense exercise.

In contrast to humans, resting glycogen concentrations range from 450 to 650 mmol/kg dry weight in horses (human, 300 to 400 mmol/kg), with the highest concentrations found after long-term training. A unique aspect of equine muscle metabolism is that muscle glycogen concentrations in healthy horses are resistant to carbohydrate loading and slow to replete after exercise. In humans, glucose is taken up into skeletal muscle cells through glucose transporter GLUT4 under the influence of insulin.

The reason why glycogen repletion is so slow in horses has remained a conundrum, and traditional biochemical and immunohistochemical studies have failed to explain why GLUT4, so important in human skeletal muscle, is not activated by insulin to rapidly restore muscle glycogen levels.

Where do genomics, proteomics, and glucose transporters fit into this?

"Omic" technologies allow us to obtain a comprehensive understanding of biological systems in muscle that was not previously possible.

Examples of "omics" include proteomics, transcriptomics, genomics, metabolomics, lipidomics, and epigenomics, which correspond to global analyses of proteins, RNA, genes, metabolites, lipids, and methylated DNA or modified histone proteins in chromosomes, respectively. To date, proteomics, transcriptomics, genomics, and metabolomics have been used by a small number of researchers to study equine skeletal muscle in health and disease. The results of these studies have helped us to capture the myriad of events that can combine to cause muscle dysfunction in horses and recently to study limitations in glycogen repletion in horses.

Using mRNA measurements, Valberg, Pagan, and colleagues studied the gene expression of all known glucose transporters in the skeletal muscle of Thoroughbreds. We did this before and after three days of intense exercise and for 72 hours after recovery. In doing so, one group of horses received a ration high in starch (HS) and another group received a ration low in starch and high in fat (LS-HF). The study showed that the recovery of muscle glycogen stores was incomplete after 72 hours.

Moreover, recovery was lower for the LS-HF ration than for the HS ration. The exciting finding in this study was that horses express nine different glucose transporters in their muscle. That is, the body had copied the genetic code of nine different transporters to make the corresponding proteins.

Surprisingly, training or recovery did not significantly affect the expression of GLUT1 and GLUT4, as we see in humans. Rather, the expression of GLUT6 and GLUT10 was significantly increased after 24 hours for the HS diet and after 72 hours for the LS-HF diet.

Thus, it seems that different glucose transporters are important in horses compared to humans. The next step in this research is to follow up and confirm that the mRNA is being translated to form specific glucose transporters such as GLUT6 and GLUT10 in skeletal muscle. The transcriptomic results led to a new hypothesis that GLUT6 and GLUT10 may play an important role in replenishing skeletal muscle glycogen stores in horses and that the lack of GLUT4 expression may contribute to slow glycogen recovery even in horses fed insulin-stimulating high-starch diets.

The study of skeletal muscle is complicated by the fact that muscle fibers are not all the same and can be classified into type I, IIa, and IIx subtypes. These different fiber types are active at different points during exercise and have very different contractile and metabolic characteristics. New technologies allow for studies of single fibers that may shed more light on the nuances of muscle metabolism in horses. We recently applied a single-cell approach using spatial transcriptomic studies to isolate the region of Purkinje cells that are dysfunctional in horses with shivers. Within the cerebellum, we were able to precisely localize the disorder to the axons of Purkinje cells rather than the body of these cells. This shows the power of the spatial transcriptomic approaches.

How does this work help veterinarians and owners manage their horses?

"Omic" technologies provide the opportunity to integrate studies of exercise adaptations in the whole horse with the high-precision assessment of skeletal muscle fiber function. Further, they offer the possibility to decipher the regulation of skeletal muscle and harness this knowledge for the development of nutritional and training strategies as well as develop therapeutics to optimize performance and alleviate disease.

By integrating proteomic and transcriptomic studies of horses with recurrent exertional rhabdomyolysis (RER), we were able to determine that abnormal intracellular calcium regulation is fundamental to the underlying basis for this disease.

This showed that medications such as dantrolene, which slow calcium release from the sarcoplasmic reticulum storage site in muscle, should be highly effective in managing RER. A key role of epinephrine was highlighted in these studies that indicated why excitement has the potential to trigger excessive calcium release from the sarcoplasmic reticulum and trigger rhabdomyolysis in RER-susceptible horses.

We also learned that Standardbreds with RER have higher signals of oxidative stress and inflammation than Thoroughbreds with RER. Based on this, we now recommend supplementation with ReSolvin EQ™ to decrease inflammatory signals in the muscle of Standardbreds with RER.

Further, the proteomic and transcriptomic studies of horses with myofibrillar myopathy (MFM) led to the development of MFM Pellet™, which significantly decreases muscle pain and exercise intolerance in these horses.

What do you want veterinarians and owners to know about these new technologies after the presentation? What might surprise them?

"Omic" technologies are the wave of the future in research and are already being applied to develop new approaches to treat muscle diseases.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on myopathies written by you or someone you admire in the field?

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Omega-3 and Omega-6 Fatty Acids in Equine Nutrition

Joe Pagan, MS, PhD

Can you provide some background for feeding fat to horses?

For the past 40 years, we have been studying fat as an energy source for horses, and fat is now universally accepted as a useful and essential energy source, especially for performance horses. For most of that time, we added fat simply to fuel muscle contraction. In that instance, fat is broken up into small, two-carbon bits that when mixed with oxygen in the mitochondria are used to produce ATP. We looked at fat as firewood—chop it up and burn it!

If we look at fat from that perspective, the type of fat does not matter. We have since learned the type of fat and specifically the polyunsaturated fats play a lot of other roles besides just being firewood. When we discuss fat in a feed, we usually describe it as crude fat, which is mostly in the form of triglycerides, which are a glycerol molecule attached to three fatty acids. These fatty acids can be saturated, monounsaturated, or polyunsaturated.

Explain the differences in those fatty acids.

Saturated fat is a string of carbons connected to hydrogens with a hydroxyl group on one end and a methyl group on the other. There are no double bonds between the carbons in saturated fat.

Adding double bonds changes the names of the fat. Add a single double bond to the string, and it becomes a monounsaturated fat; add two or more bonds and it becomes a polyunsaturated fat. If we look specifically at polyunsaturated fatty acids, the two we are most interested in are omega-3 and omega-6. The placement of the double bond determines how a polyunsaturated fatty acid is characterized. If the first double bond is after the third carbon from the methyl end, it is an omega-3. Conversely, if the first double bond is after the sixth carbon from the methyl end, it is an omega-6.

You can further divide polyunsaturated fatty acids by their chain length, i.e., how many carbons are in the fat. Short-chain polyunsaturated fatty acids contain 18 or fewer carbons, while long-chain polyunsaturated fatty acids have 20 or more carbons. Most of the polyunsaturated fatty acids that we put into horse feed are derived from plant sources and are short-chain fatty acids. On the omega-3 side, that is alpha-linolenic acid (ALA), and on the omega-6 side, it is linoleic acid (LA).

Horses cannot produce these fatty acids in their body, so they are considered essential fats. Omega-3s and omega-6s can be used as firewood, i.e., they can be burned for energy production, but what we learned is that they can also be converted into long-chain fatty acids. That conversion is controlled by enzymes, specifically desaturases that add double bonds and elongases that add carbons. These long-chain polyunsaturated fatty acids have several interesting metabolic functions that reach far beyond being an energy source.

One of the main functions of these long-chain polyunsaturated fatty acids is that they can be converted to secondary messengers or lipid mediators that have a lot of physiologic functions in the horse after they have been incorporated into and then released from cell membranes. Much of this function involves the regulation and resolution of inflammation.

Each of the long-chain fatty acids produces lipid mediators that have different functionality. Typically, the omega-3 long-chain polyunsaturated fatty acids produce lipid mediators that tend to reduce or resolve inflammation, whereas the omega-6 long-chain polyunsaturated fatty acids are inclined to produce inflammatory lipid mediators.

If we look at a horse's natural forage-based diet, it tends to be higher in short-chain omega-3 fats and lower in short-chain omega-6 fats. We've looked at the composition of pastures on our research farms in Florida and Kentucky, and we found the concentrations of omega-3 and omega-6 were often similar.

The amount of omega-3 in these pastures tended to be three times the amount of omega-6. If a horse is strictly consuming pasture, it's going to be consuming three times as much omega-3 as omega-6. When we looked at other forages besides pasture (hays, haylage, pelleted forage), we found quite a large difference in the amounts of omega-3 and omega-6.

Well-preserved timothy hay had the highest concentration but only about half of what was in pasture. Other conserved forages had lower concentrations of omega-3 and omega-6 fats. Given this, it looked as though the natural environment skewed toward providing more omega-3s than omega-6s.

How did the omega-3/omega-6 landscape change with the introduction of concentrates?

We add fat to horse feeds primarily through vegetable oils and cereal grains. The most popular vegetable oils, corn oil and soy oil, are quite high in LA, a short-chain omega-6 fat. Conversely, oil from flax seed contains predominantly ALA, a short-chain omega-3.

Fish oil is not high in short-chain fats but is high in long-chain fats in the form of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Other oils fed to horses do not have polyunsaturated fats as a predominant fat source. Canola oil and olive oil have high concentrations of monounsaturated fat. In rice oil, the predominant fat is monounsaturated. In coconut oil and palm oil, the highest concentration of fat is saturated.

Because we make horse feed with cereal grains like oats and corn that naturally contain oil and then we typically add soy oil, most commercial feeds tend to be highest in omega-6 fats and with different concentrations of omega-3 depending on the formulation. By feeding conserved forages and concentrates with added fat, we have changed the typical fat intake from a diet high in omega-3s to one high in omega-6s.

What types of fatty acids are incorporated into the horse's cell membranes?

We have done a lot of studies on changes in the composition of cell membranes because of different types of fat intake. The tissue that is most studied has been blood. Whole blood or plasma can be used, as can red blood cells and white blood cells. We have primarily studied plasma and red blood cell fatty acid composition in horses.

For our purposes, we tend to concentrate on red blood cells because the fatty acids have been incorporated into the cell membranes and that is the path we are interested in. Typically, researchers assume that red blood cell fatty acid content is indicative of all tissues in the body.

We have begun looking at other tissues, however, and have found the glandular portion of the stomach has a different composition than red blood cells. When we looked at the plasma or red blood cells taken from many horses, we observed a lot of variation in the amount of LA, a higher variation than we see in some other fatty acids. LA is one of the predominant fats in both plasma and red blood cells.

To see how diet affects the variability of these fats, we looked at two groups of horses: 13 Thoroughbreds in training in Florida fed a 5% fat textured horse feed, timothy hay, and bahiagrass pasture (a typical performance horse diet that was higher in LA than ALA), compared to 16 Thoroughbreds on pasture in Kentucky that were fed only a vitamin/mineral supplement (they were consuming more omega-3 than omega-6). When we compared these two groups, we found that pastured horses had lower LA and higher ALA than horses fed grain. Therefore, the amount of LA and ALA in plasma reflected their diets.

When we looked at arachidonic acid (AA) and di-homo-gamma-linolenic acid (DGLA), they were higher in grain-fed horses than pastured horses. If we looked at long-chain omega-3 fats, we found that concentrations of EPA and DHA were significantly higher in pasture-fed horses than grain-fed horses. In short, both omega-3 and omega-6 fatty acids in red blood cell membranes were reflective of the dietary intake of those fats.

Why would there be less EPA and DHA in horses fed diets high in LA?

To transform short-chain fatty acids to long-chain fatty acids, enzymes are used by both omega-3 and omega-6 pathways. They compete for the use of these enzymes. Excess LA intake might inhibit omega-3 fats from being desaturated, leading to a reduction in the amount of DHA and EPA. It has recently been discovered that LA (the short-chain fatty acid) can also produce inflammatory mediators without being elongated.

Are high levels of LA in and of themselves a problem?

An interesting study was recently published in Finland. Researchers looked at mature horses—both normal and those with osteoarthritis. When they studied synovial fluid, they found that horses with osteoarthritis had higher concentrations of LA in their synovial fluid than controls.

Earlier we had also looked at how high dietary fat affects the ability of older horses to clear glucose from their blood. We found that when older horses were fed a high-fat diet (30% calories from fat) it impaired their glucose tolerance after being given a frequently sampled glucose tolerance test. When we added fish oil to the diet, it reversed the glucose intolerance. That study and other studies have indicated that high levels of fat may have an adverse effect on glucose metabolism and that long-chain omega-3 fats might have a beneficial effect. In conclusion, in nonobese, aged geldings, a moderate intake of NSC improved glucose tolerance, and a high level of dietary fat impaired glucose tolerance. The glucose intolerance could be reversed by supplementation with fish oil.

What other research have you done with fish oil?

Fish oil has a high level of long-chain polyunsaturated omega-3 fatty acids. As we have shown in multiple studies, we can affect the concentration of EPA and DHA in red blood cells with supplementation. We have done studies that have shown a reduced incidence of inflammation after strenuous exercise with EPA and DHA. Rather than reducing LA in the diet or even fixing the ratio of LA to ALA, one solution is simply adding DHA and EPA. We know we can manipulate EPA and DHA, but we do not know if the higher LA is still a problem.

EPA and DHA are omega-3s. We have tended to think that all omega-6s are bad or inflammatory. However, DGLA has some anti-inflammatory properties. We have discovered that if horses are fed GLA, a short-chain omega-6 fatty acid, you can enhance the amount of DGLA. One lipid mediator of DGLA is prostaglandin E1 that, in synthetic form (misoprostol), is used to inhibit gastric acid and enhance mucosal resistance to injury in the stomach.

Could you increase the amount of DGLA by feeding GLA?

Yes. GLA converts linearly to DGLA relative to the amount of GLA fed. We subsequently performed a study where we looked at the addition of EPA and DHA along with GLA and compared those with supplementation with short-chain polyunsaturated fatty acids. We compared similar amounts of short-chain polyunsaturated fatty acids with long-chain omega-3 polyunsaturated fatty acids and GLA, the precursor to DGLA.

We fed these diets at the same levels to Thoroughbreds in training for three months. When we fed the short-chain polyunsaturated fatty acids LA and ALA, we did not get elongation into EPA in the red blood cell membranes. When we fed EPA, we got a significant increase in EPA in red blood cell membranes. We also observed the same thing with DHA. When fed directly, EPA and DHA increased. When fed GLA, we observed an increased concentration of GLA and, more importantly, an increased concentration of DGLA and a reduced concentration of LA in red blood cell membranes.

Through dietary manipulation, we are able the change the ratio of AA (long-chain omega-6) to these other three long-chain fats (EPA, DHA, and DGLA) from three times as much AA to a 1:1 ratio. Using a standardized exercise test on the racetrack, horses that were supplemented with GLA, EPA, and DHA had significantly reduced heart rate during intense exercise compared to unsupplemented controls. Post-exercise muscle biopsies were harvested to look at mitochondrial function. The supplemented horses had increased citrate synthase in their muscles, indicating mitchondrial biosynthesis.

Bronchoalveolar lavage (BAL) of the horses after exercise was conducted. We looked for concentrations of red blood cells in the BAL fluid. While none of the horses were bad bleeders, fewer red blood cells were found in horses fed long-chain polyunsaturated fatty acids compared to control horses. We were interested in what effects these fats would have on cytokines in response to an exercise stressor. One cytokine, interleukin-10 (IL-10), has anti-inflammatory properties that can help prevent tissue damage during exercise. When horses were fed long-chain polyunsaturated fatty acids, they had higher concentrations of IL-10 compared to controls and horses fed short-chain polyunsaturated fatty acids. We also looked at gastric ulcers when fed these diets. When supplemented with polyunsaturated fatty acids, we were able to significantly reduce severe squamous ulcers compared to unsupplemented horses.

What are the next steps in working out the optimal fatty acid composition of equine rations?

More research is necessary to determine if the overall concentration of LA in the diet affects performance horses regardless of its ratio to ALA or our ability to manipulate long-chain polyunsaturated fatty acids. Future research includes feeding fats that are not high in polyunsaturated fatty acid but higher in monounsaturated or saturated fats without affecting the delicate balance of polyunsaturated fats.

Heat Stress: A Comparative Approach Between Species and Disciplines

Mike Davis, DVM, PhD, DACVIM

Why do animals create heat and how do they deal with it?

Animals create heat because nearly every chemical reaction that occurs in a living cell flows "downhill" from an energy standpoint; when you consider the net amount of energy stored in the chemical bonds of the substrates and products, there will be less energy stored in the products.

The difference is released during the chemical reaction in the form of heat. As a general statement, more chemical reactions mean more heat production and fewer chemical reactions mean less heat production. With the production of heat being a foregone conclusion in any living subject, everything that makes up that living being has been designed based on the assumption that some heat will be present.

Most animals depend on a certain amount of heat always being present for everything to work correctly. But most mammals can only work correctly within a narrow range of body temperature, and body temperature at any given time is determined by the balance of heat production and heat dissipation.

Given that some animals, like athletic mammals, can have a 20-fold range of body heat production rates and may exist in environments with even larger ranges of heat content, they will need to have processes at their disposal to both conserve heat and dissipate heat to strike the proper balance.

What are the mechanisms in place to dissipate heat?

All animals are obligated to follow the laws of physics when it comes to dealing with heat. Heat moves through four different processes: radiation, convection, conduction, and evaporation. All of them occur at the interface between the body and the environment, and while it is virtually impossible to completely control any of these, there are physiological processes that can influence some of them.

The best-known example is evaporation, either sweat from the skin or moisture from the lining of the respiratory tract. The body can increase heat loss through evaporation by increasing the amount of sweat produced as well as increasing the amount of air that moves over the respiratory tract. On the other hand, conduction of heat from the skin can be inhibited by reducing the amount of air that comes and goes from the skin surface. This can be accomplished by increasing the density of the hair coat and in some cases causing the existing hair to "stand up" to increase the thickness of the hair coat without increasing the amount of hair.

What happens when these mechanisms fail, either partially or completely? What can go wrong?

Failure in either direction—failing to conserve heat or failing to dissipate heat—can result in the subject deviating from that narrow range of body temperature that allows the body to function. We will focus on only one direction—failing to adequately dissipate heat—for this presentation. What can go wrong depends a lot on how badly thermoregulation has failed. In mild cases, the adverse consequences reflect the body's reallocation of resources to mitigate the situation and prevent more severe failure. For example, we rely on the cardiovascular system to transport oxygen and glucose to muscles to support exercise, but we also use it to transport heat to the skin to be dissipated.

If we get too hot, the body may decide to "rob Peter to pay Paul," i.e., reduce blood flow to the muscles to increase it to the skin, thus reducing exercise capacity. Many other similar processes can kick in, all resulting in one thing: decreased performance because the athlete is too hot. If the situation is more severe, then there are more severe consequences.

Our recent research has shown that hot muscle tends to produce more oxidative stress, resulting in damage to the muscle that will need to be repaired. Also, too much heat will result in leakage of the lining of the stomach and intestines, resulting in the absorption of toxins that can activate inflammatory cascades and cause widespread damage to the athlete's tissues. In even more severe cases, body temperature strays outside of the range required for basic function, so basic functions fail. The athletes will lose proper neurological function, resulting in seizures, coma, and possibly death.

How do you define heat stress? In horses, what are the signs of heat stress that owners might recognize? Does this differ between species?

We all must speak the same language when discussing heat-related issues in athletes, because there is quite a bit of "sloppy" terminology that causes a lot of confusion. "Heat stress" is the increase in body temperature that causes any change in physiology that, in attempting to mitigate the increase in body heat, impairs another function.

A great example is a panting dog. That dog is changing its breathing pattern specifically to attenuate an increase in body temperature, but in doing so is impairing its ability to smell. Another example would be reducing blood flow to various organs to perfuse skin for heat dissipation. It is important to recognize that when defined in this manner, "heat stress" does not cause harm or injury; it is merely an indication that the body has become too hot to do everything, so it is starting to pick and choose. Next up in the sequence is "heat injury," which, as the name implies, means that injury has occurred.

This could be to muscle (often is), gastrointestinal tract (also frequently the case), or other tissues. It does not mean that overt clinical illness has occurred, but something has been damaged and will need to be repaired. Finally, there is "heat stroke," which is used to denote overt clinical illness because the systems have been so injured and so altered that the subject is no longer capable of even normal bodily function.

When considering species, there is only a major difference in the signs of heat stress, and that is because different species take different steps to increase the dissipation of excess body heat. However, to some extent, it is a matter of degrees.

Everyone recognizes that a dog will start panting to increase heat dissipation but, in reality, all species have this reflex to varying degrees. And while a dog won't divert blood flow to the skin to increase heat dissipation to the extent that a horse or a human will do so, if you get a dog hot enough it will at least try to do that as a last resort (after it has diverted blood flow to the tongue, gums, nose, and face).

Does heat stress affect equine athletes involved in various disciplines differently, e.g., between endurance athletes and high-intensity, short-duration athletes?

Heat stress can have different implications to different equine athletes, based on their discipline. A racing Quarter Horse that develops heat stress in a matter of 30 seconds does not have a huge problem because they are done exercising; the increased rate of heat production that produced the heat stress is done, and they can make cooling a priority because the race is over.

On the other hand, an endurance horse may develop heat stress only partway through its event, as it is faced with an extended period of continued heat production and is unable to actively cool without interrupting performance. In addition, an endurance horse is far more likely to be dehydrated, thus impairing cardiovascular function and making it more likely that the adjustments needed to find an appropriate heat balance will adversely influence exercise.

Heat stress (and the following stages of heat injury and heat stroke) is the product of both exercise intensity and duration but, in practice, duration is the more important factor.

How does conditioning factor in heat stress?

A major element in athletic conditioning, particularly aerobic conditioning, is an increase in peak cardiovascular capacity. This plays a direct role in the relative resistance to heat stress due to the role of the cardiovascular system in moving the heat through the body to be dissipated.

For a given exercise intensity, a well-conditioned horse is likely to be less susceptible to heat stress because it is better equipped to distribute blood flow to both the working muscle and the heat dissipation surfaces. But it is important to recognize the qualifier for a given exercise intensity. A well-conditioned horse with a greater cardiovascular capacity may also be capable of running faster, so instead of being less susceptible to heat stress, it may simply develop heat stress at a higher speed.

How can horse owners prevent heat stress?

There are numerous things that horse owners can do to prevent heat stress. First, emphasizing effective cardiovascular conditioning, for the reasons listed above. Second, preserving the improved cardiovascular conditioning by maintaining hydration: the lion's share of improved cardiovascular performance that results from athletic conditioning is due to increased plasma volume, which is basically water. If the horse is allowed to become dehydrated, then that benefit disappears. And can disappear quickly; depending on conditions, you can erase weeks' worth of conditioning in a matter of hours. Being prepared (both mentally and logistically) to cool the horse effectively is another area that owners can emphasize. Finally, be smart about deciding how hard to exercise the horse given the environmental conditions. If your conditioning program requires a certain level or duration of high-intensity exercise, make a point of picking a time of day that gives the horse the best chance of maintaining an acceptable body temperature.

Have you worked with biomarkers to predict or assess heat stress in horses or other animals?

I have done some work in both horses and dogs in this area, and have consulted on studies in humans. There is a general desire to come up with some sort of "formula" or device that can decide on whether an athlete is too hot but, in my opinion, the best approach remains a subjective assessment, with assessment improving in quality with a greater understanding of exactly how the athlete (whether it is a dog, horse, or human) thermoregulates.

Please provide two or three points about heat stress that you would really like veterinarians to take home with them.

- Heat stress, heat injury, and heat stroke always have two elements in the equation: rate of heat generation and rate of heat dissipation. Restoring normal heat balance can and should address both.
- Despite a lot of beliefs to the contrary, the best scientific evidence supports rapid cooling of an overheated athlete. There is no such thing as cooling an athlete too fast, although it is possible to cool them too much. If they are demonstrating signs of heat injury, get them to a normal body temperature as quickly as you can, but not lower than normal body temperature.
- Water application/immersion is by far the most effective practical means of cooling a hot athlete, and all organized athletic events should have that capacity on-site and available without restriction.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on heat stress written by you or someone you admire in the field?

When it comes to equine-specific publications, nothing beats the impressive effort that preceded the 1996 Atlanta Olympics. Faced with conducting a bunch of very high-profile equestrian events under intense public scrutiny in what can be described charitably as a hot, humid environment, the equine exercise physiology community stepped up to the task, learned a huge amount about thermoregulation in horses, and can rightfully claim to have been instrumental in the fact that there were no equine heat-related injuries at that event.

Any textbook published since then in the field of equine exercise physiology will have that complete record and is foundational knowledge in this day and age. I am a fan of *Equine Sports Medicine and Surgery* by K.W. Hinchcliff, A.J. Kaneps, and R.J. Geor.

More recently, the journal Experimental Physiology produced an entire issue (Volume 107, Issue 10)—a state-of-the-art review—of what is known about exertional heat stroke. I was a co-author on the paper addressing pathophysiology, but all of it is excellent reading. It is not equine-specific, but then again neither is exertional heat stroke. Bottom line: someone seeking to know the most current information on the topic should read this open-access issue.

Should You Sweat It? Anhidrosis in the Equine Athlete

Samantha Brooks, PhD
Sally DeNotta, DVM, PhD, DACVIM

What is anhidrosis? What is its prevalence?

Anhidrosis refers to a temporary or complete loss of the ability to sweat. In horses, it is characterized by the reduced or lack of sweat response to increased body temperature. This is a poorly understood condition with a varying prevalence in the horse population. In Florida, one study found that 6%–25% of Thoroughbreds were diagnosed with anhidrosis at least once in their careers. For non-racing horses, the incidence of anhidrosis varies between 2% and 11% of the horse population.

What are the symptoms of anhidrosis?

Owners may notice a partial or complete loss of sweat response, rapid breathing (tachypnea), elevated body temperature (hyperthermia), reduced appetite (anorexia), decreased water intake, hair loss (alopecia), dull hair coat, and depression. Severe cases result in collapse due to overheating, leading to convulsions, and without timely intervention, death.

Anhidrotic horses suffer considerably during work and warm seasons, do not perform well athletically, and have a reduced quality of life. These horses need intensive medical management and restricted physical activity in warm weather. They often must retire early from breeding or competition.

Can you briefly mention the horse's "cooling system"?

Healthy horses have a remarkably effective cooling system. Their sweat glands, distributed over most of the body surface, can move large volumes of water. To disperse excess body heat, the horse relies mostly on sweat evaporation. Still, the equine heat regulatory system can be overwhelmed, resulting in critical overheating.

Environmental temperatures exceeding 77° F (25° C) combined with 70%–90% relative humidity will begin to compromise heat loss through sweating, increasing body temperature. Without the sweat response, traveling a distance of roughly two miles at the gallop is estimated to increase a horse's core temperature by 11° F.

Why does the cooling system fail?

Overtraining and/or electrolyte imbalance can lead to the exhaustion of cooling mechanisms and a temporary reduction in the ability to sweat. Drugs such as antihistamines and macrolide antibiotics can also lead to temporary anhidrosis, but this condition usually resolves in a matter of days or weeks once the drug is withheld. In contrast, chronic idiopathic anhidrosis (CIA) occurs for more than one consecutive summer season and does not resolve despite changes in housing, diet, and exercise schedule. Veterinarians can use an intradermal sweat test (with terbutaline) known as QITST to quantify the sweating capacity of horses. It can also aid in the diagnosis of CIA. However, the factors that distinguish acute but reversible anhidrosis from a permanent loss of sweating ability are still unknown, and the underlying cause of chronic idiopathic anhidrosis remains unclear.

What are other trigger factors for the onset of anhidrosis?

Environmental factors such as overtraining, failure to acclimate after travel to a hotter climate, electrolyte imbalance, and dehydration can trigger an acute anhidrotic episode.

However, CIA defies these trigger factors. A University of Florida study revealed that, at the individual level, the risk for anhidrosis varies significantly by breed, with Thoroughbreds and Warmbloods at higher risk. Researchers also observed that the odds of manifesting anhidrosis are 21.7 times higher in horses with a family history of the condition. These findings strongly support the theory that there is a hereditary/genetic component contributing to anhidrosis in horses.

Are there any treatments for anhidrosis?

Numerous "treatments" are advertised for this condition. Most are based on speculation, anecdotal reports, and at best, uncontrolled and unreplicated studies conducted in a small set of horses. Some attempted therapies include dietary supplements, electrolytes, methyl-dopa, clenbuterol, and thyroid supplementation. Alternative therapies such as acupuncture and herbal supplements are also popular but were shown to be ineffective long-term in at least one report. To date, no treatment for anhidrosis has passed even the minimal standard for evidence-based medicine.

Can you provide some management tips for chronically anhidrotic horses?

- Work horses during the cooler parts of the day such as evening or early morning.
- Observe the horse closely for signs of overheating such as rapid respiratory rate and high body temperature.
- When introducing a new horse to a hot and humid climate, allow the horse to adjust to turnout time and light work.
- Start a horse's spring/summer workout regime earlier, so that it is more fit for the hottest parts of the season.
- Always make sure shade is available. Misters and fans can be used for added cooling.
- Hosing off a horse during the hottest parts of the day will help keep its internal body temperature down.

The information provided in the preceding portion of the Q&A was originally published as "Anhidrosis in the Horses (Non-Sweaters)—What Do We Know?" a University of Florida Extension publication written by Laura Patterson Rosa, Martha F. Mallicote, Robert J. Mackay, and Samantha A. Brooks. The original publication, which contains a complete list of references, can be found at: https://journals.flvc.org/edis/article/view/117703/126670.

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Dr. Samantha Brooks has provided the answers for the following questions.

Can you tell us how you became involved in genetics?

I followed a pre-vet track in my undergraduate degree, but of all the classes I took, genetics was the one that most often captured my interest, especially once I joined a new major in "agricultural biotechnology." That major required a capstone project in research, and I was lucky enough to find Dr. Ernest Bailey as a mentor for that work. Equine genetics seemed to be the perfect blend of horse health and molecular biology for me.

How did you become involved in anhidrosis particularly?

Anhidrosis is not a problem specific to tropical and subtropical climates, but it is more obvious in places where it is tough to escape the heat, like Florida. There was already a strong history of anhidrosis research at the University of Florida, led early on by Dr. Rob Mackay.

Yet, the anhidrosis project was driven really by the passion of two people: Dr. Laura Patterson Rosa, my doctoral student at the time who had lost a beloved Thoroughbred mare that she showed as a jumper to anhidrosis, and Dr. Martha Mallicote, who along with Rob convinced me that the time was right to start to tackle this very frustrating disease using genetics.

Can you explain to people who aren't geneticists how you went about studying the genetics of anhidrosis?

To look for heritable factors contributing to anhidrosis, we used a technology that examines markers found evenly across the entire genome of the horse. Each of these markers has two different possible types, and tracking these types in each horse gives us a way to distinguish one region of the genome from another.

We gathered a population of horses of similar breeds all displaying signs of idiopathic anhidrosis, as well as a group of horses from the same breeds with no known history of the disease.

By comparing the types observed at all the genetic markers across these two groups we identified a set of markers, all found in one region of the genome, where the marker types lined up with the diagnosis of anhidrosis. Then we can use the reference genome as a road map, studying what is known about that region, what genes are likely found there, and to look specifically for genetic variants found specifically in the anhidrosis group.

How did you isolate the gene that is responsible for anhidrosis?

Once the genome mapping step gave us a region likely to contain a heritable risk for anhidrosis, we studied the functions of genes in the region, and research from other species (humans and lab mice mostly) that tied these genes to other diseases. This comparative approach pointed to the gene we focused on, based on its role in ion transport.

What insights did you glean from this study?

There is a genetic component to chronic idiopathic anhidrosis, so breeders should think carefully about using breeding stock that might have retired from their careers due to this disease. Testing for this risk allele is also available to breeders, though it is important to note that it does not completely explain the disease. More work is needed to understand all the genetics and environmental triggers at play.

The implicated gene highlights a role for ion transport, though not in the way we might have envisioned. Just feeding electrolytes clearly will not solve the problem, though it could be an important part of the supportive care plan for a horse with anhidrosis. It also suggests that we need to study the possibility that horses with anhidrosis may also have alterations in their pain sensation and, potentially, risk for arrhythmia.

Given the role of this gene in human diseases, like cystic fibrosis, and the millions of research dollars spent to target these pathways pathologically, there may be an avenue to pursue preventatives/treatments for horses adapted from the arsenal of drugs developed to modulate these channels in human patients. All that is needed is the research funding to pursue these compounds.

How will this information be useful going forward? What is the next step?

Testing for the identified risk locus may be useful for breeders and clinicians looking for an additional diagnostic, as a start. Pursuing some of the more impactful applications, like additional phenotypes and possible treatments will require more research support from the industry.

Is there a way for horse owners to achieve early detection?

Genetic testing can be done in parents of prospective foals, and on embryos prior to transfer to a recipient mare. Certainly, it is easy (and economical!) to incorporate comprehensive genetic testing into a prepurchase exam. Finding a risk allele does not guarantee the horse will develop the condition, nor does absence ensure the horse will never develop it. However, it does provide an important hint at the future to keep in mind during the decision-making process.

What do you want people to know when they leave your presentation?

This project was carried in large part by the horse owners who voluntarily enrolled their horses, and initial funding was provided by the American Quarter Horse Foundation. It is just a glimpse of what could be accomplished if the horse industry could pull together to support horse health research.

As a side note, we are not continuing the anhidrosis research, as we have run out of funding for it. Due to some administrative politics, we are no longer allowed to apply for grants from the American Quarter Horse Foundation, or other nonprofit groups supporting horse health research. Those in the audience who are Florida residents and think work like this is important are urged to contact their representatives in state government to voice their support for horse health research in Florida.

Can you recommend three to five seminal references (articles, book chapters, books, etc.) on anhidrosis written by you or someone you admire in the field?

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New Methods for Studying Heat Stress in Horses

Joe Pagan, MS, PhD

Briefly explain the history of measuring muscle temperature in horses.

Understanding how much heat a horse produces and how quickly the horse can dissipate it is certainly an important factor in deciding how to manage performance horses. Historically, making those measurements proved an invasive process, particularly to measure changes in muscle temperature or core body temperature. This entailed implanting thermistors into muscle and running thermistors through catheters into the cardiovascular system or, at the other extreme, using inexact measurements like rectal temperatures under field conditions.

How has this study changed recently?

An important innovation has come about, and that is the use of microchips that can measure temperature. These are the same types of microchips used for identification in dogs, cats, and horses, but now they measure temperature.

Traditionally, microchips are put in the nuchal ligament in the neck, but recently a research group at the University of Queensland experimented with taking these microchips and placing them in various muscle tissues in the horse. The researchers then performed studies to look at how well the temperatures taken by microchips correlated with core body temperature, which they measured by insertion of a catheter deep in the cardiovascular system.

The researchers implanted these chips into different muscles, one of which is the middle gluteal muscle, a major muscle that overlies the hip of the horse. The middle gluteal muscle is commonly biopsied in exercise physiology studies to look at the effects of exercise on muscle metabolism.

The second location is the pectoral muscle. They then performed a series of standard exercise tests on a treadmill, and they looked at how well the central venous temperature (core temperature) correlated to muscle temperature and rectal temperature. They inserted a thermistor further into the rectum than a normal handheld thermometer. During and after exercise, the microchips in muscles were highly correlated with core temperature.

At Kentucky Equine Research, we have recently taken this validated technology and have applied it to 12 Thoroughbreds that we use in our research program in Florida. Each of these horses had microchips implanted into its middle gluteal muscle and pectoral muscle. These horses were physically fit, as they had been in training on a high-speed treadmill for several months. We conducted several studies first to see what kind of muscle temperature occurred in response to treadmill exercise. We also looked at how various types of cooling methods affected recovery, muscle temperature, heart rate, and respiratory rate.

One advantage of these microchips is they allow researchers to measure muscle temperature while the horse is exercising. For each of the studies we did, horses performed a standard exercise test on the treadmill. These tests typically involve exercise at increasing speeds up to maximum speed and then decreasing speed incrementally—from a gallop to a trot, trot to walk, walk to stop. We found that the horses' heart rates responded predictably when we reached the peak speed. Peak heart rate (HR) occurred at peak speed. As soon as the treadmill speed was reduced, heart rate dropped and continued to drop until the end of the test. Muscle temperature did not decrease in the same pattern at all, and continued to increase even as the horse slowed down. By the end of exercise when the horses' heart rates had recovered to a significant degree, the muscle temperature was still high. The cardiovascular system responds largely to the extra energetic needs of the muscle by delivering oxygen to the muscle.

In these studies, we found that resting muscle temperature was around 37° C (99 F°). As soon as we began exercise, muscle temperature increased and, in many of our studies, muscle temperature would reach 41–42° C (106–107° F).

In one study, peak heart rate reached an average of 208 beats per minute (bpm), which was 170 bpm over pre-exercise heart rate. Immediately post-exercise heart rate was only 44 bpm above pre-exercise, while respiration rate was still 105 breaths per minute higher than pre-exercise. Muscle temperatures were 4.2° C (middle gluteal muscle) and 4.8° C (pectoral muscle) higher. In a number of these studies, we used recovery protocols that might be like those used in the field. Some would entail hosing the horses right after exercise. In those studies, we typically would use regular tap water (out of the hose), and we would hose the horses for five minutes and then walk them for 10 minutes.

In another study, we walked horses for 10 minutes and then hosed for five minutes. In that study, after we had walked the horses for 10 minutes and hosed for 5 minutes, their respiratory rate had dropped to only 20 breaths per minute above pre-exercise and heart rate had dropped to 14 bpm above pre-exercise values. Muscle temperatures did not recover nearly as fast as heart rate or respiratory rate. The pectoral and middle gluteal muscle temperatures were still >39° C. At a time when the horses were considered cooled and put back into their stalls, their muscle temperatures were only halfway recovered.

In some of these studies, we also measured rectal temperature using a handheld digital thermometer. This measurement did not reflect the muscle temperature at all. It greatly underestimated the muscle temperature immediately after exercise and lagged real muscle temperature. Rectal temperature is not a reliable way to understand how hot a horse is, especially after exercise.

In one study, we purposely wanted to see how muscle recovered with passive cooling (walking the horses for 15 minutes and not applying water) versus more active cooling protocols (hosing the horses, walking the horses under misting nozzles on the mechanical walker, and placing the horses under cooling mats). These cooling mats are manufactured by Gentherm Medical and are typically used in human operating rooms.

Cold water (4° C or 40° F) circulates through the mats. We placed these under traditional sheets so we could continue to apply coolness to the horses as they stood tied in a wash stall for 45 minutes. We compared these horses to those that did not get anything but 15 minutes of walking and 45 minutes of standing under fans. We saw a major difference in how well the muscles cooled. We did this to develop a model for studying exercise-induced heat stress.

We are in the process now of seeing if active versus passive cooling affects muscle integrity, measures of inflammation, gastrointestinal integrity, and other processes that are normally attributed to heat stress. All of the lab work for many of these studies is in process, so details of the studies are not finalized.

An interesting side to this is that we tested all the horses using the terbutaline testing technique to assess anhidrosis. Using that technique, you grade the degree of compromise the horse has from sweating on a scale of 0 to 5. A score of 0 indicates a free sweater, while a score of 5 indicates complete anhidrosis. When we measured our research horses, many of them were compromised sweaters, from mild (1/5) to compromised (4/5).

We had noticed in our initial studies that the degree to which the muscle heated during exercise was consistent and variability between horses narrow. When we looked at how well the horses cooled, there was a lot more variability, even when they were treated with the same types of cooling modalities.When we took those horses and divided the ones with compromised sweating versus the free sweaters, this explained the difference in their ability to cool. The ones that have compromised sweating had a more limited ability to cool than the ones considered free sweaters.

This brings a whole new area that we can investigate in terms of how well horses handle and dissipate heat. The horses that were compromised sweaters tended to have higher respiration rate immediately after exercise and the drop in muscle temperature in their middle gluteal muscle was slower than those horses that were free sweaters.

In one study, we hosed the horses immediately after exercise. In that study, in a less hot and humid environment, the horses' respiration rate recovered to almost resting at the end of the hosing period (5 minutes). Interestingly, we found horses that were compromised sweaters did not recover their middle gluteal muscle temperature as quickly as the free sweaters. This raises an intriguing question, even though we are in the early days, whether causing a horse to rapidly reduce its respiration rate, particularly one that has a compromised ability to sweat, is really what you want to accomplish. In normal sweaters, their main means of heat dissipation is sweating. In compromised horses, evaporation and convection through respiration become more prominent methods of heat dissipation. Hosing effectively reduces respiration rate, but do you want that to slow respiration if the horse cannot cool itself in some other fashion?

In another study, we looked at precooled horses. Horses were precooled for 30 minutes before a standardized exercise test using the cooling mats. After the test, we hosed the horses for 5 minutes and then walked them for 10 minutes. We found that precooling decreased peak heart rate during exercise and decreased the middle gluteal muscle temperature during exercise, but that seemed to be more effective in anhidrotic horses. Precooling also decreased the respiration rate in the horses that were free sweaters but not in the compromised horses after exercise.

Because this is brand-new technology, we are in the early days of understanding different cooling methodologies and potential nutritional interventions to aid recovery. Having the microchip technology available to us is especially encouraging, as this is a repeatable, noninvasive method for accurately measuring recovery from exercised-induced heat stress.

Electrolytes and Hydration

Joe Pagan, MS, PhD

What are electrolytes?

Electrolytes are a class of minerals that disassociate into electrically charged ions in water. They play an important role in maintaining osmotic pressure, fluid balance, and nerve and muscle activity.

The major electrolytes are sodium, potassium, and chloride, and lesser ones include calcium and magnesium. Electrolytes are important all the time for the normal functioning of the horse, but they are particularly relevant for performance horses because large quantities of these electrolytes are lost in sweat. Sodium and potassium are positively charged, so they are considered cations. Chloride is negatively charged, so it is considered an anion.

Sodium is the principal cation in extracellular fluid and determines the volume of extracellular fluid—fluid outside the cell. Interestingly, almost 50% of a horse's sodium is in its skeleton, so it is not readily available to influence extracellular fluid. The rest of the sodium is distributed through blood, muscle, other tissues, and the gastrointestinal tract.

Potassium, which is the other major cation, is primarily found in the intracellular fluid—fluid inside the cell. Cells maintain their electrical charges by pumping sodium out of the cell in exchange for potassium going into the cell. The majority of potassium (75%) is in the muscle.

Chloride is the main anion, and it can be found in practically all body fluids, including sweat, saliva, and gastrointestinal fluids, to name just a few, and is distributed uniformly throughout most tissues. Chloride plays an important role in acid-base balance.

Describe how horse sweat differs from human sweat.

The horse uses sweating as its primary method of thermoregulation during exercise. Horse sweat is different than human sweat, though, as it is considered hypertonic or isotonic, meaning the concentration of electrolytes in horse sweat is about equal to (isotonic) or higher than (hypertonic) the concentration of electrolytes in the blood. Horses produce a lot of sweat. Given the same surface area, horses generate three times the amount of sweat as humans.

Can you measure electrolyte losses in horses?

We can calculate electrolyte losses based on how much sweat is lost, yet measuring sweat is not easy. Sweat carries heat to the skin and dissipates it through evaporation. If evaporation is working as it should, the sweat dries quickly, before it is seen. The surest way to measure sweat loss is through weighing the horse before and after exercise. While this can be achieved easily in a research setting, it is not practical in the real world.

What are some basic electrolyte recommendations?

How much electrolyte the horse requires in its diet depends on many factors, including the duration and intensity of exercise, ambient conditions (heat and humidity), and whether the horse is already acclimatized to the heat.

Let us start at the beginning, with horses that are not exercising. Electrolyte requirements for average-sized horses (500 kg or 1,100 lb) are about 10 grams of sodium, 25 grams of potassium, and 40 grams of chloride daily. These maintenance requirements can be met by forage, which is typically high in potassium, and supplemental salt in the diet. Most of the things we feed horses—forages and grains, for example—are low in sodium so we must provide a source of sodium. Most horse feeds are fortified with salt. As insurance to meet salt requirements, all horses should have free-choice access to a salt block or have 30 g(1 oz) of salt added to their feed each day. When horses start to sweat, requirements increase quickly, and electrolytes should be replaced through supplementation.

What kind of electrolytes should you provide?

Many electrolytes on the market contain large amounts of sugar. Manufacturers add sugar to boost palatability, as sugar is more appetizing than salt. But sugar was not lost in the sweat. Owners should make sure they choose an electrolyte supplement formulated to mimic the losses in sweat.

In addition to an improvement in palatability, there is another reason some believe that sugar is added to these electrolyte formulas: to improve the uptake of electrolytes. We've done two studies that indicate this is not true. In one study, we gave an electrolyte mix that included sodium, chloride, and potassium in the level calculated in sweat, the same electrolyte mix with a small amount of dextrose (sugar), the same electrolyte mix with a large amount of dextrose, and a control. The electrolytes were dissolved in one liter of water and then dosed using a nasogastric tube.

We tested blood hourly for four hours after dosing. The uptake of electrolytes was the same for all three electrolyte treatments. Sodium did increase after feeding all three of the electrolytes, as did osmolality. Osmolality is a measure of the concentration of all chemical particles in the blood. An increase in sodium and an increase in osmolality stimulate thirst. We also measured voluntary water intake. In all three electrolyte treatments, the horses drank significantly more water than the control.

We conducted a second similar experiment except we gave the electrolyte mixed with water containing deuterium oxide (D20), which is a stable, nonradioactive isotopic form of water. We found the electrolytes absorbed at the same rate, but the control had a higher increase in D20 in plasma at 30 and 60 minutes after dosing, suggesting that electrolyte solutions delay water uptake compared to pure water.

In sum, find an electrolyte with not too much sugar and the correct ratio of sodium, chloride, and potassium. Unless the exercise is of long duration, like that of an endurance horse, or furosemide was given, do not worry about calcium. If you are in an endurance situation, where a horse is producing lots of sweat, calcium losses become more meaningful.

Although calcium is present in sweat at a much lower concentration than sodium, chloride, and potassium, we typically add calcium to electrolyte supplements intended for endurance horses.

If I lead a horse to water, how do I make it drink?

Since sweat is isotonic or hypertonic relative to blood, after exercise the sodium content of blood is often the same or lower than pre-exercise and the horse is not thirsty. One trick to stimulate thirst is to give an electrolyte paste after exercise. A paste is like a powdered electrolyte except it is in a convenient, easy-to-administer form. Providing horses electrolytes through paste or powder will increase the concentration of sodium in the blood, which will stimulate thirst and will encourage horses to drink. An important consideration is that horses must have access to water after electrolyte administration. Electrolytes tend to draw out water from the blood and into the gastrointestinal tract. Giving the horse electrolytes without water is never advised. The horse should have plenty of water available.

The second trick came by way of research. In one study, we fed 2.5 kg (5 lb) of grain with free-choice water. We fed hay based on three treatments: hay fed two hours before the grain, hay fed with the grain, and hay fed four hours after the grain. We found the horses drank water in response to hay, not grain. The horses that got their hay two hours before their feed drank soon after they ate their feed. The horses that got hay and grain together did not take the big drink of water till two hours later, and the horses that got hay four hours after being fed grain did not take a drink until five hours after being fed grain.

Hay draws out fluid from other areas of the body (saliva, for example), so you partially dehydrate the horse with the hay. That type of dehydration stimulates the horse to drink. One of the best ways to get a horse to drink is to give it hay.

One school of thought involves teaching horses to drink salty water, which is water mixed with the ideal concentration of electrolytes. Controlled studies have proven this to be effective, but that means you must train the horses to accept salty water well before the competition. If you decide to offer salty water for the first time at a competition, it can backfire. It is best to give horses a source of fresh, unsalted water in addition to salt water, as the last thing we want to teach horses is to be wary of water.

When horses are heavily exercised, they lose a lot of sweat and electrolytes. It is not necessary to completely replete the electrolytes that day. A sound rule of thumb: with heavy electrolyte loss, replace 40% of lost electrolytes on the first day, 40% on the second day, and 20% on the third day.

If horses consume more electrolytes than they need and they are drinking water, it is no problem because horses have a great ability to regulate electrolytes. Excess electrolytes are passed in the urine. The dangers involve not giving horses enough electrolytes and not allowing the horse plenty of opportunity to drink after electrolyte consumption. It is important to match electrolyte and water intake. The horse will typically do that on its own when they consume electrolytes.

While we normally think of electrolyte losses in performance horses as being primarily through sweat, horses can lose electrolytes through increased urination if given furosemide.

Describe electrolyte loss in horses administered furosemide.

Furosemide is a diuretic given to horses susceptible to exercise-induced pulmonary hemorrhage (EIPH). We have done several studies looking at electrolyte and fluid losses when furosemide is used.

A lot of researchers have used furosemide as an alternative to getting a horse to sweat instead of exercise. The ratio of electrolyte loss, however, is not the same with furosemide as it is with exercise.

Through complete urine collection, we have quantified electrolyte losses after furosemide administration. Horses lose about 2% of body weight when given the normal dose of furosemide, about 10 liters of urine. This is similar to the amount of sweat a horse would lose in a strenuous competition, so fluid loss is nearly the same.

Sodium and chloride are lost with furosemide administration, though the amount of potassium lost is not as high in urine for horses given furosemide compared to sweat. Potassium loss was not increased with furosemide. However, another mineral lost in appreciable amounts is calcium. So, with horse sweat, horses lose sodium, chloride, and potassium; with furosemide administration, horses lose sodium, chloride, and calcium.

What kind of electrolyte do you give performance horses administered furosemide?

Normal electrolyte losses can be replaced with Restore®, which contains the ratio of sodium, chloride, and potassium that mimics sweat loss. Our research with furosemide led to the creation of a new product.

If a horse has been given furosemide (barrel racer, racehorse), we developed Race Recovery™, a hybrid electrolyte intended to replace losses associated with both sweat and furosemide use. It contains more calcium than a higher sweat-replacement electrolyte because we know there are meaningful amounts of calcium lost in urine after furosemide administration.

The other thing we discovered from all the work with furosemide was that furosemide caused horses to lose fluid as urine but they did not drink any more than horses that were not given furosemide. Water is typically withheld for four hours after furosemide administration. Horses given furosemide do not have higher levels of sodium in the blood, so horses are not signaled to drink. The horses aren't thirsty, and they do not drink, so they do not replace lost fluids right away. It takes a long time for these horses to bounce back. It may take several days for horses to recoup body weight.

If you give exercising horses or furosemide-treated horses a hypertonic (concentration higher than blood) paste, it stimulates them to drink, and they replete the lost fluids quicker than if they don't get some stimulus to drink. For horses that have just exercised and lost sweat, the stimulation can just be added to the next grain meal.

Post-competition strategy includes giving the horse hay and then a couple of hours later giving feed with added electrolytes (30-60 g; 1-2 oz of electrolytes). Do not oversupply electrolytes in the grain meal, as there is danger in making it too salty, which will negate what you are trying to accomplish.

In 100 words or less, provide some simple, sound electrolyte recommendations.

For horses at maintenance: free-choice salt (or 1 oz/day) and hay.

For horses in regular training: if they are sweating, provide an electrolyte that matches the composition of sweat.

For horses in heavy competition: provide an electrolyte that matches the composition of sweat and replace electrolytes over two to three days.

For horses given furosemide: give hypertonic paste post-exercise to stimulate thirst and provide additional calcium.

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