EQUINE VETERINARY EDUCATION/AMERICAN EDITION

VOLUME 36 NUMBER 3

MARCH 2024



AAEP®

# EQUINE VETERINARY EDUCATION

American Edition | March 2024



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The official journal of the American Association of Equine Practitioners, produced in partnership with BEVA.


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References:
[1] Nogradi N, Couetil LL, Messick J, Stochelski MA, Burgess JA. Evalutation of an Omega-3 Fatty Acid Containing Feed Supplement in the Management of Horses with Chronic Lower Airway Inflammatory Diseases. J Vet Intern Med 2015; 29:299-306
[2} Couetil LL, Cardwell J.M, Gerber V, Lavoie J.P, Leguilette R, Richard E.A. Inflammatory Airway Disease of Horses. ACVIM Consensus Statement of J of Vet Intern Med 2016; 30:503-515 p. 508-510.

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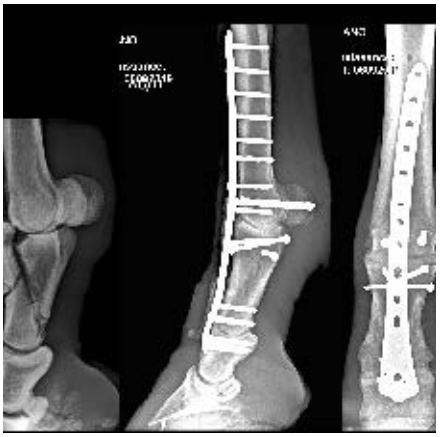

**AAEP NEWS** In this issue



*Cover photo by Dr. Ruth Sobeck.*

# EQUINE VETERINARY EDUCATION
## AMERICAN EDITION
**MARCH 2024 • VOLUME 36 • NUMBER 3**



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***Published monthly. Deadlines are the fifth of the preceding month.***

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**AAEP Mission Statement:** To improve the health and welfare of the horse, to further the professional development of its members, and to provide resources and leadership for the benefit of the equine industry.




*Equine Veterinary Education* is a refereed educational journal designed to keep the practicing veterinarian up to date with developments in equine medicine and surgery. Submitted case reports are accompanied by invited reviews of the subject (satellite articles) and clinical quizzes. Tutorial articles, both invited and submitted, provide in-depth coverage of issues in equine practice.

*Equine Veterinary Education* (American Edition ISSN 1525-8769) is published monthly by the American Association of Equine Practitioners, an international membership organization of equine veterinarians. Office of publication is 4033 Iron Works Parkway, Lexington, KY 40511. Periodicals Postage paid at Lexington, KY and additional mailing office. POSTMASTER: Send address changes to: Equine Veterinary Education, 4033 Iron Works Parkway, Lexington, KY 40511.

Communications regarding editorial matters should be addressed to: The Editor, Equine Veterinary Education, Mulberry House, 31 Market Street, Fordham, Ely, Cambridgeshire CB7 5LQ, UK. Telephone: 44 (0) 1638 720250, Fax: 44 (0) 1638 721868, Email: sue@evj.co.uk.

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All subscription inquiries should be addressed to: Subscriptions Department, AAEP, 4033 Iron Works Parkway, Lexington, KY 40511, Telephone: (859) 233-0147, Email: jcooney@aaep.org. Subscription rates: AAEP annual membership dues include $40 for a subscription to *Equine Veterinary Education*. Other subscriptions at $151.80. Single copies $37.50.

Canadian Subscriptions: Canada Post Corporation Number 40965005. Send change address information and blocks of undeliverable copies to AAEP, 1415 Janette Avenue, Windsor, ON N8X 1Z1, Canada.





Printed by: Sheridan, Hanover, PA.

# From the president: Out of the abyss

*By Katie Garrett, DVM, DACVS*

*Dr. Katie Garrett*

In January, I had the opportunity to attend the AVMA Veterinary Leadership Conference in Chicago and address the AVMA House of Delegates during a session examining challenges facing the veterinary profession. It's no secret that many of today's challenges aren't confined within the equine sector or the small animal sector, but instead are faced by our colleagues across the practice spectrum.

I spoke about the solutions and resources created to date by the subcommittees that make up our Commission on Equine Veterinary Sustainability. Many of our solutions are not only relevant to equine practice but to veterinary medicine overall, eliciting an overwhelmingly positive reception from the audience.

Reflecting on that day, I think back to a sentence penned by Dr. Emma Read on this page in the December 2022 issue. She wrote: "Just as equine seemingly led the way into the abyss, I am positive that we will be the ones to effectively lead the way out." It appears her vision has become reality.

Leading the way have been the dozens of member volunteers who have given generously of their time and expertise by serving on subcommittees devoted to the key areas of compensation, emergency coverage, practice culture, internships and the student experience. I am extremely proud of what has been accomplished in such a short time and would like to share some of these take-home, immediate-use tools with you, all of which are accessible by scanning the QR code or visiting https://tinyurl.com/2v6e9z23.

The Compensation Subcommittee has been busy "myth busting" using results of its salary survey that, among other things, found that the average salary for a recent graduate in equine practice (not including internship positions) was $89,000 per year. This is much higher than previously reported. They've also completed and distributed the results of a fee survey to assist our members with setting appropriate fees for services, which can impact compensation.

The Emergency Coverage Subcommittee has identified strategies to ease the burden of on-call responsibilities that are a major source of stress and burnout for equine practitioners. Knowing that there's no one-size-fits-all solution since every practice is unique, the subcommittee has published a toolkit that describes in detail a variety of successful emergency coverage models that are working in the "real world." In addition, social media is being used to educate horse owners on how to appropriately utilize their veterinarian and understand that their veterinarian may not be available to them 24-7-365.

We all want to work at a place where we feel valued and respected, and this has been the focus of our Practice Culture Subcommittee. They identified seven pillars that contribute to the individual culture of a practice and then developed a handbook with actionable resources for practice owners to evaluate their culture and then improve upon it to help make every practice a desirable place to work.

Our Student Subcommittee has prioritized outreach to veterinary students and conducted a survey of students to determine their needs. They established a speakers' bureau of practitioners who are visiting veterinary schools to connect with students and share their positive experiences as equine veterinarians. Speakers visited 25 veterinary schools in 2023 with plans to visit the rest in 2024. Steps are also underway to strengthen the role of the student chapter faculty advisor to ensure that student members have easy access to AAEP's clinical skills workshops and other programs.

Finally, the Internship Subcommittee has focused on improving the internship experience for both the intern and the practice. They have created a comprehensive best practices handbook to help practices provide a high-quality, educational internship experience to new graduates as well as to help potential interns understand the expectations of an intern position. The subcommittee has provided veterinary students with additional tools to help them evaluate internships to identify positions that are a good fit for their personal career goals. Identifying and applying for internship and externship opportunities will become easier than ever when the AAEP rolls out a redesigned website this year.

Our work on sustainability issues is certainly commendable but far from done. We must continue striving to make equine practice a sustainable, fulfilling and rewarding career for those of us in it and those who will come after us. However, I believe we have taken the first steps in the right direction to ensure a healthy supply of equine veterinarians for the future. And if you have a success story to share, please let us know by emailing Sally Baker at sbaker@aaep.org to share how you've embraced the innovation occurring throughout equine practice.

## Board of directors sets priorities for the year ahead

*By David Foley, Executive Director*

The AAEP board convened Jan. 21 for its winter meeting with all directors present. Following is a synopsis of the meeting.

After introductory remarks by AAEP President Dr. Katie Garrett and approval of the minutes from the December 2023 board meeting, discussion turned to the strategic plan. Specific actions taken were:

- Approval of a motion to form a standing committee for Students and Early-Career Veterinarians. At its prior meeting, the board approved a task force on the recent graduate to consider the feasibility of a young members subsection of membership and recommend ways to facilitate input from recent graduates. This task was redirected to the Member Engagement Committee.
- Approval of a motion to form a task force on the Foreign Equine Veterinarian charged with identifying the best means to create a pathway for foreign graduates who wish to enter the U.S. workforce, where appropriate.
- Tabling of a joint proposal from the Wellness Committee and Practice Culture Subcommittee for a strategic planning session for the Commission on Equine Veterinary Sustainability until a more prescriptive and targeted approach to the session is formulated.

Considerable discussion ensued on the topics of membership, member communication and governance. The Member Engagement Committee will conduct needs assessments and/or focus groups with solo practitioners, mid-career members and members less than five years post-graduation to determine how the association can better enfranchise these membership segments. In addition, the board approved a motion to engage the services of an association communications consultant to provide guidance on improving communications amongst all membership categories in terms of content, frequency and modality. Finally, the board approved a motion to form a governance review task force charged with reviewing the AAEP bylaws and creating a mechanism for member feedback.

The board then considered a recommendation from the Welfare & Public Policy Advisory Council on revised guidelines for Equine Transportation. After much discussion, the board took no formal action, as there were a few areas that needed further clarification. The board will provide feedback to the council members for their consideration.

During a segment on The Foundation for the Horse, the board approved The Foundation's latest strategic plan while Drs. Sara Langsam and Erin Contino volunteered to serve as liaisons to The Foundation's Horses at Risk and Research working groups, respectively. Dr. Jackie Christakos remains the liaison to the Disaster Medicine working group.

The meeting's focus shifted to other business, including a mid-year update on the AAEP and The Foundation for the Horse finances, which remain healthy; a recap of feedback from the 2023 Annual Convention; and discussion of AAEVT Guidelines. In addition, Dr. Sarah Reuss discussed horse care guidelines that the Horse Owner Education Committee drafted jointly with the AVMA. A motion to approve the guidelines passed.

The meeting concluded with establishment of the following priority areas for 2024:

- Sustainability: Merge the Student and Internship subcommittees at the appropriate time later in 2024 to cohesively address priorities of our youngest members; and pursue the practice culture/wellness planning session and the foreign DVM pathway. The Sustainability focus is themed "Recruit-Retain-Rekindle-Reinvent" with initiatives under each heading.
- Committee Review: Develop a process for reviewing/evaluating all committees and their charges and institutionalize a regular review process. This will include a process for committee chair/new member orientation, goals and accountability.
- Communication: Evaluate and implement consultant recommendations for improving member communications.
- Governance/Bylaws: Form a task force to review the AAEP bylaws for possible revisions.
- Education: Launch a wet-lab series initiative and review the convention assessment for potential enhancements to the meeting.
- Membership Initiatives: Initiate a category review, including possible solo and mid-career categories.

The next board meeting is July 15 in Lexington, Ky.

## 5 things to know about AAEP this month

1. Improve the veterinary experience for both the practice and client by downloading and sharing the new Veterinarian-Client Agreement for Effective Equine Care available at aaep.org/node/38237.
2. Thinking of starting an ambulatory practice? Don't miss the March 27 Virtual Wednesday Round Table episode. Register to watch live at learn.aaep.org.
3. Check out the catalog of resources from each subcommittee of the Commission on Equine Veterinary Sustainability at https://tinyurl.com/2v6e9z23.
4. Honor the achievements and service of an admired colleague by nominating the individual for an AAEP Annual Award by June 1 at aaep.org/about-aaep/annual-awards.
5. Achieve cost and time savings when purchasing practice supplies by becoming an AAEP Inner Circle member on Vetcove. Learn more about this free benefit at aaep.org/dashboard/vetcove.

## Acquire expert insights during 2024 Virtual Wednesday Round Tables

Much like the release of pent-up anticipation when a new season of your favorite streaming show drops, the wait is over for Season 4 of the AAEP's Virtual Wednesday Round Tables! The twice-monthly sessions on Zoom resumed March 13, offering members a double dose of online education and engagement with subject matter experts.

Available as a complimentary benefit of your membership on the second and fourth Wednesday of each month through October, the Virtual Round Tables are 90-minute virtual discussions similar to convention Table Topics. To participate in a particular session, simply register in advance through AAEP Anywhere by going to learn.aaep.org and clicking the Virtual Wednesday Round Tables banner.

Each month's Virtual Round Tables feature one clinical and one non-clinical topic. Following is the tentative schedule of upcoming sessions:

March 27 – Start Your Mobile Practice
April 10 – Interpreting Dental Radiographs


The 2024 season began March 13 with a session entitled The Lameness Exam: What Gives You Trouble? If you were unable to attend the live session, watch a recording on-demand through AAEP Anywhere. On-demand sessions are available approximately 48 hours later and include mentioned resources such as PowerPoint slides, images and more. CE credit is not offered.

*The AAEP thanks its Virtual Wednesday Round Table sponsor:*

CareCredit a Synchrony solution

## Hear from AAEP leadership on new Practice Life podcast


While the winter board meeting recap on page IV provides an overview of the AAEP's priorities for the current year, you can go a little deeper by listening to the February episode of the AAEP Practice Life podcast.

Entitled "AAEP Leadership: What's in Store for 2024," the episode features co-hosts Drs. Jessica Dunbar and Mike Pownall speaking with AAEP President Dr. Katie Garrett, President-Elect Dr. Tracy Turner and Executive Director David Foley. The topics range from governance issues and sustainability efforts to the 2024 annual convention and the anticipated launch of a wet labs initiative during the second half of the year.

The discussion included creation of a Governance Task Force comprised of members from diverse backgrounds to examine and recommend potential changes to ensure that AAEP's bylaws effectively serve the association and its members into the future. As Dr. Garrett notes, "The world changes, and we need to change with those times. Not just looking at things like how officers end up being officers but also things like the structure of the General Membership Meeting, how are bylaws actually able to be changed—can we modernize that so you don't have to be there in person at the General Membership Meeting, are we using appropriate communications channels to reach our members?"

Download or listen to the 28-minute episode on iTunes or at podcast.aaep.org.

THE ART OF HORSE
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*The AAEP Practice Life podcast is sponsored by Boehringer Ingelheim.*

## Strengthen the veterinary team-client partnership with new resource

Foster healthy lines of communication and a positive partnership between your staff and horse owners with the Veterinarian-Client Agreement for Effective Equine Care, presented on the next page.

Developed collaboratively by the AAEP's Horse Owner Education Committee and the AVMA, the one-page agreement seeks to strengthen the veterinary team-client partnership and encourage a supportive, safe and inclusive environment for all. The agreement helps set expectations between your team and clients in order to improve the veterinary experience for everyone.

We encourage you to download and share the agreement with your team along with the supporting rationale to encourage conversation. Then introduce the agreement to your clients by displaying the agreement prominently in your clinic, posting on your practice website and social media, integrating into new client welcome materials or other means.

Download the agreement to share with your team and your clients at aaep.org/node/38237.

## Consider 'going green' with digital *EVE*

If you are among the 62% of membership survey respondents in 2023 who signaled a willingness to forgo a print copy of *Equine Veterinary Education* in favor of a digital version, it's easy to make the switch. Simply update your preferences:

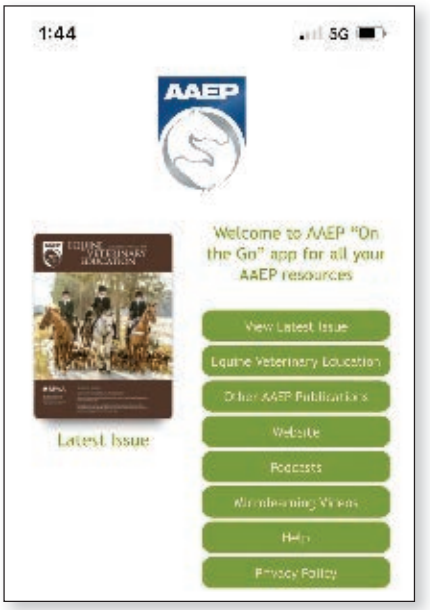
1. Go to aaep.org/dashboard
2. Select "Update Your Contact Information" (you may be asked to log in)
3. Scroll down and check the box beside "Do not send me a printed version of *Equine Veterinary Education*"

The digital edition of *EVE* is generally available on the AAEP website and On-the-Go app prior to its print counterpart dropping in the mail. Before making the switch, check out a recent digital issue at aaep.org/equine-veterinary-education or on the app. To download the app, search "AAEP On-the-Go" at your app store.

If you should ever change your mind and want to resume receiving a print issue, simply follow the previous steps and uncheck the box. A similar checkbox appears for Convention *Proceedings* if you would prefer to access the compilation of papers presented at the annual convention through the AAEP website or On-the-Go app in lieu of receiving a print copy.

Why consider switching? In 2023, over 76,000 copies of *Equine Veterinary Education* totaling more than 6.7 million pages were printed and mailed to AAEP members. Switching to digital would eliminate your paper consumption of more than 1,000 pages per year as well as emissions related to transit of print copies via ground and/or air to your mailbox.

Of course, if you like receiving a print copy of the journal in your mailbox every month, there is no need for you to do anything. We'll keep mailing it to you.




If you have questions about the digital AAEP publications, contact John Cooney, publications coordinator, at jcooney@aaep.org or (859) 233-0147.

# Veterinarian-client agreement for effective equine care

We are committed to cultivating beneficial relationships with our clients and patients. A strong partnership—rooted in mutual trust and respect—is essential to support the best possible care for all parties. Any behaviors that suggest differently will not be tolerated.

| AS A CLIENT, YOU CAN EXPECT TO: | IN RETURN, WE ASK THAT YOU: |
|---|---|
| Be treated with consideration, respect, and compassion by all members of our team in all communications and interactions. | Demonstrate consideration and respect towards all members of our team, other clients, and patients. Bring any concerns directly to us so we can work with you toward a solution. |
| Know who is providing your horse's care, and be assured that the provided care is appropriate and competent. | Accept that in certain cases your preferred veterinarian may not be available, and understand that anyone we trust to see your horse is qualified to do so. |
| Be presented with a range of care options that address your horse's needs. Be free to accept, decline, or discuss recommended options and their risks and benefits, and to seek a second opinion. | Allow us the opportunity to answer any questions you have about your horse's health status, recommended diagnostic or treatment options, or next steps if unclear. Follow the agreed-upon plan to the best of your ability, and contact us with your questions and any needs for assistance. |
| Be seen on time, or be notified of any delay, knowing that life-threatening illnesses or injuries will be prioritized over routine appointments. | Be ready on time with your horse appropriately restrained for your appointment, or call ahead if you're going to be late or need to reschedule or cancel. |
| Be informed of the estimated costs of veterinary services as well as available payment options, free of assumptions. | Meet agreed-upon financial responsibilities for provided veterinary services. Recognize that many veterinarians now require payment at the time of service for all clients. |
| Be provided with contact information for veterinary emergency services when needed. | Reserve after-hours calls for true emergencies. Understand that your regular veterinarian may not be the one to see your horse. |
| Be contacted in a timely manner, whether that involves answering questions, providing follow-up, or scheduling appointments. | Use the practice phone number and/or email to contact us, unless your veterinarian instructs you differently. |
| Have your horse treated with care and compassion. | Work with your horse on ground manners and basic restraint. This will minimize stress for your horse and promote safety for all. |
| Have us arrive with a properly equipped vehicle, stocked to treat most issues that your horse may have. | Provide us with a safe working area on your farm that is well lit, with running water and electricity whenever possible. |
| Be provided with veterinary care that complies with applicable laws and regulations. | Accept that veterinarians can only recommend treatments and prescribe drugs within a current veterinarian-client-patient relationship.* |
| Have medical and personal information held in confidence. | Provide in writing any individuals with whom information can be shared (e.g., multiple owners, trainer, barn manager, or insurance company). |
| Have accurate medical records maintained by us, and have copies or summaries provided to you on your request, in accordance with state laws. | Maintain detailed and easily accessible records of your horse's health and medical history, including vaccinations, deworming, and treatments provided outside our practice. |

**A veterinarian-client-patient relationship exists when your veterinarian knows your animal/facility well and recently enough to be able to treat your animal's medical condition, you have agreed to follow your veterinarian's instructions, and other requirements for this relationship have been met as defined by applicable federal and state law*

**NOTE:** Despite everyone's best efforts, things can go wrong. As partners in your horse's care, let's extend each other the grace to work through and learn from any issues, and continue to cultivate our partnership for the benefit of all horses.

*mcm-0124*

## Reward excellence with an AAEP award nomination

Deadline to nominate is June 1

Pay the ultimate compliment to a richly deserving colleague by nominating that individual for an AAEP annual award. The presentation of annual awards is a highlight of each year's President's Luncheon at the annual convention as those serving the horse and profession in outstanding ways are celebrated in front of hundreds of their peers.

The AAEP is accepting nominations in the following categories until June 1:

The **Distinguished Educator – Academic Award** honors an individual educator who by his or her actions and commitment has demonstrated a significant impact on the development and training of equine practitioners.

The **Distinguished Educator – Mentor Award** honors an individual who by his or her actions and commitment has demonstrated a significant impact on the development and training of equine practitioners through mentoring.

The **Distinguished Life Member Award** recognizes a member who has demonstrated outstanding or extraordinary service to the AAEP over the course of their career.

The **Distinguished Service Award** recognizes an individual who has provided exemplary service to the AAEP or a similar organization to the benefit of the horse, horse industry or the profession of equine veterinary medicine.

The **George Stubbs Award** recognizes the contributions made to equine veterinary medicine by individuals other than veterinarians.

The **Sage Kester Beyond the Call Award** is named in honor of its first recipient, the late General Wayne O. "Sage" Kester, DVM, and represents the highest honor bestowed by the AAEP upon a current or former member. The award is presented to an individual who has made significant and long-lasting contributions to equine veterinary medicine and the community.

The **Lavin Cup (The Equine Welfare Award)** honors a non-veterinary organization or individual that has demonstrated exceptional compassion or developed and enforced rules and guidelines for the welfare of horses.

The **AAEP Research Award** recognizes an individual who has recently completed research that has or will make a significant impact on the diagnosis, treatment or prevention of equine disease. Nominations are open to all individuals whose research is acknowledged by presentation or publication and by peer review as a significant advancement in equine medicine or innovation in equine science. Nominees must have had their research presented or published during the two years prior to when nominations are submitted to the AAEP.

Visit aaep.org/about-aaep/annual-awards for nomination forms as well as additional information about the awards and selection process. Nomination forms are also available from Sue Stivers at (859) 233-0147 or sstivers@aaep.org.

Award recipients will be honored at the AAEP's 70th Annual Convention in Orlando, Fla., Dec. 7–11, 2024.

*Dr. Ryland Edwards (left) accepts the Distinguished Educator – Mentor Award from 2023 AAEP President Dr. Rob Franklin during the 69th Annual Convention.*

## Digest latest colic research at international symposium

The 14th International Equine Colic Symposium will bring together equine veterinarians and researchers to exchange the latest information about colic over two-and-a-half days at Surgeons Quarter in Edinburgh, Scotland, July 10–12, 2024.

The triennial meeting is hosted alternately by the British Equine Veterinary Association and The Foundation for the Horse. The format is 12-minute oral presentations followed by three minutes for questions and discussion on topics that include, but are not limited to, surgical techniques, treatment, parasitology, gastric ulceration, endotoxemia, pharmacology of intestinal motility, complications and more. Beyond presentations, there will be plenty of opportunities for connection with other attendees during social events.

The registration rate is £475. For more information or to register and secure housing, visit http://tinyurl.com/2enbcy9z.

## Patient and personal well-being share spotlight at Resort Symposium

Sun and science were on the syllabus when 150 practitioners from across North America and as far away as New Zealand gathered in Costa Rica Jan. 22–24 for education and memory-making at the AAEP's 25th Annual Resort Symposium.

Three half-day educational sessions equipped practitioners with the latest knowledge to treat, manage and prevent equine gastric ulcer syndrome; effectively image the stifle, digital tendon sheath, cervical spine and back; and treat and rehabilitate horses with neck and back pain. Attendees also explored the surgical options for kissing spine, stifle issues and tendon sheath injuries as well as when and how to use orthobiologic therapies.

After sessions, attendees relaxed, recharged and strengthened bonds while basking in the warm Pacific sun poolside, beachfront or on group excursions with colleagues.

If bucket-list travel and uniquely casual CE sounds like a perfect pairing to start a new year, mark your calendar for the 26th Annual Resort Symposium, which will be held Jan. 20–22, 2025, at the Four Seasons Resort in Maui, Hawai'i. Additional information about the meeting will be announced later this year.

*Attendees enjoy the camaraderie of colleagues at the Welcome Reception.*

*The AAEP thanks Zoetis for its ongoing sponsorship of the annual Resort Symposium.*

## Welcome to new AAEP members!

We are delighted to welcome the following practitioners who joined the AAEP between Oct. 1–Dec. 31, 2023:

Justin Adam, DVM, Santa Maria, CA
Morgon Allbaugh, VMD, Starkville, MS
Kelly Anders, DVM, Eaton, CO
Fabio A. Aristizabal Mena, DVM, Scottsdale, AZ
Rebecca D. Armstrong, DVM, PhD, Castro Valley, CA
Zachary Arnold, DVM, Guthrie, TX
Michelle Elizabeth Bakker, BVMS, Bonsall, CA
Alexis Morgan Baney, DVM, Greensburg, PA
Clayton Barton, DVM, Hurricane, UT
Tad Bender, DVM, Escondido, CA
Mauricio José Bittar, DVM, Rio Claro/SP, São Paulo, Brazil
Larissa Bokan, DVM, Bement, IL
Jason Brownridge, DVM, Wellesley, ON, Canada
Lorna Marion Bryce, DVM, Columbia, MO
Cecilia Calestani, DVM, Wellington, FL
Elisa Holthausen Caminoto, DVM, PhD, Sao Jose Do Rio Preto, Brazil
Gustavo Ferrer Carneiro, DVM, Jaboatão, Pernambuco, Brazil
Carlos Carvajal de La Cerda, MVZ, Lexington, KY
Brent N. Cassady, DVM, Atlanta, GA
Carolina Cervera Torres, MVZ, Stephenville, TX
Andraya Cole, DVM, Deerfield, MA
Katie Comerford, DVM, Mechanicsburg, PA
Carina Joppe Cooper, DVM, PhD, Sherwood Park, AB, Canada
Casey Cooper, DVM, Three Forks, MT
Carissa Coulson, DVM, Allegan, MI
Madeline Courville, DVM, Wilton, CA
Jason R. Crawford, DVM, DACVPM, Kensington, MD
Erin Culligan, DVM, Duchess, AB, Canada
Sabrina Dagher-Arevalo, DVM, Fort Sam Houston, TX
Rachel Davis, DVM, North Vernon, IN
Katie Chanel De Jong, DVM, Halls Creek, NSW, Australia
Marcos Guilherme De Souza, DVM, Millsap, TX
Dustin Dorris, DVM, Austin, TX
Benjamin Bastien Dubois, DVM, Ranson, WV
Ghislaine Dujovne, DVM, Davis, CA
Janea Eberly, DVM, Albuquerque, NM
Dominic Eickert, DVM, Columbus, NC
Lindsey Elster, DVM, Amanda, OH
Hernan Guerrero, DVM, Cuidad de La Costa, Uruguay
Selene Guillén, DVM, Calexico, CA
Ricardo X. Gutierrez Cruz, MVZ, Naucalpan, Estado, DM, Mexico
Alfredo Guzman, DVM, Chula Vista, CA
Nicola Hardgrove, DVM, Hermosa Beach, CA
Babiche Alida Heil, DVM, Pullman, WA
Kaylin Henry-Moses, DVM, Ocala, FL
Kate Huggler, DVM, Penfield, NY
Hafiz M. Taimoor Ihsan, DVM, Bonsall, CA
Taylor Isberner, DVM, Ankeny, IA
Monica Ayon Jaime, DVM, Calexico, CA
Friedrich Janetzko, DVM, Hannover, Germany
Martyna Malwina Jargielo, DVM, Lafayette, IN
Kate Johansen, DVM, Tulsa, OK
Carlin Jones, DVM, Selkirk, NY
Thomas Kanach, DVM, Avon, IN
Molly Kearney, BVSc, MRCVS, Corvallis, OR
Nathan Keefer, DVM, Petaluma, CA
Travis Kelter, DVM, Calgary, AB, Canada
Anna Kucera, DVM, San Antonio, TX
Sandra Kurras, DVM, Bakum, Germany
Claudia Michell Lefranc Lopez, MVZ, Monterrey, Nuevo León, Mexico
Daniela Lemos, DVM, Krum, TX
Renzo Leoncini, DVM, Palaia, Italy
Heather Lerseth-Fliehs, DVM, Groton, SD
Jing Li, DVM, Beijing, China
Lisa Maria Lidbeck, DVM, Lund, Skåne, Sweden
Marco Livini, DVM, Milan, Milano, Italy
Antonio Luciani, DVM, San Lazzaro Di Savena, Italy
Katie Luoma, DVM, Forest Lake, MN
Tiffany Lynn, DVM, Johnson City, TN
Meagan MacDiarmid, DVM, Napan, NB, Canada
Fernanda Machado, DVM, Azusa, CA
Margo Machen, DVM, PhD, Upland, CA
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Sheila McDonald, DVM, Kelowna, BC, Canada
Rachel McEwen, DVM, Halifax, NS, Canada
Walter-John Howard McGowan, DVM, Spring, TX
Katherine Michele Miley, DVM, Germantown, NC
Travis Miller, DVM, Vinton, LA
Percy Flores Modomo, DVM, Makati City, Metro Manila, Philippines
Kate Moeller, DVM, The Dalles, OR
Jessica Morgan, DVM, PhD, DACVSMR, Woodland, CA
Tyler Mort, MV, Ghent, NY
Hailey Mueller, DVM, Independence, KS
Joan M. Murnane, DVM, PhD, McLouth, KS
Azalea Navarro, DVM, North Riverside, IL
Brad Nelson, DVM, PhD, Fort Collins, CO
Tyler Newton, DVM, Bend, OR
Jared Anthony Nyman, DVM, Redmond, OR
Melissa Ochoa, DVM, Show Low, AZ
Mark O'Connor, DVM, Bozeman, MT
Cameon Ohmes, DVM, De Soto, KS
Carla Olave, DVM, West Lafayette, IN
Sarah Jeanne Osborne, DVM, Panama, NY
Ana Paula Monteiro Pacheco, DACVIM, Corvallis, OR
Summer Pepper, DVM, Norco, CA
Lauren Platt, DVM, Highlands Ranch, CO
Rijk-Jan Pleijter, DVM, Oosteind, N-Brabant, Netherlands
Jessica Ragauskas, DVM, MPH, Prebbleton, New Zealand
Jose Manuel Ramirez Uribe, MVZ, Lomas del Huizachal, Naucalpan, Mexico
Abigail Reising, DVM, Wheaton, IL
David Rendle, BVSc, Tiverton, United Kingdom
Kacy Riley, DVM, Pittsburg, TX

## Members in the News

### Dr. Julie Dechant receives distinguished teaching award

*Dr. Julie Dechant (left) receives her award from Dr. Joie Watson.*

Dr. Julie Dechant, clinical faculty, equine surgical emergency and critical care in the department of surgical and radiological sciences at the University of California, Davis, received the 2023 Zoetis Distinguished Veterinary Teacher Award.

Dr. Dechant received her veterinary degree from the University of Saskatchewan. In 2004, she joined the UC Davis faculty where she teaches all four years of the DVM curriculum and is assistant director of the large animal clinic. Additionally, Dr. Dechant serves as the faculty advisor for the school's AAEP student chapter and the Camelid Medicine Club.

### Dr. Kenton Morgan named Missouri Veterinarian of the Year

*Dr. Kenton Morgan*

The Missouri Veterinary Medical Association presented its 2024 Veterinarian of the Year Award to MVMA executive board member Dr. Kenton Morgan, who in 2023 retired as managing veterinarian, equine technical services with Zoetis.

Dr. Morgan received his veterinary degree from the University of Missouri and has been an active advocate for organized veterinary medicine throughout his career. He served on the AAEP's board of directors from 2013–2015 and as chair or member of numerous councils and committees, culminating in his receipt of the Distinguished Life Member Award in 2023.

### Three members appointed to Louisiana State Racing Commission

*Dr. Larry Findlay Sr.*

*Dr. Travis Miller*

Drs. Patrick Bernard, Larry Findlay Sr. and Travis Miller have been appointed to the Louisiana State Racing Commission by Governor Jeff Landry.

Dr. Bernard is an AAEP Honor Roll member and past president of the Louisiana Veterinary Medical Association. He received his veterinary degree from Auburn University and served on the Pediatrics, Political Liaison and Research committees in the early 1990s. *Photo unavailable.*

Dr. Findlay is the founding owner of Delta Equine Center in Vinton, La. He currently serves on the AAEP's Racing Committee and previously served on the Pediatrics Committee. Dr. Findlay received his veterinary degree from Louisiana State University.

Dr. Miller practices at Acadiana Equine Hospital in Opelousas, La. A veterinary graduate of Tuskegee University, Dr. Miller interned at Peterson Smith Equine Hospital in Ocala, Fla., prior to joining his current practice.

## Welcome new members, continued

Bethany Marie Roof, DVM, Columbia, MO
Caroline Rossner, DVM, Phenix, VA
Amy Santonastaso, VMD, Blacksburg, VA
Kazushige Sato, DVM, Hidaka-gun, Hokkaido, Japan
Laura Schapman, DVM, Almont MI
Svea Marie Schmidt, DVM, Stromberg, RLP, Germany
Juliano S. Schmitt, DVM, Lemoore, CA
Anna Rose Schultz, DVM, Salida, CO
Kerstin Schwichtenberg, DVM, Agassiz, BC, Canada
Kathleen Shaughnessy, VMD, Newberg, OR
Zoe Shelton, DVM, The Dalles, OR
Courtney Sherman, DVM, Cowan, TN
Kat Sippel, DVM, Perrysburg, OH
Holly N. Smith, DVM, Lynden, WA
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Christina Eiry Spence, DVM, Edson, AB, Canada
Isabell Stamm, DVM, Ponoka, AB, Canada
Ali Sturtevant, DVM, Wentzville, MO
Michael Swagemakers, DVM, Sint Oedenrode, Noord-Brabant, Netherlands
Veronica Tamez Castro, MVZ, Chino Hills, CA
Christina Thompson, DVM, Yucaipa, CA
Taylor Tuso, DVM, Ocala, FL
Pete R. Van Dyke, DVM, Walla Walla, WA
Andrew van Eps, DVM, Kennett Square, PA
Jana Vanaga, VMD, Jelgava, Jelgavas novads, Latvia
Gustavo Vautier, DVM, Ocala, FL
Israel Vazquez Lopez, MVZ, Mexico City, DIF, Mexico
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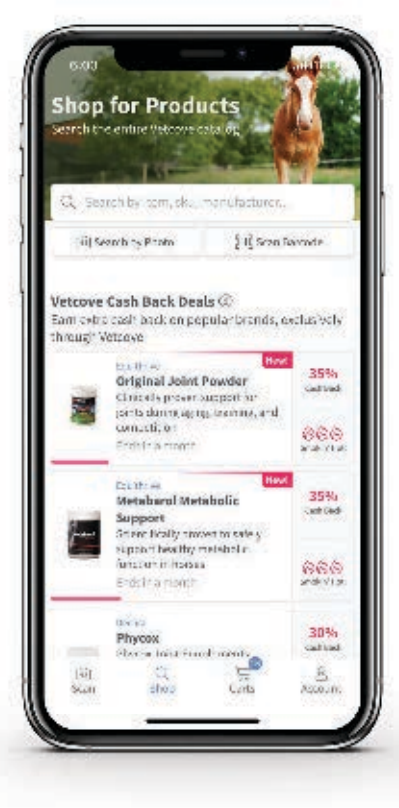


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INDUSTRY

## AAEP Educational Partner Profile: Dechra Veterinary Products

Dechra Veterinary Products has emerged as a leader in equine medicine by offering a specialized range of approved equine products. Dechra has just launched **Zycosan® (pentosan polysulfate sodium injection)**, the first FDA-approved pentosan to the market. Some of Dechra's other niche products include **OSPHOS® (clodronate injection)**, **Zimeta® (dipyrone injection)** and **SucroMate® Equine (deslorelin acetate)**.

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Innovations in equine regenerative medicine are an important focus for our well-known brands, **Orthokine® vet irap** and **Osteokine® PRP**. In addition to this line, Dechra also markets **Equidone® Gel (domperidone)** and **Phycox® EQ Granules Joint Supplement** containing the patent ingredient, phycocyanin. In the fall of 2021 Dechra launched the **Provet™ APC system** which can process PRP in as little as 3 minutes with a convenient, stall-side centrifuge weighing only 4.4 pounds.

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Received: 6 December 2023 | Accepted: 6 December 2023

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Equine Veterinary Education BEVA

RESEARCH HIGHLIGHTS

# Highlights of recent clinically relevant papers

## STANDING FRACTURE REPAIR OUTCOMES

*This study by Victoria Colgate and co-workers in the United Kingdom aimed to determine short-term (survival to discharge) and long-term (return to racing) outcomes of horses undergoing standing repair of metacarpal/metatarsal III (MC/MTIII) and proximal phalanx (P1) fractures, and to compare pre- and post-surgical racing performance.*

Clinical records of 245 horses undergoing standing repair of MC/MTIII or P1 fractures were reviewed. Data on signalment, fracture configuration and complications were collected, and full race records were retrieved from the Racing Post Database. Chi-squared and Mann–Whitney *U* tests were used to determine any difference in variables between horses that raced after surgery compared to those that did not. McNemar change and Wilcoxon signed-rank tests were used to compare pre- and post-surgical racing performance.

The proximal phalanx was fractured in 41.2% (101/245) of cases and 58.8% (144/245) had a condylar fracture. Of the repaired fractures, 64.9% (159/245) were bicortical and 35.1% (86/245) unicortical fissure fractures. Ninety-eight per cent of horses survived to hospital discharge, and 75.1% raced after surgery, a median of 241 days later. Horses that raced post-surgery were significantly less likely to have suffered from complications during hospitalisation than those that did not race again (17.3% vs 36.5%). Comparing pre- and post-operative racing performance, there was no significant difference in earnings per start (median £628.00, interquartile range [IQR] 115.90–1934.80 vs £653.20, 51.00–1886.40) or proportion of horses winning (51%; 95% CI 41%–61% vs 54%; 95% CI 44%–64%), or being placed first–third (77%; 95% CI 68%–85% vs. 71%; 95% CI: 62%–80%) in at least one race.

Standing fracture repair is a viable treatment option for MC/MTIII or P1 fractures that returns horses to the racetrack within an acceptable time frame and can restore pre-surgical athletic ability.

## RADIOGRAPHIC ABNORMALITIES OF THE SPINE

*In this study, Maria Pressanto and co-workers in Italy and the United Kingdom investigated the prevalence and severity of radiographic abnormalities of the interspinous spaces (ISSs) in the thoracolumbar vertebral column of unbroken yearlings and compared these findings with a group of older trained Thoroughbred horses without perceived back pain.*

A total of 102 horses were included (47 yearlings and 55 trained horses). Each horse underwent a digital radiographic study of the thoracolumbar vertebral column (T7-L3) and each space graded for narrowing of the ISSs, increased opacity, radiolucency and modelling of the cranial and caudal margins of two contiguous dorsal spinous processes (DSPs). This generated both an individual anatomical space score for each space and a total score for each horse for subsequent comparison.

Narrowing and impingement were detected in a third of the examined ISSs. DSP increased opacity, radiolucencies and modelling were found in over half of the yearlings. The median total score per horse was 33 (0–96) in the yearlings and 30 (0–101) in trained horses, indicating no significant difference in radiographic abnormalities. Likewise, the median total score per anatomical space was 112 (25–259) and 127.5 (24–284) in the yearlings and trained horses. No differences were found between the groups for number of radiographic abnormalities, scores and total score.

This study reported the incidence of DSP radiographic abnormalities in Thoroughbred horses. The absence of difference in occurrence between yearlings and older horses supported a developmental rather than acquired aetiology.

## PASSIVE TRANSFER FAILURE TESTING

*This study by Sevim Kasap and co-workers in Turkey compared the diagnostic performance of serum glutaraldehyde coagulation test (GCT) and colostrum BRIX% to detect failure to transfer passive immunity (FTPI) diagnosis with the results of the SNAP foal test; and evaluated the results of serum GCT and colostrum BRIX% measurements in foals that developed diarrhoea in the 0–1-month period.*

Residual serum and colostrum from 298 samples collected from newborn foals and their dams for clinical purposes were used. Foals were classified as FTPI positive (IgG < 8 g/L) or negative (IgG ≥ 8 g/L) using the SNAP foal test. The sensitivity and specificity of serum GCT and colostrum BRIX% for diagnosing FTPI in all foals were compared. The relationships between the results of the serum GCT and colostrum BRIX% and the development of diarrhoea in the 0–1-month period in foals with and without FTPI were evaluated.

Serum GCT and colostrum BRIX% were significantly different between the foals classified according to the SNAP test as with and without FTPI. Using a cut-off value for serum GCT of >10 min, the sensitivity for the detection of FTPI was 100% (95% CI 92.9%–100%)

and specificity 100% (98.3%–100%) while using BRIX% a cut-off value of ≤24%, the sensitivity was 92% (80.9%–97.8%) and specificity was 98% (95.3–99.3). In the subgroup of foals without FTPI using a colostrum BRIX% cut-off value of ≤26, the sensitivity for prediction of diarrhoea in the 0–1 month period was only 72.4% (52.8–87.3) with specificity 54.3% (47.6–61.1) but the test performance was not robust (ROC AUC 0.61).

Using serum GCT (>10 min), and colostrum BRIX% (≤24%), both economical and practical to use in the field, gave results comparable with the SNAP foal IgG test for the detection of FTPI (<8 g/L). The ability to accurately predict diarrhoea in the first month of life with these tests was limited.

## EFFECTS OF POLL FLEXION ANGLES

*In this study, Paula Tilley and co-workers in Portugal aimed to evaluate the effects of two riding poll flexion positions with a difference of only 15° on the respiratory systems and behaviour of horses through an evaluation of dynamic airway collapse* via *over-ground endoscopy, the pharyngeal diameter, pleural pressure, arterial oxygenation and lactate, heart and respiratory rates (HR/RR) and the occurrence of conflict behaviours*.

Twenty high-level dressage and 20 show-jumping horses underwent a 40-min ridden test at a ground angle of 85°; 3 weeks later, they underwent a ridden test at a 100° ground angle (the angle between the ground and the line from the forehead to the muzzle) and in a cross over design. Using a mixed model for repeated measures, Wilcoxon/Friedman tests were carried out according to the experimental design and/or error normality. For both groups, at 100°, conflict behaviours and upper airway tract abnormalities were more frequent, the pleural pressure was higher, and the pharyngeal diameter was lower. At 85°, relaxation behaviours were more frequent. Lactate was higher at 100° only in the dressage horses. Compared to the first test at 85°, the HR/RR were lower at the beginning of the second test (at 100°) but higher at the end.

The differences identified in these dressage and show-jumping horses support the idea that an increase of just 15° in riding poll flexion can have negative effects on the respiratory system and behaviour of a horse and therefore on its welfare.

## MEDIUM-TERM OMEPRAZOLE TREATMENT

*In this study, Bethanie Clark and co-workers in Australia and Hong Kong evaluated changes in serum gastrin and chromogranin A (CgA) concentrations in response to medium-term (57-day) omeprazole treatment and after omeprazole discontinuation*.

Fourteen mature Thoroughbred racehorses in simulated race training received 2.28 g of oral omeprazole PO q24h for 57 days within a 61-day period, excluding a withholding period applied mid-protocol during which treatment was stopped as part of a concurrent study. Serum samples were collected on day 0 before omeprazole treatment, on day 1 of each week of the treatment period, and for an additional 5 weeks after discontinuation of treatment. Serum gastrin and CgA concentrations were analysed using radioimmunoassay (RIA) and ELISA respectively.

Median serum gastrin concentrations increased 2.5-fold from baseline to day 7 but did not increase further during the omeprazole treatment period. Median serum gastrin concentrations returned to baseline within 2–4 days after administration of the last dose of omeprazole. No effect of treatment or discontinuation was seen in serum CgA concentrations.

Sue Wright

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Received: 22 June 2022 | Accepted: 25 April 2023

DOI: 10.1111/eve.13819_1

Equine Veterinary Education

CASE REPORT

# Pan-carpal arthrodesis as a treatment for distal radial fracture in a pony

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## SUMMARY

Complete, displaced radial fractures carry a poor prognosis in adult horses. In foals and ponies, fracture repair may have a good outcome following internal fixation. Distal radial fractures are difficult to stabilise due to limited bony purchase. Therefore, carpal arthrodesis may provide a viable option for salvaging the horse. In this report, we describe pan-carpal arthrodesis as a treatment for distal radial fracture in an adult pony. A 12-year-old pony, 180 kg bodyweight, was presented due to left front non-weight bearing lameness. Diagnosis of a closed, comminuted, displaced distal radial fracture, with antebrachiocarpal joint involvement was made radiographically (Figure 1). Due to limited bony purchase in the distal radius, incomplete reduction during surgery and high load on the fracture line, pan-carpal arthrodesis was performed, by placing 14 and 15-hole broad 5.5 mm locking compression plates on the dorsolateral and dorsomedial aspects of the limb. Recovery was uneventful. The implants were subsequently removed in a minimally invasive standing procedure, due to surgical site infection. Removal was performed on two separate occasions, three and four months post-operatively. Two years following surgery, the skin is intact and without signs of infection. The horse is sound at a walk and trots freely. Carpal flexion was pain-free and the range of motion was limited to approximately 55 degrees. Radiographs revealed adequate healing of the fracture and failure of the arthrodesis. In essence, unintentionally, we performed temporary trans-articular stabilisation by repairing a distal radial comminuted fracture, while preserving partial carpal range of motion. This technique, of temporary trans-articular stabilisation, could potentially be further modified and applied intentionally on other distal or proximal long bone fractures in horses.

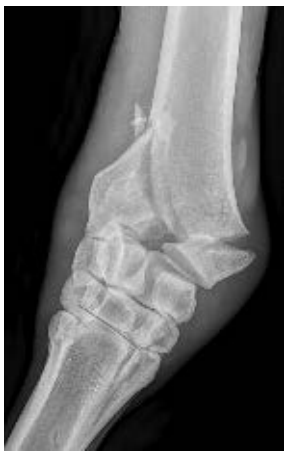

**FIGURE 1** Dorsopalmar projection of left front limb radiographs taken on admission demonstrating a comminuted, displaced and unstable distal radial fracture.

**KEYWORDS**
horse, radial fracture, internal fixation, carpal arthrodesis, temporary trans-articular stabilisation

### Key points

- Distal radial fractures carry a poor prognosis in the adult horse due to difficulty in stabilisation and limited bony purchase.
- Temporary trans-articular stabilisation is a feasible technique for distal or proximal long bone fracture in horses, while preserving the range of motion of adjacent joints.
- Presence of infection crucially inhibits the fracture healing process.

Presented in part, as a poster at the annual summit of the European College of Veterinary Surgery, 2021.

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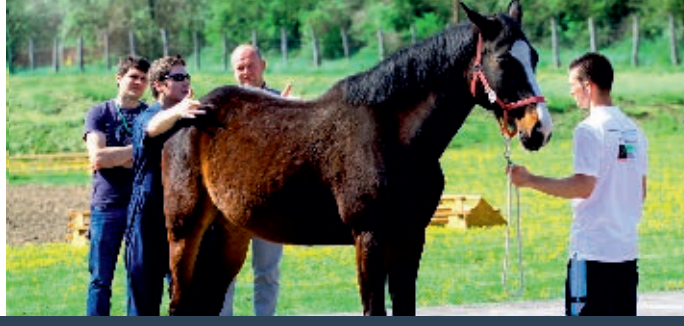

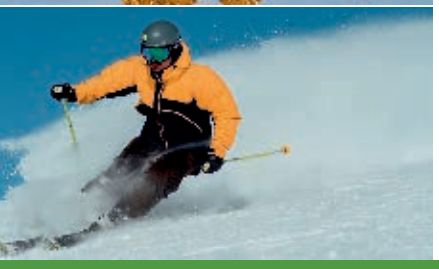

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**Mar 22 - 23** **Ultrasound of the Mid-Distal Limb & Stifle for Equine Practitioners**
Chicago Equine Medical Center, IL

**Apr 5 - 6** **Sports Horse Medicine & Orthopedics**
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**Jun 7 - 8** **New Developments in Equine Endoscopy & Respiratory Conditions**
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Received: 17 July 2023 | Accepted: 17 July 2023

DOI: 10.1111/eve.13871

Equine Veterinary Education BEVA

CLINICAL COMMENTARY

# Distal radial fracture repair in adult horses

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Multifragmentary and comminuted distal metaphyseal fractures are the most common radius fracture configurations in adult horses (Auer, 2020a, 2020b). Repair of these complex fractures is rarely attempted as the prognosis is generally unfavourable (Auer & Watkins, 1987). Achieving adequate fracture reduction and a stable construct are particularly challenging. In foals, however, fractures of the radius occur most frequently as transverse or short oblique fractures in the mid-diaphyseal region (Watkins, 2006). As these fracture configurations are more reliably reconstructed and the smaller case size is more suited to the available implants, foals are better candidates for internal fixation. Complex distal radius fractures in adult ponies may be associated with a more favourable prognosis because of their low bodyweight.

Following anatomical reconstruction, long-bone fractures in horses are most commonly stabilised using orthogonal double plate fixation with one plate placed on the tensile surface of the bone (Nixon et al., 2020). Surgeons rely on the implants and repaired bone to share responsibility for transmitting forces through the construct. To ensure this, fractures must be fully reduced and repaired with adequate bone purchase in the fracture fragments. Multifragmentation and comminution add another dimension of difficulty to the repair (Auer & Watkins, 1987). Undeniably, small fragments, which cannot be stabilised with lag screws, as well as fragments located in the cortex subjected to compressive loads, increase the risk of construct failure (Watkins, 2006). Difficulties in long-bone fracture repair arise when a fracture segment is insufficient in length to allow adequate purchase using standard implants (Watkins, 2006). In the case of distal metaphyseal fractures and especially when multifragmentation is present, bone purchase in the distal epiphysis of the radius is limited and deemed insufficient to achieve adequate stability. The authors of the accompanying case report, therefore, decided to include the third metacarpus and carpal bones in the construct and thereby inevitably perform a pancarpal arthrodesis (Haddad & Kelmer, 2024).

Two 5.5 mm locking compression plates (LCPs) were used to bypass the area of the fracture and transmit the shear, bending and rotational forces from the healthy segment of the radius to the third metacarpus (Auer, 2018; Nixon et al., 2020). The chosen plates were fixed to the bone with one locking head screw at each extremity of the plates, prior to lag screw fixation of the fracture. Locking head screws lock directly into the plate and do not rely on the friction between the plate and the bone to achieve stability (Levine & Richardson, 2007). Starting an LCP plate fixation with two locking head screws does not compress the plate to the bone. Hence, the construct acts as an internal fixator, biomechanically similar to an external fixator placed under the skin.

Locking compression plates have recently been introduced as an alternative to external fixation with satisfactory outcome (Woon et al., 2010; Xiao et al., 2016). Biological bridge plating has been used in human surgery for highly comminuted fracture of the distal femur (Singh et al., 2018). This technique uses the plate as an extra medullary splint fixed to the two main fragments. The complex fracture zone is virtually left untouched; however, it is bridged by the plate. Length, alignment and rotation are restored, but anatomical reduction of each fragment is not attempted. This concept combines the relative stability provided by the plate with the preservation of natural fracture biology to achieve rapid callus formation and fracture consolidation. Bridge plating techniques are applicable to all long-bone fractures with complex fragmentation and conventional plate fixation is not suitable. We have used this concept of biological fracture fixation for treatment of highly comminuted fractures of the proximal phalanx in horses (Rossignol, 2022) (Figure 1). The LCP distal femoral plate is used for double arthrodesis of the proximal interphalangeal joint and metacarpo/tarso phalangeal joint, based on biological bridge plating.

In the accompanying case report, although not being the authors' initial intent, the medial and lateral plates were removed at 3 and 4 months, respectively, because of implant infection (Haddad & Kelmer, 2024). The high-motion radiocarpal joint remained mobile while the intercarpal joints were partially fused. The chosen double plate construct ended up serving as a temporary transarticular bridging construct providing adequate stability to enable fracture healing. In human medicine, temporary transarticular bridging is recommenced for cases suffering from complex periarticular and intra-articular fractures (Poulsen et al., 2023). The standard surgical approach for most of these fractures has traditionally been open reduction internal fixation with transarticular

  

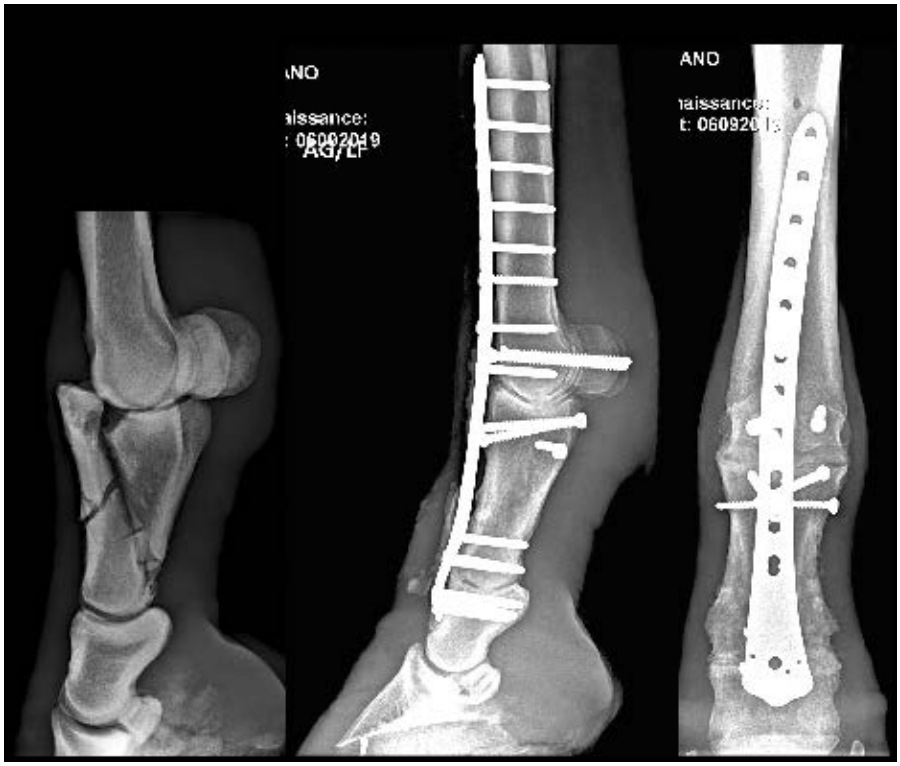

**FIGURE 1** Use of the locking compression plates (LCP) distal femur plate as a bridging plate to treat comminuted fracture of the proximal phalanx.

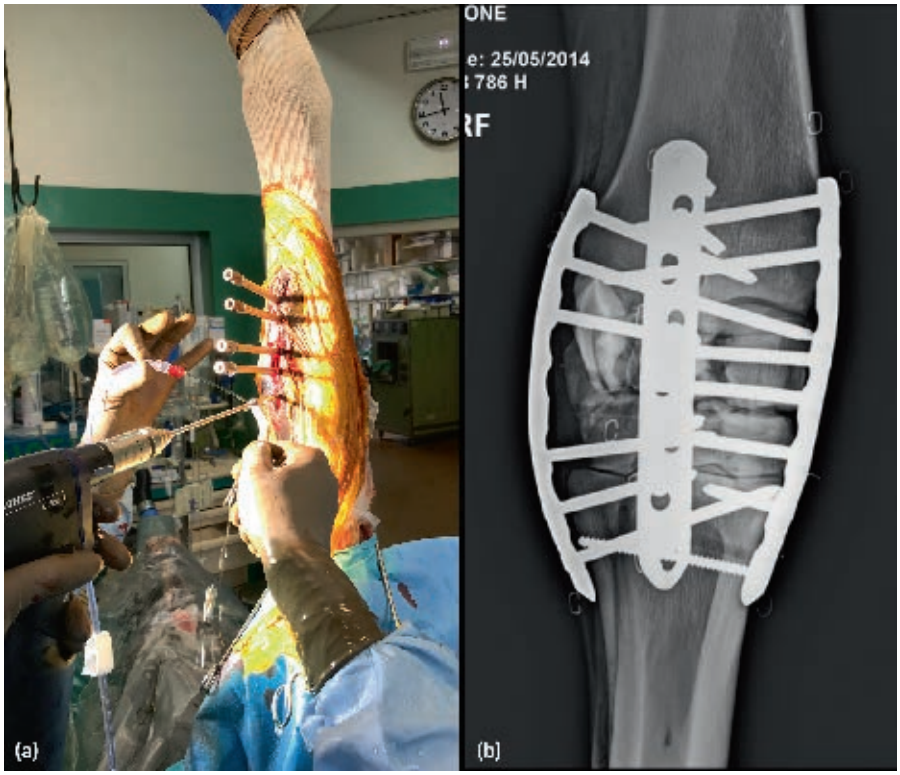


**FIGURE 2** Pancarpal arthrodesis using three short locking compression plates (LCPs) placed using a minimal invasive technique. (a) Peroperative view. (b) Final X-ray.

screws, which inevitably causes articular surface damage leading to various degrees of osteoarthritis. Temporarily stabilising the joint while sparing the subchondral bone and articular cartilage, reduces the risk of developing osteoarthritis. The case is estimated to regain natural joint motion following implant removal (Poulsen et al., 2023). Anatomic restauration of the joint surface prior to fracture stabilisation is of course mandatory to prevent development of severe osteoarthritis. The incidence of symptomatic osteoarthritis using this technique is low (Poulsen et al., 2023). If painful osteoarthritis develops or the preserved range of motion is severely reduced, performing an arthrodesis may become mandatory. In the accompanying case report, the case would have been expected to develop osteoarthritis since cartilage debridement had been performed in the antebrachiocarpal and middle carpal joints. The pony is, however, sound at walk, which does not justify any further intervention (Haddad & Kelmer, 2024).

Pancarpal carpal arthrodesis is a salvage procedure that is performed in cases of carpal instability due to complex fractures or end-stage osteoarthritis (Curtiss et al., 2018). Successful surgery has been described using two LCP plates placed, similarly to the accompanying case report, along the dorsomedial and dorsolateral aspect of the limb, on either side of the extensor carpi radialis and centred on the carpus (Carpenter et al., 2008). A more recent technique using three short LCP plates allows more complete engagement of the three relevant carpal bones within each row, increasing the stability of the construct in both plans, potentially more efficient in the latero-medial plan (Curtiss et al., 2018). The plates are short (6–8 holes), which prevents the stress from being concentrated at the plate ends, over the mid-diaphysis of the radius and third metacarpal bone (Figure 2).

Extensive cartilage debridement is absolutely mandatory for successful arthrodesis, especially in acute trauma cases with healthy joint surfaces. Articular cartilage should be debrided through the calcified cartilage layer, exposing the subchondral bone to enable rigid osseus bone union once the joint is stabilised in the weightbearing position (Zubrod & Schneider, 2005). As much articular cartilage as possible is removed arthroscopically from the radiocarpal and middle carpal joints using a combination of manual (bone curette) and motorised (arthroscopic burr) instruments. Alternatively, the cartilage can be debrided blindly through stab incisions using a bone curette. A 4.0 or 4.5 mm drill bit is used to drill across the carpometacarpal joint in multiple directions to remove the articular cartilage (Curtiss et al., 2018). While ankylosis can occur naturally in some low-motion joints, spontaneous bony bridging of the articular surface in high-motion joints, such as the radiocarpal and carpometacarpal joints, is very rare and relies on extensive cartilage debridement and rigid stabilisation to be successful (Zubrod & Schneider, 2005).

Performing pancarpal arthrodesis using a minimally invasive approach can help to prevent wound closure complications and reduce the risk of infection (Curtiss et al., 2018). As noted in the accompanying case report, open reduction and internal fixation through multiple large incisions can result in excessive skin tension when suturing (Haddad & Kelmer, 2024). The mechanical characteristics of LCP plates facilitate minimally invasive osteosynthesis because careful contouring and plate bone contact are not essential to ensure construct stability, especially when placed following the rules of an external fixator (Nixon et al., 2020). The authors of the accompanying case report chose to use two 5.5 mm LCP plates. These implants were developed exclusively for use in horses (Auer, 2020a, 2020b). The 5.5 mm and broad 4.5 mm LCP plate have the same width but the 5.5 mm LCP plate is thicker (6 vs. 5.2 mm). Most surgeons are reluctant to use the 5.5 mm LCP plate for distal limb fracture repairs in anticipation of difficulties to cover the implants during closure. Fracture healing relies on a favourable interaction between the stability of the fracture fixation construct and the biological environment provided by the soft tissues enveloping the fracture (Watkins, 2006). If the biological environment is substantially compromised, fracture healing is unlikely. Infection is the most common biological limitation to fracture healing in the equine case. It is a consequence of bacterial contamination resulting from a penetrating injury at the fracture site or acquired during open reduction and internal fixation (Watkins, 2006). Since infection is directly related to the degree of soft tissue injury and surgery time, strenuous closure is a substantial risk factor.

Fracture fixation of multifragmentary distal radial fractures is challenging because it requires a particularly stable construct even though bone purchase is very limited. If the articular surface can be fully reconstructed and the fracture reduced with lag screws, using a temporary external fixation construct or neutralisation plating to stabilise the fracture and protect the lag screw fixation may be an option. Otherwise, a pancarpal arthrodesis should be performed to restore comfortable weightbearing on the limb. Even though the authors of the accompanying case report did not initially intend to perform a temporary fixation of the fracture, infection, requiring early removal of the implants and failure of the radiocarpal joint fusion led to successful healing of the fracture while preserving part of the carpal range of motion. These kinds of 'happy accident' have often happened in the history of science and medicine and have played a role in innovation.

## AUTHOR CONTRIBUTIONS

F. Rossignol and A. Campos both contributed to the preparation of the manuscript and gave their final approval of the manuscript.

## CONFLICT OF INTEREST STATEMENT

No conflicts of interest have been declared.

## FUNDING INFORMATION

No funding received.

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**How to cite this article:** Rossignol, F. & Campos, A. (2024) Distal radial fracture repair in adult horses. *Equine Veterinary Education*, 36, 117–120. Available from: https://doi.org/10.1111/eve.13871

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**Reference:
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Received: 25 July 2022 | Accepted: 7 June 2023

DOI: 10.1111/eve.13852_1

Equine Veterinary Education BEVA

CASE REPORT

# Suspected idiopathic vestibular syndrome in the horse: Eight cases (2014–2021)

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## SUMMARY

A descriptive case series of the presentation, diagnosis, treatment and outcome of eight horses presenting to a UK equine veterinary hospital with suspected idiopathic peripheral vestibular syndrome over a 7-year period (2014–2021). During the 7-year period (between 2014 and 2021), eight horses were diagnosed with suspected unilateral IVS. The median age of presentation was 11 years (range 7–16) and there were 5 geldings and 3 mares. The median duration of clinical signs before presentation was 5 days (range 1–21) and most horses presented with an acute onset of clinical signs that did not worsen with time. Idiopathic vestibular syndrome (IVS) in the horse causes unilateral clinical signs including; head tilt (7/8), pathological strabismus (7/8), nystagmus (6/8), ataxia (6/8) and ipsilateral facial nerve dysfunction (5/8). Diagnosis is aided by neurological examination, endoscopy of the upper airway and cross-sectional imaging (i.e. magnetic resonance imaging and computed tomography). Computed tomography of the head was performed in every case (8/8). Additional diagnostic tests included upper respiratory tract (including guttural pouch) endoscopy (6/8) radiography of the head (1/8) or cervical region (1/8) and analysis of cerebrospinal fluid (1/8). There were no apparent abnormalities detected on any of these tests. Treatment varied between cases including; prednisolone (3/8), phenylbutazone (1/8), physiotherapy (2/8) or none (3/8). Horses were stabled during hospitalisation but turnout was encouraged following discharge. Long-term follow-up revealed complete resolution in four cases, partial improvement in two cases and no improvement in two cases. This case series indicates that horses with IVS can make a full recovery (4/8) with return to ridden exercise at their previous level (5/8). Treatment with anti-inflammatory medication did not appear to influence outcome. No recurrence was reported at long-term follow-up.

**KEYWORDS**
ataxia, head tilt, nystagmus, peripheral

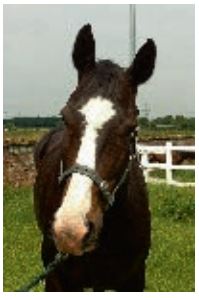

**FIGURE 1** Horse with head tilt—direction of head tilt is determined by the position of the poll. Horses with idiopathic vestibular syndrome demonstrate poll deviation towards the lesion (ipsilateral).

### Key points

- Idiopathic vestibular syndrome should be suspected in horses demonstrating acute onset vestibular dysfunction (head tilt, strabismus, nystagmus and ataxia) with no structural abnormalities identified on investigation.
- Facial and vestibulocochlear nerve dysfunction can occur concurrently.
- Full recovery from this condition is possible and does not appear to be associated with administration of anti-inflammatory medication.

This work was undertaken at the Leahurst Equine Hospital of the University of Liverpool.

Received: 3 September 2023 | Accepted: 23 September 2023

DOI: 10.1111/eve.13900

Equine Veterinary Education BEVA

CLINICAL COMMENTARY

# Vestibular disease in horses: Recognition, localisation and common causes

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## INTRODUCTION

Wilson et al. summarised the presentation, diagnosis, treatment and outcome in 8 horses with idiopathic peripheral vestibular syndrome (Wilson et al., 2024). When vestibular disease is encountered in the horse, it can be difficult to discern whether the associated clinical signs are central or peripheral in origin. However, appropriate localisation will narrow differential diagnoses and aid in the pursuit of appropriate diagnostic tests and ultimately treatment protocols. This clinical commentary aims to review the anatomy of the vestibular system, illustrate key clinical findings to distinguish central versus peripheral lesions and highlight common differential diagnoses and appropriate diagnostic tests.

## ANATOMICAL OVERVIEW

The central aspects of the vestibular system refer to those within the brain and spinal cord, whereas the peripheral portion exists outside these components, within the petrous portion of the temporal bone (de Lahunta & Glass, 2009). The peripheral sensory apparatus, comprised of bony and membranous labyrinths within the petrous temporal bone, contains a set of 'motion detectors' known as the semicircular canals and otolith organs. Hair cells (receptor cells) with cilia become deformed by gravitational forces, generating action potentials that progress through the vestibular nerve.

The central component arises after the vestibular nerve exits the petrous temporal bone and enters the calvarium through the internal acoustic meatus at the rostral medulla oblongata. Most of its axons synapse on the vestibular nuclei in the brainstem, while some proceed to the cerebellum. Information from the vestibular nuclei is transmitted via the medial longitudinal fasciculus to the motor nuclei of the oculomotor (CN III), trochlear (CN IV) and abducens (CN VI) nerves, coordinating eye movement. Other information is transmitted to the spinal cord to influence extensor and flexor tone. Balance during rest and movement is achieved through the vestibulo-ocular (physiological nystagmus) and vestibulospinal (extensor tone) reflexes (Aleman, 2022).

Normal physiological nystagmus results in rapid, involuntary movement of the eyes in the direction of head movement (de Lahunta & Glass, 2009). The eye should slowly move opposite the direction of the head and then exhibit a fast phase in the direction of the head turn towards a central position (Kent et al., 2010). Horses with vestibular disease might also have pathologic, spontaneous nystagmus when the head is still, with slow phase towards the side of the lesion and fast phase away (King, 1987). Pathologic nystagmus might only occur when the head is in a specific position (positional nystagmus), and pendular nystagmus without a discrete slow or fast phase has also been observed (Eggers, 2019). Additionally, vestibular disease can result in ipsilateral strabismus (abnormal eye position, most often deviated ventrally), which might be persistent or inducible by head elevation.

The normal vestibular apparatus yields equivalent facilitation of ipsilateral extensors and contralateral flexors in the limbs and trunk via vestibulospinal tracts (de Lahunta & Glass, 2009). Typically, a unilateral lesion within the vestibular apparatus will result in the body of the horse being 'pushed' (tilting, leaning or moving) towards the direction of the lesion by opposing extensors (Kent et al., 2010). Meanwhile, the cerebellum provides ipsilateral inhibition to the extensors. When it is affected, this will be absent resulting in a contralateral direction of leaning (paradoxical vestibular disease), described more comprehensively below.

## DETECTING AND DIFFERENTIATING CENTRAL AND PERIPHERAL VESTIBULAR DISEASE

A horse experiencing either central or peripheral vestibular disease might exhibit any constellation of head tilt, circling, pathologic

See Related article: https://doi.org/10.1111/eve.13852.

nystagmus, pathologic strabismus and vestibular ataxia (balance loss) as well as recumbency (Figure 1). Unlike with general proprioceptive ataxia, animals with pure vestibular ataxia maintain appropriate strength and awareness of limbs despite loss of equilibrium and balance.

In the authors' opinion, identification of vestibular disease is most straightforward in the moderately affected, ambulatory horse. Such affected horses show a wide-based, tentative gait, obvious head tilt and tendency to drift or lose balance to one side. Abnormal nystagmus and strabismus might or might not be obvious. Detecting vestibular disease in severely affected, recumbent horses can be more challenging; many neurological syndromes, from severe spinal cord damage to diffuse neuromuscular weakness, can result in recumbency, and type of ataxia cannot be assessed in a recumbent horse. Signs of vestibular disease in a recumbent animal include continuous or intermittent nystagmus, which might be elicited when the horse's head is lifted and turned and a strong preference for one lateral recumbency over the other. Animals with vestibular disease prefer to lie on the affected side and often show great reluctance to be turned to the opposite recumbency; if forced to turn over, the animal might thrash, struggle and flip back to the preferred side.

Mild vestibular dysfunction also can be difficult to detect. If a horse's signs are not obviously reflective of vestibular disease but it is suspected, placement of a blindfold or putting the animal in a poorly lit environment might exacerbate clinical signs and make the practitioner more convinced (Aleman, 2015). This method of exacerbation is effective because blindfolding or turning off lights removes the horse's ability to compensate visually for deficits. This should only be done if deemed safe.

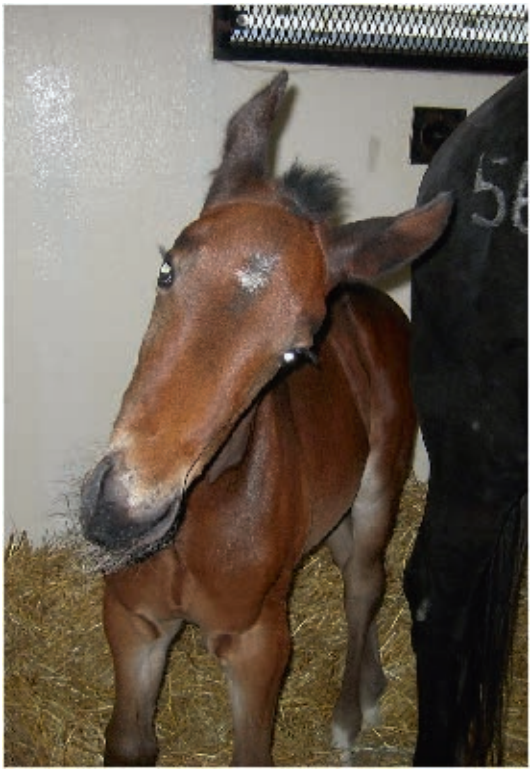

**FIGURE 1** Foal with left peripheral vestibular disease due to otitis media-interna. Note left head tilt and mild ventral strabismus of the left eye.

Signs of vestibular disease might be more severe in the early stages and less overt later as adaptation occurs. This is particularly true for abnormal nystagmus, which might be continuous initially but gradually decrease so that after 24–48 h it is only occasionally noticeable or inducible. Some horses with acute vestibular disease might stagger and collapse, struggling to rise. As Aleman (2022) describes, these signs can be confused with seizures, and combining anamnesis and clinical evaluation can help distinguish conditions.

Unilateral vestibular disease, as reported in horses with idiopathic vestibular syndrome, results in signs ipsilateral to the lesion. The horse will tilt its head, lean and circle towards the side of the lesion. These animals might display pathologic nystagmus at rest; both eyes are affected by the fast phase of the nystagmus *away* from the lesion. Ventral strabismus might be present or inducible in the ipsilateral eye. The horse is alert without proprioceptive deficits; even when losing its balance severely, the horse scrambles and moves its legs quickly, without evident weakness, to regain its balance. Ipsilateral deafness might be present but is difficult to discern clinically. Horses with peripheral vestibular disease might show deficits associated only with the vestibulocochlear nerve or might show both cranial nerve VIII and cranial nerve VII (facial nerve) deficits due to the proximity of the latter nerve to the former (Aleman, 2022). The presence of cranial nerve deficits outside of cranial nerves VII and VIII suggests central disease.

If a horse has bilateral peripheral vestibular disease, it will typically *not* exhibit a head tilt, leaning, drifting or nystagmus, but rather exhibit a wide-based stance and could stagger in attempts to maintain balance without a particular 'sidedness' (Aleman, 2022). If the disease is bilateral, there will be no nystagmus (pathological *nor* physiological), as there will be no ascending or descending positional information or resultant response. Bilaterally affected horses might display low and wide-swinging head and neck carriage as they attempt to maintain balance (de Lahunta & Glass, 2009). Complete deafness might be present.

Unlike uni- or bilateral peripheral vestibular disease, central vestibular disease is frequently accompanied by additional signs disparate from the vestibular system. Cranial nerve signs beyond CN VII and CN VIII increase clinical suspicion for central vestibular disease, suggestive of pathologic change elsewhere within the brainstem. Other signs of intracranial disease, including altered mental status or behaviour, cerebellar or general proprioceptive ataxia and seizures, similarly point the practitioner towards a central vestibular lesion. These findings (altered mental status, proprioceptive or cerebellar ataxia, and cranial nerve deficits in addition to cranial nerves VII and VIII) are considered 'firm' indicators of central disease. 'Softer' indicators that might suggest central disease include pathologic nystagmus with the fast phase directed towards the lesion/head tilt or changing direction with changes of head position (Troxel, Drobatz,

& Vite, 2005). Note that unilateral central disease can mimic unilateral peripheral disease if only the vestibular nuclei on one side are affected.

Paradoxical vestibular disease is a specific central phenomenon that occurs when the head tilt and loss of balance are contralateral to the side of the lesion. Typically, this occurs when the lesion is located within the cerebellum, specifically the flocculonodular lobe and caudal cerebellar peduncle (de Lahunta & Glass, 2009). Lesions in this region obliterate the inhibition that normally occurs via Purkinje neurons on the vestibular nuclei, leading to excitatory responses. Clinically this results in a contralateral head tilt because of the excessive stimulation on the affected side and relatively decreased stimulation on the normal side. Because the caudal cerebellar peduncle also contains general proprioceptive afferent fibres, abnormalities in postural reactions and proprioceptive ataxia are observed; these deficits occur on the ipsilateral side of the lesion.

## COMMON CAUSES OF EQUINE VESTIBULAR DISEASE AND RECOMMENDED TESTS

To reach a diagnosis of 'idiopathic' vestibular disease, as documented in the Wilson et al. case series, all likely alternative differential diagnoses should be investigated and excluded. One of the most common aetiologies of vestibular signs and specifically peripheral vestibular disease is temporohyoid osteoarthropathy (THO). The disease arises due to osseous proliferation at the temporohyoid joint with resultant pathologic fracture of the petrous temporal bone, leading to damage to the vestibulocochlear and/or facial nerves (Blythe et al., 1984; de Lahunta & Glass, 2009; Walker et al., 2002). Guttural pouch endoscopy and cross-sectional imaging are the most diagnostically useful tools (Figure 2a–e; Espinosa et al., 2017; Hilton et al., 2009).

Otitis media and interna can lead to vestibular signs. Otitis can be the result of bacterial, viral, fungal or even parasitic infections,

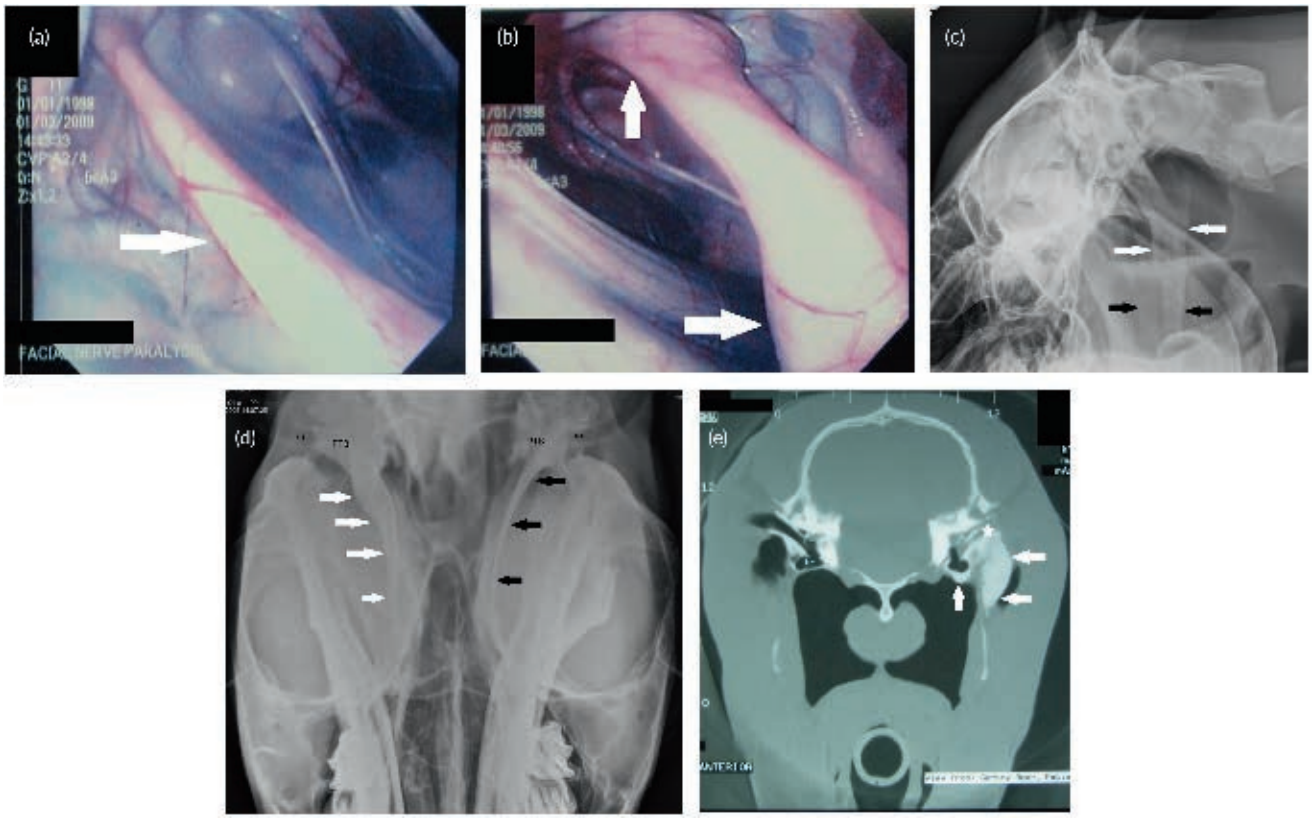


**FIGURE 2** (a) Endoscopic view of a normal right stylohyoid bone within the guttural pouch. Note the thin, smooth appearance of the bone (white arrow). (b) Endoscopic view of an abnormal left stylohyoid bone within the guttural pouch. Note the thickened, irregular appearance of the bone (white arrows). This horse had temporohyoid osteoarthropathy (THO). (c) Severe changes to the stylohyoid bone can be detected radiographically. This laterolateral projection of the head (rostral to the left) shows the two stylohyoid bones (black arrows) with osseous proliferation of one stylohyoid bone towards its dorsal aspect (white arrows). (d) Asymmetry in the stylohyoid bones in horses with THO can sometimes be detected in a dorsoventral radiographic projection. This severely affected horse has marked irregular osseous proliferation of the affected stylohyoid bone (white arrows) compared to the normal stylohyoid bone (black arrows). The petrous portions of the temporal bones are denoted (PTB); the affected side is sclerotic. The air-filled ear canals are marked (**). (e) Axial computed tomographic image of a horse with THO at the level of the ear canal and middle ear. The air-filled normal bulla is marked (**). The abnormal bulla on the affected side has a sclerotic ventral margin (vertical white arrow) with a large bony callous projecting laterally from the proximal stylohyoid bone (horizontal white arrows). The ear canal on the affected side is filled with a soft-tissue opacity (likely exudate or blood instead of air, marked by white star).

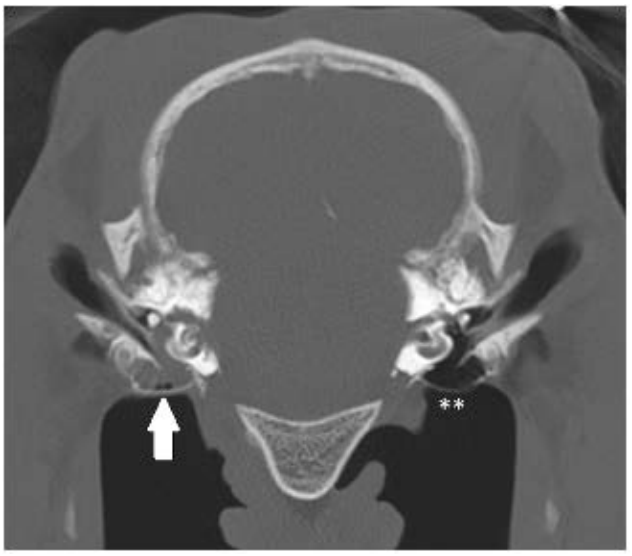


**FIGURE 3** Axial computed tomographic image at the level of the inner and middle ear from a foal with unilateral otitis media-interna. The normal air-filled bulla is marked (**). The affected bulla is mostly filled with a soft-tissue opacity (white arrow).

as well as neoplasia and less common causes (Blythe, 1997). Clinical signs can include headshaking, aural discharge if the horse has accompanying otitis externa and ear droop in addition to classic uni- or bilateral vestibular signs. Aural examination and endoscopy are challenging in horses, making cross-sectional imaging often the most useful diagnostic tool (Figure 3).

Trauma, most often related to collision from a kick, running into a fixed object or flipping over, can fracture the petrous temporal bone, damaging the vestibulocochlear nerve and causing peripheral vestibular syndrome. More severe trauma can cause central vestibular disease, often through intracranial haemorrhage or displaced fractures affecting the brain and brainstem. Vestibular disease is a common sequela of basilar skull bone fractures, as occur when a horse flips over backwards. As with THO and otitis media-interna, traumatic injuries are best evaluated through careful clinical examination and cross-sectional imaging.

An important differential diagnosis for central vestibular disease in horses in the Americas is equine protozoal myeloencephalitis (EPM). In the authors' practice, EPM is one of the most common causes of vestibular disease in horses. Despite affecting the brainstem, it can mimic peripheral disease if protozoa localise to the vestibular nuclei unilaterally. Diagnosis is best achieved through cerebrospinal fluid (CSF) and serum immunologic analyses to permit calculation of serum:CSF titre ratios or specific antibody indices. Therefore, CSF collection and EPM testing would be an important step in working towards a diagnosis of idiopathic vestibular disease in the Americas, whereas only 1/8 horses in the Wilson et al. report underwent CSF collection. Other infectious aetiologies affecting the brainstem and/or cerebellum, such as aberrant parasite migration (*Halicephalobus gingivalis*) or Lyme neuroborreliosis (*Borrelia burgdorferi*), typically cause aberrations in CSF that would point the examiner away from idiopathic disease. In a case report by Toth et al. (2012) evaluating meningitis and meningoencephalomyelitis in 28 equids, vestibular signs were present in almost half of cases. Equine brain abscesses have also been documented in cases of acute vestibular signs (Raphel, 1982). Thus, diagnostic tests should include CSF analysis when possible.

## CONCLUSION

Equine vestibular disease is likely to be encountered by the equine general practitioner. Knowledge of vestibular system anatomy and signs associated with peripheral versus central disease are essential for accurate localisation and construction of an appropriate differential diagnoses list. Wilson et al. (2024) have highlighted an important potential cause of peripheral vestibular disease, namely idiopathic vestibular disease. Diagnosis of such can only be reached after exclusion of all other pertinent possibilities. As Wilson et al. (2024) point out, guttural pouch endoscopy and cross-sectional imaging (computed tomography) assist in excluding structural abnormalities from consideration. Although CSF analysis was not commonly performed in their UK population of horses, this step is imperative in the Americas, an endemic region for *S. neurona*. Even in the UK and elsewhere, CSF analysis ideally would be performed prior to reaching a diagnosis of idiopathic disease, as other infectious and inflammatory diseases can affect the vestibular system and mimic peripheral disease.

### AUTHOR CONTRIBUTIONS

S. Colmer drafted the manuscript. A. Johnson provided substantial editing and revisions.

### CONFLICT OF INTEREST STATEMENT

No conflicts of interest have been declared.

### FUNDING INFORMATION

No funding was received.

### ETHICS STATEMENT

Not applicable to this clinical commentary.

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**How to cite this article:** Colmer, S.F. & Johnson, A.L. (2024) Vestibular disease in horses: Recognition, localisation and common causes. *Equine Veterinary Education*, 36, 122–126. Available from: https://doi.org/10.1111/eve.13900


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Received: 29 December 2021 | Accepted: 25 August 2023

DOI: 10.1111/eve.13895_1

Equine Veterinary Education BEVA

CASE REPORT

# Antemortem diagnosis of renal haemangiosarcoma in a Hanoverian gelding

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## SUMMARY

A 27-year-old Hanoverian gelding was presented to the Michigan State University Veterinary Medical Center for evaluation of lethargy, weight loss, stiffness, and more recent colic signs. At presentation, the gelding was alert with mild tachycardia (60 beats/min). Other physical findings were normal except for a mass detected in the area of the left kidney on rectal palpation. Laboratory analyses revealed anaemia (RBC count $4.9\times10^{12}$/L; rr 6.9–$10.4\times10^{12}$/L; haematocrit 25%; rr 30%–45%). hypoproteinaemia, hypoalbuminaemia, and hyperbilirubinaemia (39.3 μmol/L; rr 1.7–35.9 μmol/L), consistent with haemorrhage and haemolysis. Transabdominal ultrasonography revealed a 14×22.5 cm mass in the region of the left kidney with loss of normal renal architecture. The right kidney had multiple 0.5–2 cm diameter hyperechoic nodular structures without shadowing artefacts. An increased amount of hypoechoic peritoneal fluid was present around the liver and spleen, but no other abnormal findings were imaged in the remainder of the abdomen or the thorax. Abdominal paracentesis yielded a sample of serosanguinous fluid (presumed blood contamination). Percutaneous needle biopsy of the right kidney yielded a diagnosis of haemangiosarcoma. Palliative care was initiated with dexamethasone. 19 days after hospital discharge the gelding became recumbent, and humane euthanasia was performed. Necropsy examination confirmed disseminated haemangiosarcoma, including both kidneys, adrenal glands, spleen, heart, and lungs, with the largest neoplasm in the left kidney.

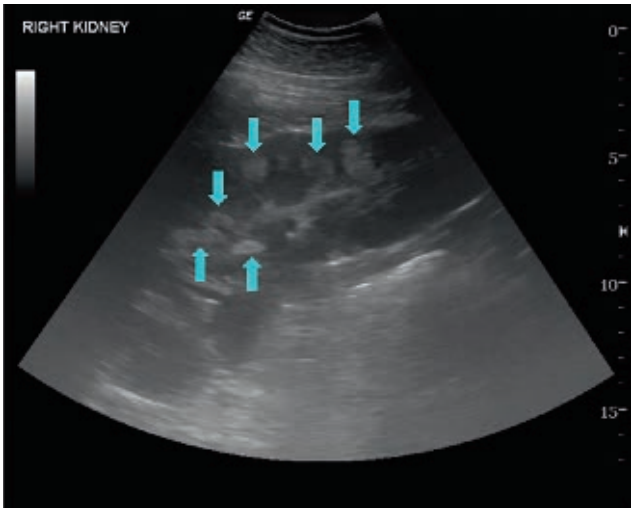


**FIGURE 1** Transabdominal ultrasonographic image of the right kidney revealing multiple hyperechoic nodules (arrows) in the renal parenchyma.

**KEYWORDS**
horse, disseminated, kidney, neoplasia

### Key points

- Disseminated haemangiosarcoma is an uncommon neoplasm in horses that is rarely diagnosed antemortem.
- Clinical signs of haemangiosarcoma are non-specific and can include lethargy, recurrent colic, icteric or pale mucous membranes, and weight loss.
- Disseminated haemangiosarcoma carries a poor prognosis.

Received: 1 October 2023 | Accepted: 5 October 2023

DOI: 10.1111/eve.13902

Equine Veterinary Education BEVA

CLINICAL COMMENTARY

# Ultrasonography as an aid to the antemortem diagnosis of internal neoplasia in the horse

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In the accompanying article by Williams et al. (2024) an aged gelding was diagnosed with renal haemangiosarcoma antemortem, with abdominal ultrasound and ultrasound-guided biopsy playing a key role in reaching the diagnosis. Obtaining an antemortem diagnosis of internal (abdominal and thoracic) neoplasia in the horse is challenging, with a definitive diagnosis obtained antemortem in only 34%–67% of horses in two case series of horses with intestinal neoplasia (Spanton et al., 2020; Taylor et al., 2006). Clinical signs are typically vague and non-specific, and include weight loss, lethargy, poor appetite and colic in cases of abdominal neoplasia. There are also no pathognomonic findings on routine haematological and serum biochemical analysis, although anaemia, hyper- and hypo-proteinaemia and increased levels of acute phase proteins can be seen (Spanton et al., 2020; Taylor et al., 2006). Whilst biomarkers such as thymidine kinase-1 (which can be increased in horses with lymphoma and multiple myeloma) can be measured, they are neither sufficiently specific nor sensitive to be able to confirm a diagnosis (Drozdzewska & Gehlen, 2023). In dogs, a so-called 'liquid biopsy' has been utilised as an aid in the diagnosis of neoplasia. DNA is released into the circulation when cell death occurs via apoptosis or necrosis and is broken down into fragments called cell-free DNA (cfDNA). Tumour cells also release cfDNA, termed circulating tumour DNA (ctDNA). Next-generation sequencing can be used to detect genomic alterations in the cfDNA of an animal that can indicate the presence of tumour cells in the body (Flory, 2023). Whilst the technology is not available in horses, it is something that will likely become available in the future, adding additional testing to the investigation of neoplasia in horses.

Unlike in small animals and people, the abdomen cannot be easily palpated in adult horses, and thus ultrasound is routinely performed to further investigate suspected disorders of both the abdominal and thoracic cavities (Freeman, 2003; Hillyer, 1994; Janvier et al., 2016). Further imaging via radiography can be pursued in horses with suspected thoracic disease, although its use is typically restricted to larger clinics and hospitals due to the equipment required. A systematic approach should be used with the aim of identifying abnormalities such as masses, increase in size of an organ, thickening of intestinal walls, increased volumes of peritoneal and pleural fluid and abnormal echogenicity of organs (Freeman, 2003; Hillyer, 1994). Although ultrasound can provide useful information, it is important to also remember the limitations of the technique, which primarily relate to the size of the horse and the topographical anatomy of the abdomen (Freeman, 2003; Hillyer, 1994). For example, it is estimated that as little as 20% of the liver can be viewed by transcutaneous ultrasound and as such, focal hepatic masses could be missed. Interestingly, in the case described by Williams et al. (2024), ultrasonographic examination did not identify multiple masses in the spleen which were subsequently identified at post-mortem examination, serving as a reminder that a 'negative' ultrasound does not rule out disease.

Whilst ultrasound cannot provide a definitive diagnosis of intestinal neoplasia, abnormal findings on ultrasound are commonly reported in horses with intestinal neoplasia (Janvier et al., 2016; Spanton et al., 2020; Taylor et al., 2006) (Figures 1–3). In one case series, 21/23 horses that had abdominal ultrasound performed had abnormalities identified including thickening of intestinal walls and identification of a mass (Spanton et al., 2020). To obtain a definitive diagnosis, samples for histopathology are usually required as tumours do not typically exfoliate into pleural or peritoneal fluid. Historically, many owners have not wished to pursue a definitive diagnosis when there has been a high index of suspicion of neoplasia. However, it is the author's experience that more owners are now wishing to pursue further diagnostics in an attempt to obtain a definitive diagnosis, as was the case in the accompanying report. If a mass or area of abnormal echogenicity within an organ is identified ultrasonographically, an ultrasound-guided biopsy or fine needle aspirate can be performed to obtain a sample for further analysis. Whilst the technique requires some practice to master, once the clinician becomes comfortable with the procedure, even relatively small lesions can be accurately sampled. For an excellent review of ultrasound-guided techniques, the reader is referred to Vaughan et al. (2009).

  

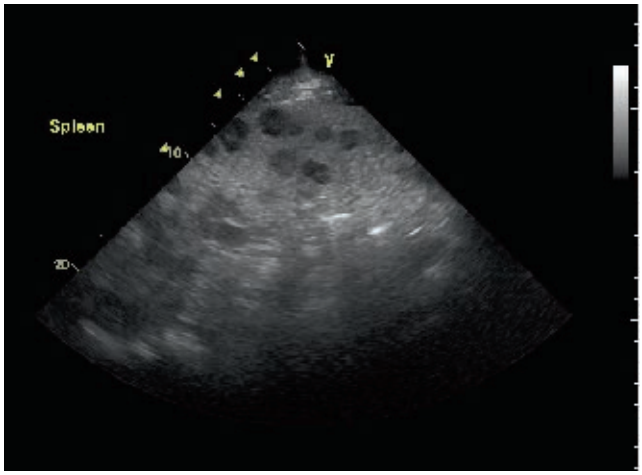


**FIGURE 1** Multiple circular hypoechoic masses within the spleen in a grey gelding with weight loss. Similar lesions were identified within the liver and on the pleural surface. Although external melanoma was not present, internal melanoma was strongly suspected and confirmed at postmortem examination.

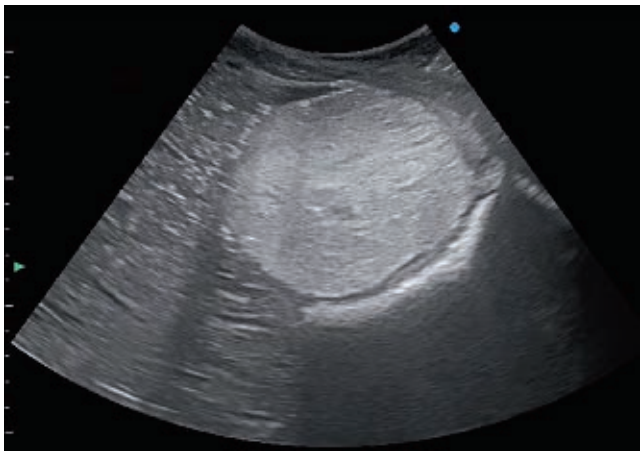

**FIGURE 2** Large spherical mass identified via transabdominal ultrasonography in a pony with recurrent colic episodes. The mass was removed via an exploratory laparotomy and subsequently diagnosed as a lipoma. The pony made an uneventful recovery from the surgery and the colic episodes resolved.

Biopsy of both the left and the right kidneys was required in the aged gelding before a diagnosis of haemangiosarcoma was made (Williams et al., 2024). The initial biopsy of the left renal mass was non-diagnostic, with a second ultrasound-guided biopsy of the visible masses within the right kidney performed, allowing for the final diagnosis. In comparison to hepatic biopsy, for example, renal biopsy is relatively rarely performed in horses, with only 151 biopsies reported from 14 institutions over a 26-year period (Tyner et al., 2011). In this case series, biopsy specimens yielded sufficient tissue for a histopathologic diagnosis in 94% of cases but had only fair (72%) agreement with post-mortem findings. Complications occurred in 11.3% of cases and were more likely when the left kidney

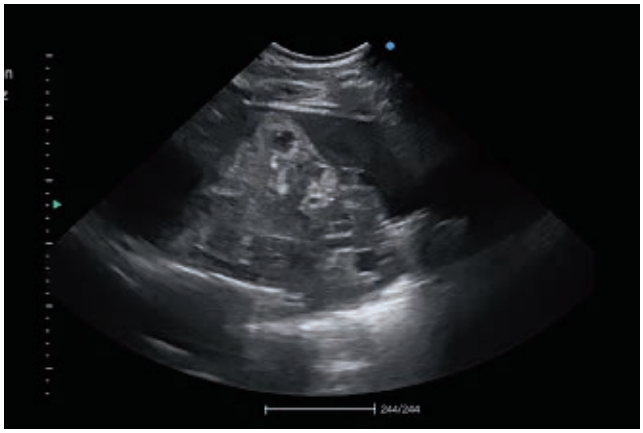

**FIGURE 3** Mass of mixed echogenicity located in the pleural cavity surrounded by echoic pleural fluid in an aged mare with signs of respiratory compromise. Neoplasia was strongly suspected based on the ultrasound findings and further diagnostics were not pursued.

was sampled and when a diagnosis of neoplasia was made, as occurred in the case reported by Williams et al. (2024).

Despite advances in the early diagnosis of neoplasia in people and small animals, antemortem confirmation of internal neoplasia remains challenging in horses and may not always be pursued when a high index of suspicion for neoplasia exists, as treatment options are typically very limited. In the case report by Williams et al. (2024), an antemortem diagnosis was pursued in a horse where neoplasia must have been strongly suspected based on the ultrasound findings. Whilst no additional treatment options became available after the definitive diagnosis was made, the authors reported that this allowed the owner to spend time with the horse at home before ultimately euthanasia was performed. As for this case, there are currently typically limited treatment options if a diagnosis of internal neoplasia is suspected. However, pursuing an antemortem diagnosis in these cases can be justified so long as the welfare of the horse is not compromised in pursuing that diagnosis or in delaying humane euthanasia, when appropriate. Ultimately, if we become more adept at making these diagnoses, especially if additional antemortem tests/biomarkers become available, then therapeutic options may also be expanded, as has happened in many cases of small animal neoplasia.

## FUNDING INFORMATION

No funding received.

## CONFLICT OF INTEREST STATEMENT

No conflicts of interest have been declared.

## ETHICS STATEMENT

Not applicable to this clinical commentary.

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**How to cite this article:** Johns, I. (2024) Ultrasonography as an aid to the antemortem diagnosis of internal neoplasia in the horse. *Equine Veterinary Education*, 36, 128–130. Available from: https://doi.org/10.1111/eve.13902

Received: 10 June 2022 | Accepted: 20 October 2023

DOI: 10.1111/eve.13915_1

Equine Veterinary Education BEVA

CASE REPORT

# *Listeria monocytogenes* encephalitis in a donkey foal

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## SUMMARY

This is the first known case report of encephalitic listeriosis in a donkey (*Equus asinus*). A 5-day-old female donkey foal presented for a 2-day history of fever, progressive weakness and loss of suckle reflex. Upon arrival, the foal's vital parameters were normal, but the foal was in lateral recumbency. A neurological examination revealed deficits of cranial nerves III (delayed pupillary light responses), V (atrophy of facial musculature on left side of the face), VII (no palpebral response on the left, slow palpebral response on the right) and VIII (slow, periodic bilateral horizontal nystagmus). Enophthalmos of the left eye and left third eyelid prolapse were present, with diminished muscle tone on the left side of the face. There was no damage to the globe or cornea on closer evaluation (including fluorescein staining) of the eye. There was little response to manual manipulation or noxious stimuli. The findings of the neurological examination localised the lesion to the cerebrum and/or brainstem. Samples collected for diagnostic evaluation included blood and cerebrospinal fluid (CSF). Haematological and biochemical values showed evidence of nonspecific inflammation (mild leucocytosis, an elevated plasma protein concentration characterised by a hypoalbuminaemia and hyperglobulinaemia and hyperfibrinogenaemia). The CSF showed a pleocytosis consisting mainly of nondegenerative neutrophils and had a normal protein concentration, and no bacteria were seen on Gram stain. A blood culture was negative after 8 days of incubation. No bacterial growth was isolated from the cerebral spinal fluid following 5-day enrichment in thioglycolate broth. After 24 hours of treatment with intravenous fluids, anti-inflammatories, antibiotics, nasogastric feedings of the dam's milk and supplemental nasal oxygen, little improvement was observed, and the owners elected to euthanise the foal. Post-mortem examination revealed multifocal, irregularly distributed hepatic necrosis and multifocal lymphohistiocytic encephalitis. A fresh section of brainstem was submitted for aerobic culture, and after a 2-day enrichment in Fraser broth, *Listeria monocytogenes* was isolated and identified by matrix-assisted laser desorption ionisation time of flight mass spectrometry (MALDI TOF). This case highlights the challenges of diagnosing listeriosis premortem. This donkey foal demonstrated neurological abnormalities more commonly seen in ruminant listeriosis cases. Contrarily, the abnormalities (sepsis, seizure or ataxia) documented in young horses with confirmed or suspected listeriosis have not localised to the brainstem. When diagnosing *L. monocytogenes*, it is important to document findings of the neurological examination and communicate clinical suspicion to the pathologist, as special culture and/or PCR methodology is often needed to identify *L. monocytogenes*.

**KEYWORDS**
horse, brainstem, donkey, encephalitis, foal, *Listeria monocytogenes*

### Key points

- While listeriosis in equids is relatively rare, most reports of confirmed *Listeria monocytogenes* infections have involved young animals.
- The clinical signs reported in equine cases of *L. monocytogenes* infection have been diverse and are not typically like the encephalitic form in ruminants.
- Special culture media and/or PCR methodology is often needed to identify *L. monocytogenes* in diagnostic samples.

Received: 6 July 2022 | Accepted: 27 October 2023

DOI: 10.1111/eve.13918_1

Equine Veterinary Education

CASE REPORT

# Ultrasound-guided modified subcostal transversus abdominis plane block in a foal undergoing omphalectomy

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## SUMMARY

A 7-day-old, 60 kg, purebred Spanish horse filly was referred to the veterinary teaching hospital for elective omphalectomy. Clinical examination was unremarkable except for a thickening of the umbilical area. Haematology and biochemistry values were within the normal range. On abdominal ultrasound (US) a well-defined, rounded (24 mm diameter), area that contained heterogeneous material with numerous hyperechoic foci with acoustic shadowing was observed within the inner portion of the umbilical pedicle, between both umbilical arteries. Surgical correction with omphalectomy was scheduled. To provide perioperative analgesia the TAP block was considered, a fascial plane block that desensitises the nerves innervating the abdominal wall. It involves the injection of local anaesthetic into the fascial plane between the transversus abdominis muscle and either the internal oblique abdominis muscle or the rectus abdominis muscle where the ventral branches of the thoracolumbar spinal nerves are located. Under general anaesthesia, the TAP block was performed in both left and right hemi-abdominal walls with a modified subcostal approach with two-site injections. A 22-G, 40 mm spinal needle was inserted under US guidance using an in-plane technique and advanced in a caudo-to-cranial direction. A total dose of 2 mg/kg (23.6 mL) of bupivacaine 0.5% diluted to a total volume of 0.64 mL/kg was injected. During surgery, low end-tidal concentrations of isoflurane were maintained (0.7%–0.9%). Surgery was completed uneventfully. No signs of nociception were observed suggesting adequate analgesia. Therefore, this TAP block technique may be considered to provide analgesia for surgical procedures involving the abdominal wall in foals.

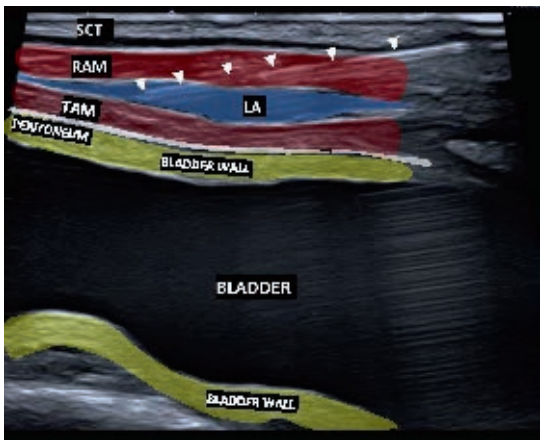


**FIGURE 1** Ultrasonographic image and photograph obtained during the TAP block technique. arrowheads, Needle position; LA, local anaesthetic; RAM, Rectus abdominis muscle; TAM, Transversus abdominis muscle.

**KEYWORDS**
horse, abdominal wall, bupivacaine, transversus abdominis (TAP) block, ultrasound-guided regional anaesthesia

### Key points

- Umbilical remnant infection is a common indication for abdominal surgery in foals.
- The transversus abdominis plane (TAP) block, between two easily identifiable fascial layers, block nerves that run within this plane desensitising the abdominal wall.
- Advantages include the use of less anaesthetic and analgesic drugs and doses, reducing adverse effects.


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Received: 23 February 2023 | Accepted: 21 June 2023

DOI: 10.1111/eve.13861

Equine Veterinary Education

ORIGINAL ARTICLE

# Perioperative pain management protocols of veterinarians in the United States for horses undergoing routine orchiectomy (castration)

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**Funding information**
Gift from Zoetis, Inc., Grant/Award Number: Not applicable.

## Summary

**Background:** Analgesic protocols related to orchiectomy or castration of young horses vary widely depending on geographic location, educational background of the veterinarian, and other demographic factors. Specific practices of equine veterinarians in the United States (US) have not been reported.

**Objectives:** To determine perioperative pain management practices of equine veterinarians in the United States as they relate to castration of healthy yearling colts.

**Study design:** Cross-sectional survey.

**Methods:** An internet-based questionnaire included items related to analgesic drugs used in association with castration of healthy yearling colts. Demographic, educational, and experiential factors associated with routine recommendation of analgesic medications after castration were analysed with calculation of odds ratios (OR) with 95% confidence intervals (CI) and multivariable logistic regression.

**Results:** Responses from 146 equine veterinarians in the United States revealed that 112/146 (76.7%) administered a nonsteroidal anti-inflammatory drug (NSAID) at the time of castration, and 107/143 respondents (73.3%) recommended administration of NSAIDs for a median of 3 days (IQR = 3–5) after surgery. Veterinarians who recommended NSAID analgesia after castration provided a higher pain severity score for horses at 24 h after a routine castration. Routine recommendation of post-castration NSAID analgesia was associated with veterinarians who were employed in a multi-veterinarian practice (OR = 2.7, 95% CI = 1.2–6.1) and completed their veterinary education in the year 2000 or more recently (OR = 2.6, 95% CI = 1.1–5.9).

**Main limitations:** Possible distribution, self-selection, and response biases as a result of convenience sampling methodology and possible recall bias of respondents.

**Conclusions:** Perioperative pain management practices vary widely among US veterinarians but the majority of veterinarians, especially more recent graduates and those in multi-veterinarian practices, recommend administration of an NSAID at the time of castration and for a median of 3 days after surgery.

**KEYWORDS**
horse, castration, nonsteroidal anti-inflammatory drug, orchiectomy, pain management

## INTRODUCTION

In recent years there have been notable advancements in recognition of behavioural manifestations of pain in horses including development and validation of a variety of tools to assess and 'measure' pain behaviours in horses. These include composite pain scales (Gleerup & Lindegaard, 2016; Pritchett et al., 2003; Sutton et al., 2013; VanDierendonck & van Loon, 2016), facial expression-based pain scales (Dalla Costa et al., 2014; VanDierendonck & van Loon, 2016), and ethograms of pain-associated behaviours in stabled and ridden horses (Dyson et al., 2018; Torcivia & McDonnell, 2021).

Several equine pain behaviour instruments have been used to evaluate pain experienced by horses after routine castration procedures with the general conclusion that there are differences in behaviour of horses after castration that may be modified or ameliorated with administration of analgesic medications (Dalla Costa et al., 2014, 2021; Lawson et al., 2020; Lemonnier et al., 2022; Lencioni et al., 2021; Sanz et al., 2009; Taffarel et al., 2015; Trindade et al., 2021; van Loon et al., 2010). In a 2020 survey of equine veterinarians in the United States (US), respondents were asked to rate the degree of pain experienced by a yearling colt 24 hours after a routine castration was performed, assuming no analgesia was provided after the procedure, using a numerical rating scale of 0 (no pain at all) to 10 (worst possible pain) (Sellon et al., 2022). Responses ranged from 1 to 10 with a median score of 5 and interquartile range of 4–7. This is very similar to responses from previous surveys of equine veterinarians in the Netherlands and Belgium (Dujardin & van Loon, 2011), the United Kingdom (Price et al., 2002), and Brazil (Lorena et al., 2013).

The goal of this study was to determine pain management practices of equine veterinarians in the United States, specifically as they relate to perioperative care associated with castration of young male horses. We hypothesised that most veterinarians prescribe nonsteroidal anti-inflammatory drugs (NSAIDs) for a minimum of 3 days after surgery and that a recommendation of post-operative analgesia is associated with more recent completion of a veterinary education.

## MATERIALS AND METHODS

### Questionnaire

The Institutional Review Board of Washington State University determined this project satisfied the criteria for exempt research as specified in the US Federal Policy for the Protection of Human Subjects with appropriate standards for ensuring informed consent, voluntary participation, and data security. An internet-based questionnaire for veterinarians, designed on a commercial internet survey site (Qualtrics), consisted of eight sections: introduction, attitudes and beliefs related to pain and pain management, assessment of pain severity, analgesic drugs, castration protocols, characteristics of current employment, and demographics including veterinary education and training (Supplementary Item S1). Responses related to the assessment of pain severity (on a scale of 0 indicating no pain to 10 indicating the worst possible pain), castration protocols, characteristics of current employment, and respondent demographics were included in this analysis. Invitations to participate in the survey were distributed through electronic mail lists and social media sites for equine veterinarians. Participation was initiated by clicking on a hyperlink that directly accessed the questionnaire.

### Data analysis

All statistical analyses were performed using commercial statistical software (SigmaStat 4.0, Systat Software, Inc.) with significance determined at $p < 0.05$ unless specified otherwise. Responses to each demographic, educational, and employment item were summarised separately. Descriptive statistics were determined as appropriate based on data distribution patterns and included mean with standard deviation, median with 25th and 75th quartiles (IQR), and 95% confidence intervals (CI).

After initial review of the data, some categorical variables from the demographic, educational, and employment sections of the questionnaire were curated into fewer categories to facilitate statistical analysis. For final analysis, current employment was classified as private clinical practice (owner, partner, associate, intern, or resident) or employment in academia, industry, or government settings; number of veterinarians in the practice was classified as 1 (solo practitioner) or multiple (>1); and number of certified veterinary technicians in the practice was classified as none or ≥1. The percentage of the practice that involved horses was classified as ≤75% or >75%. The average value of equine patients in the practice population was classified as < $10,000 or > $10,000. Respondents were classified as owning personal horses or not owning any personal horses. The year of veterinary degree completion, which was provided by respondents in categories based on decades, was classified for analysis as prior to 2000 (less recent graduates) or between 2000 and the date of the survey (more recent graduates). Board certification was classified as yes or no regardless of area of specialty certification.

Descriptive analysis of pain management practices that occurred at the time of castration was performed. This analysis prioritised evaluation of variables related to type of castration performed (standing or recumbent) and drugs administered at the time of castration.

More detailed analysis was performed regarding use of NSAID analgesia in the post-castration period. Respondents were categorised into groups designated Post-Castration NSAID Analgesia (one or more NSAID medications) and No Post-Castration NSAID Analgesia (no NSAID analgesia of any type) based on whether or not NSAIDs were recommended after routine castration of a healthy yearling colt. Post-castration pain scores were compared between groups using a Mann–Whitney rank sum test. Categorical variables from the demographic, educational, and employment sections of the questionnaires were compared between groups using Chi-square analysis with calculation of odds ratios (OR) and 95% CI. Interactions between

categorical variables were evaluated by Spearman rank correlation analysis. Categorical variables with $p < 0.25$ in univariate analysis were included in an automated backward stepwise analysis to identify a subset of independent predictors that were significantly associated with the dependent variable of Post-Castration NSAID Analgesia.

## RESULTS

### Questionnaire responses

A total of 343 equine veterinarians accessed the questionnaire. Data from 97 respondents were removed from the analysis because they failed to complete the survey. Data from an additional 63 respondents were removed from analysis because they did not confirm that they practiced in the United States and 37 respondents were removed because they indicated that they did not perform castrations. The final data set, therefore, included responses from 146 veterinarians practicing in the United States who performed castration surgeries on horses (Figure 1).

### Castration protocols

Of all respondents, 127 (87.0%) preferred recumbent castrations and 19 (13.0%) preferred standing castrations. At the time of castration, NSAIDs were administered by the majority of respondents (112/146, 76.7%). Lidocaine was injected into the testicles or spermatic cord at the time of surgery by 110 respondents (75.3%).

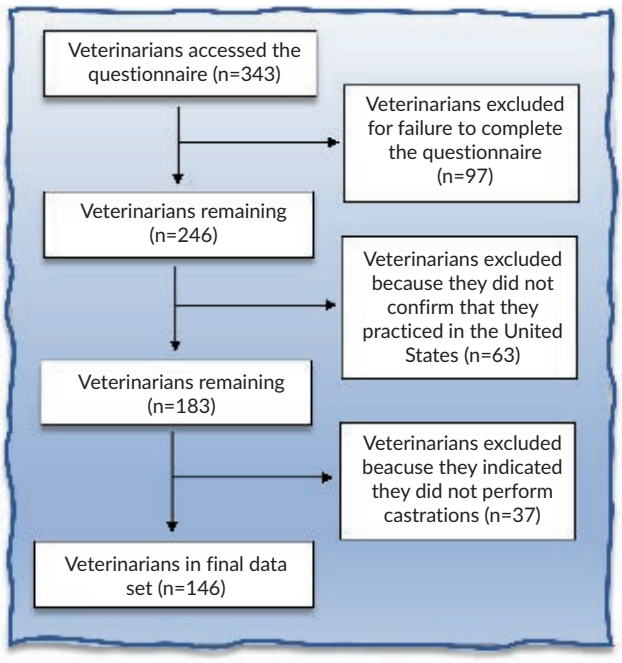


**FIGURE 1** Flow chart illustrating inclusion criteria for final data set.

There was no difference in frequency of NSAID administration at time of surgery, testicular or spermatic cord injection of lidocaine at time of surgery, or recommendation of NSAIDs after surgery between individuals who preferred recumbent approaches to castration and those who preferred standing approaches. There were 10 respondents (6.8%) who indicated that they provided none of these three types of analgesia at the time of, or after, routine castration. Of these 10 respondents, eight completed their veterinary education prior to 2000 and six worked as solo practitioners. The median pain score for post-castration horses provided by these respondents was 4 (range = 2–8). Three of these respondents rated the level of pain experienced by horses after castration as 7 or 8, yet they provided no analgesia beyond what was required for short-term anaesthesia for a recumbent castration. These three respondents all indicated that they used butorphanol, xylazine, and ketamine at the time of castration. They were all in private clinical practice comprised of 76%–100% horses and two of three graduated between 1980 and 1989.

### Post-castration analgesia

Of all respondents, 107 (73.3%) recommended use of one or more NSAIDs for treatment of healthy yearling colts after routine castration surgery. Of these 107 respondents, 94 (87.9%) also indicated that they routinely administered NSAIDs at the time of castration. There were 21 respondents (14.4%) who did not administer NSAIDs at the time of surgery and did not recommend their use after surgery. The duration of administration of NSAIDs after castration varied from one to 7 days (Figure 2) with a median duration of 3 days (IQR = 3–5 days). Respondents were permitted to select multiple types of NSAIDs. The most commonly recommended NSAIDs were phenylbutazone ($n = 77/112$, 68.8%) and flunixin meglumine ($n = 55$, 49.1%). Five respondents indicated that they recommended firocoxib for some patients. Xylazine and butorphanol were indicated as post-operative analgesic medications by one respondent each, both of whom also indicated that they recommended administration of an NSAID.

Veterinarians who recommended post-operative analgesic medications after routine castration surgery provided a higher median pain score for horses after castration as compared to those who did not make this recommendation (median values of 6 and 4, respectively, $p < 0.001$). The odds ratio and 95% CI for demographic, educational, and employment variables related to post-operative NSAID analgesia recommendations are shown in Table 1. Results of evaluation of possible interactions between these variables are shown in Table 2. Significant variables retained in the final model and positively associated with Post-Castration NSAID Analgesia included recent graduation (in the year 2000 or more recently) and employment in a multi-veterinarian practice (Table 3). The Likelihood ratio test statistic for this model was 15.287 ($p < 0.001$) with a Hosmer-Lemeshow statistics of 0.0524 ($p = 0.974$). The variance inflation factor (VIF) was 3.79 ($p = 0.8$).

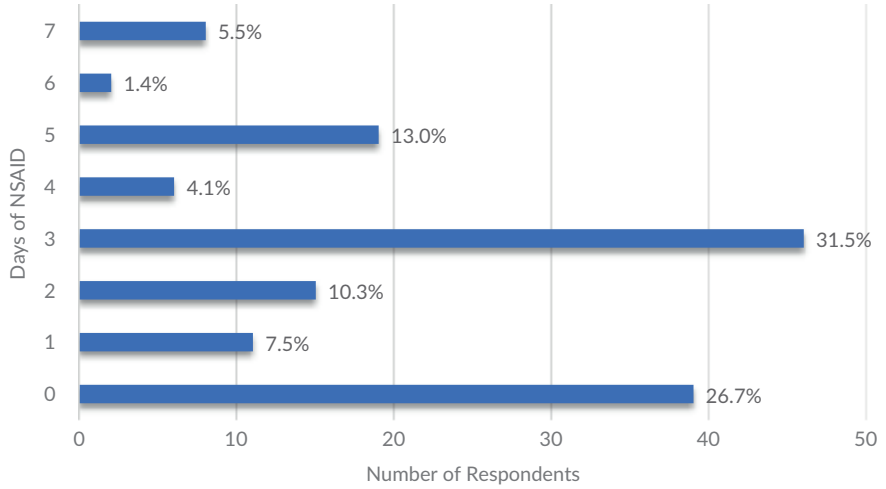


**FIGURE 2** Percentage of respondent US veterinarians who reported recommending administration of nonsteroidal anti-inflammatory drugs (NSAIDs) for the specified number of days after routine castration of a healthy yearling colt.

**TABLE 1** Odds ratios and 95% confidence intervals (CI) of categorical variables and their association with a veterinarian's recommendation of post-operative NSAID analgesia after routine castration of a healthy yearling colt.

| Categorical variables | Number | Recommend NSAID analgesia (%) | Do not recommend NSAID analgesia (%) | Odds ratio (95% CI) | *p*-Value |
|---|---|---|---|---|---|
| Academic, government or other employment | 145 | 19 (82.6%) | 4 (17.4%) | 1.9 (0.6–6.0) | 0.4 |
| Private clinical practice | | 87 (71.3%) | 35 (28.7%) | REFERENCE | REFERENCE |
| Multi-veterinarian practice | 145 | 81 (81.0%) | 19 (19.0%) | 3.4 (1.6–7.4) | 0.003* |
| Solo practitioner | | 25 (55.6%) | 20 (44.4%) | REFERENCE | REFERENCE |
| Credentialed veterinary technician in practice | 146 | 53 (77.9%) | 15 (22.1%) | 1.6 (0.7–3.3) | 0.3 |
| No credentialed veterinary technician in practice | | 54 (69.2%) | 24 (30.8%) | REFERENCE | REFERENCE |
| Practice population is ≤75% horses | 146 | 22 (75.9%) | 7 (24.1%) | 1.2 (0.5–3.0) | 0.9 |
| Practice population is >75% horses | | 85 (72.6%) | 32 (27.4%) | REFERENCE | REFERENCE |
| Average value of patients < $10,000 | 144 | 60 (68.2%) | 28 (31.8%) | 0.5 (0.2–1.2) | 0.2* |
| Average value of patients > $10,000 | | 45 (80.4%) | 11 (19.6%) | REFERENCE | REFERENCE |
| Respondent does not own horses | 146 | 29 (67.4%) | 14 (32.6%) | 0.7 (0.3–1.5) | 0.4 |
| Respondent does own horses | | 78 (75.7%) | 25 (24.3%) | REFERENCE | REFERENCE |
| Female | 145 | 78 (78.8%) | 21 (21.2%) | 2.4 (1.1–5.1) | 0.04* |
| Male | | 28 (60.9%) | 18 (39.1%) | REFERENCE | REFERENCE |
| Veterinary degree completed in 2000 or more recently | 144 | 65 (84.4%) | 12 (15.6%) | 3.4 (1.6–7.6) | 0.003* |
| Veterinary degree completed before 2000 | | 41 (61.2%) | 26 (38.8%) | REFERENCE | REFERENCE |
| Board-certified in a recognised veterinary specialty | 133 | 30 (81.1%) | 7 (18.9%) | 1.9 (0.7–4.7) | 0.3 |
| Not board-certified in a veterinary specialty | | 67 (69.8%) | 29 (30.2%) | REFERENCE | REFERENCE |
| Perform ≤10 castrations/year | 146 | 71 (73.2%) | 26 (26.8%) | 0.8 (0.3–2.2) | 0.9 |
| Perform >10 castrations/year | | 36 (73.5%) | 13 (26.5%) | REFERENCE | REFERENCE |
| Prefer standing castrations | 146 | 13 (68.4%) | 6 (31.6%) | 0.8 (0.3–2.2) | 0.8 |
| Prefer recumbent castrations | | 94 (74.0%) | 33 (26.0%) | REFERENCE | REFERENCE |

*Indicates variables included in backward stepwise regression analysis.

## DISCUSSION

In this study, >70% of US veterinarians administered an NSAID medication to healthy horses at the time of routine castration and >70% recommended continued NSAID medication for 1–7 days after surgery with a median recommendation of 3 days. Veterinarians who believed that otherwise healthy horses experienced a higher degree of pain at 24 h after castration surgery were more likely to routinely recommend NSAID analgesia in the days after castration. Demographic, educational, and employment variables positively associated with routine recommendation of post-castration NSAID analgesia were employment in a multi-veterinarian practice and completion of a veterinary degree in the year 2000 or more recently.

The reasons for these associations were not determined in this study. There were numerous significant interactions observed between variables such as the correlations between board certification and employment type, number of veterinarians in a practice, and presence of credentialed veterinary technicians in a practice. The absence of a gender effect in the final multivariable model is interesting because previous studies have shown significant differences in rating of pain severity after castration based on respondents' gender (Sellon et al., 2022) and because of the moderate correlation between gender and year of graduation that was demonstrated in initial analysis. It is quite likely that gender was not a variable retained in the final model because of its association with year of graduation. Improved ability to recognise pain behaviours in horses and increased focus on pain management in veterinary education may explain the association of post-castration NSAID use with more recent graduation.

The identification of 10 respondents who did not provide any NSAIDs before or after castration and did not use lidocaine at the time of castration was of interest. Butorphanol and xylazine are estimated to provide analgesia for a maximum of 3–4 h or 60 min, respectively, after intravenous administration (Sanchez & Robertson, 2014). It is impossible to know how long a horse may be painful after routine castration, but studies have reported behavioural evidence of pain for more than 8 h (Dalla Costa et al., 2014; Lemonnier et al., 2022; Love et al., 2013), suggesting that relying only on drugs administered at the time of surgery may be inadequate for pain control.

Rating of the severity of pain experienced by young horses after routine castration has been reported in several previous survey studies. In a 2002 study of veterinarians in the United Kingdom (UK), 70% of respondents considered pain associated with castration to be 'low' (Price et al., 2002). Using numerical rating scales in which 0 or 1 is no pain at all and 10 is the worst possible pain, several studies report median pain ratings after castration between 4 and 7 with a range from 0 or 1 to 10 (Dujardin & van Loon, 2011; Lorena et al., 2013; Sellon et al., 2022; Waran et al., 2010). This consistently broad range of scores across multiple studies in multiple countries suggest a continuing lack of consensus among veterinarians regarding the degree of pain experienced by horses after castration. These studies all used convenience sampling and questionnaires. It is not

**TABLE 2** Correlation coefficient ($p$-value) of the Spearman rank correlation analysis of all variables considered in univariate analysis.

| Variable | Number of veterinarians | Credentialed technicians | % of horses in practice | Value of patients | Personal horses | Gender | Graduation year | Board certification | Number of castrations | Castration positioning |
|---|---|---|---|---|---|---|---|---|---|---|
| Employment type | **0.20 (0.02)** | **0.24 (0.004)** | **0.17 (0.04)** | 0.14 (0.09) | 0.09 (0.3) | 0.095 (0.3) | −0.085 (0.3) | **0.48 (<0.001)** | 0.071 (0.4) | −0.057 (0.5) |
| Number of veterinarians | | **0.51 (<0.001)** | 0.11 (0.2) | −0.077 (0.4) | −0.032 (0.7) | 0.13 (0.1) | **0.33 (<0.001)** | **0.35 (<0.001)** | −0.0065 (0.9) | −0.093 (0.3) |
| Credentialed technicians | | | **0.22 (0.007)** | −0.14 (0.09) | 0.060 (0.5) | 0.11 (0.2) | 0.10 (0.2) | **0.32 (<0.001)** | 0.053 (0.5) | 0.0062 (0.9) |
| % of horses in practice | | | | **0.26 (0.002)** | 0.017 (0.8) | 0.082 (0.3) | 0.052 (0.5) | 0.15 (0.09) | 0.063 (0.5) | −0.040 (0.6) |
| Value of patients | | | | | −0.15 (0.08) | **0.20 (0.02)** | 0.10 (0.2) | −0.11 (0.2) | 0.028 (0.7) | −0.086 (0.3) |
| Personal horses | | | | | | −0.022 (0.8) | −0.045 (0.6) | 0.10 (0.2) | −0.018 (0.8) | 0.018 (0.8) |
| Gender | | | | | | | **0.53 (<0.001)** | −0.081 (0.4) | **0.27 (0.001)** | −0.087 (0.3) |
| Graduation year | | | | | | | | −0.051 (0.6) | 0.15 (0.07) | −0.13 (0.1) |
| Board certification | | | | | | | | | **0.20 (0.02)** | **−0.18 (0.04)** |
| Number of castrations | | | | | | | | | | −0.16 (0.06) |

*Note:* Values in bold text in grey-highlighted cells indicate significant correlations with $p < 0.05$.

**TABLE 3** Final multivariable logistic regression model for 143 veterinarians describing factors associated with the dependent variable of veterinarians who recommend NSAID analgesia after castration of a healthy yearling colt.

| Variable | Coefficient | Standard error | Wald statistic | *p*-Value | Odds ratio | 95% CI |
|---|---|---|---|---|---|---|
| Multi-veterinarian practice (REFERENCE CATEGORY: solo practitioner) | 0.985 | 0.419 | 5.5 | 0.02 | 2.7 | 1.2–6.1 |
| Veterinary degree completed in 2000 or more recently (REFERENCE CATEGORY: veterinary degree completed prior to 2000) | 0.9 | 0.4 | 4.9 | 0.03 | 2.6 | 1.1–5.9 |

Abbreviation: CI = confidence interval.

known how well these pain scores might correlate with scores given by veterinarians who are observing individual horses at the specified times after surgery.

There are little previous data regarding pain management practices of equine veterinarians in the United States associated with routine castrations of young horses but there is some information from other countries. A 2005 study indicated that only 36.9% of equine veterinarians in the United Kingdom routinely provided analgesia to horses after castration (Price et al., 2005). In a 2018 study of equine veterinarians in Australia, 43% of respondents gave one or more NSAID doses after surgery (Owens et al., 2018). In the current report, nearly 75% of respondents (107/146, 73.3%) routinely recommended analgesia after equine castration. It is difficult to compare results from studies that span two decades and three continents and that used very different survey instruments, but cumulatively these reports suggest that NSAID administration during the perioperative castration period may be more common now as compared to 20 years previously. This possibility is supported by current results demonstrating the association of post-castration analgesia recommendations by US veterinarians with a more recent year of graduation.

This was a cross-sectional survey study, which used a convenience sampling strategy with electronic distribution of survey invitations and associated potential for sampling and respondent bias. In addition, the sample size was relatively small as compared to the total population of equine veterinarians in the United States (estimated in 2018 as 4125 veterinarians in equine exclusive practice and an additional 4182 in mixed animal practice in 2018) (American Veterinary Medical Association, 2018). The results, therefore, should be interpreted with some caution.

These data confirm the lack of consensus regarding the degree of pain experienced by horses after routine castration and a lack of consensus on appropriate pain management strategies for these patients. Pain management guidelines for dogs and cats are provided by professional organisations in the United States with regular updates (Gruen et al., 2022). Evidence-based guidelines for equine analgesia, including perioperative analgesia for horses undergoing routine castration, were published by the British Equine Veterinary Association (BEVA) in 2020 (Bowen et al., 2020). These guidelines included recommendations for administration of pre-castration NSAIDs with a high level of overall certainty and administration of post-operative NSAIDs for at least 3 days after surgery with a moderate level of overall certainty. No analgesia guidelines for horses have been provided by professional veterinary organisations in the United States. The American Association of Equine Practitioners (AAEP) provides practice guidelines related to many other facets of equine practice including biosecurity, vaccination, drug compounding, euthanasia, and parasite control. Given the lack of consensus within the profession related to perioperative analgesia for horses undergoing routine castration, the frequency with which castrations are performed, the welfare concerns associated with inadequate perioperative analgesia, and the analgesia guidelines available for other species in the United States and for horses in other countries, it is recommended that the AAEP develop evidence-based guidelines for this very important area of equine practice.

## AUTHOR CONTRIBUTIONS

All authors contributed to the conception, design, and analysis/interpretation of data for this manuscript. All authors have approved this final version for publication.

## ACKNOWLEDGEMENTS

The authors thank Dr Jenifer Gold and Dr Debora Mattei for assistance in survey design and Dr Clark Kogan for advice on statistical methods.

## CONFLICT OF INTEREST STATEMENT

The authors have no potential sources of conflict of interest to declare.

## FUNDING INFORMATION

This work was funded by a gift from Zoetis, Inc.

## ETHICS STATEMENT

The Institutional Review Board of Washington State University determined that this research satisfied the criteria for exempt research as specified in the US Federal Policy for the Protection of Human Subjects.

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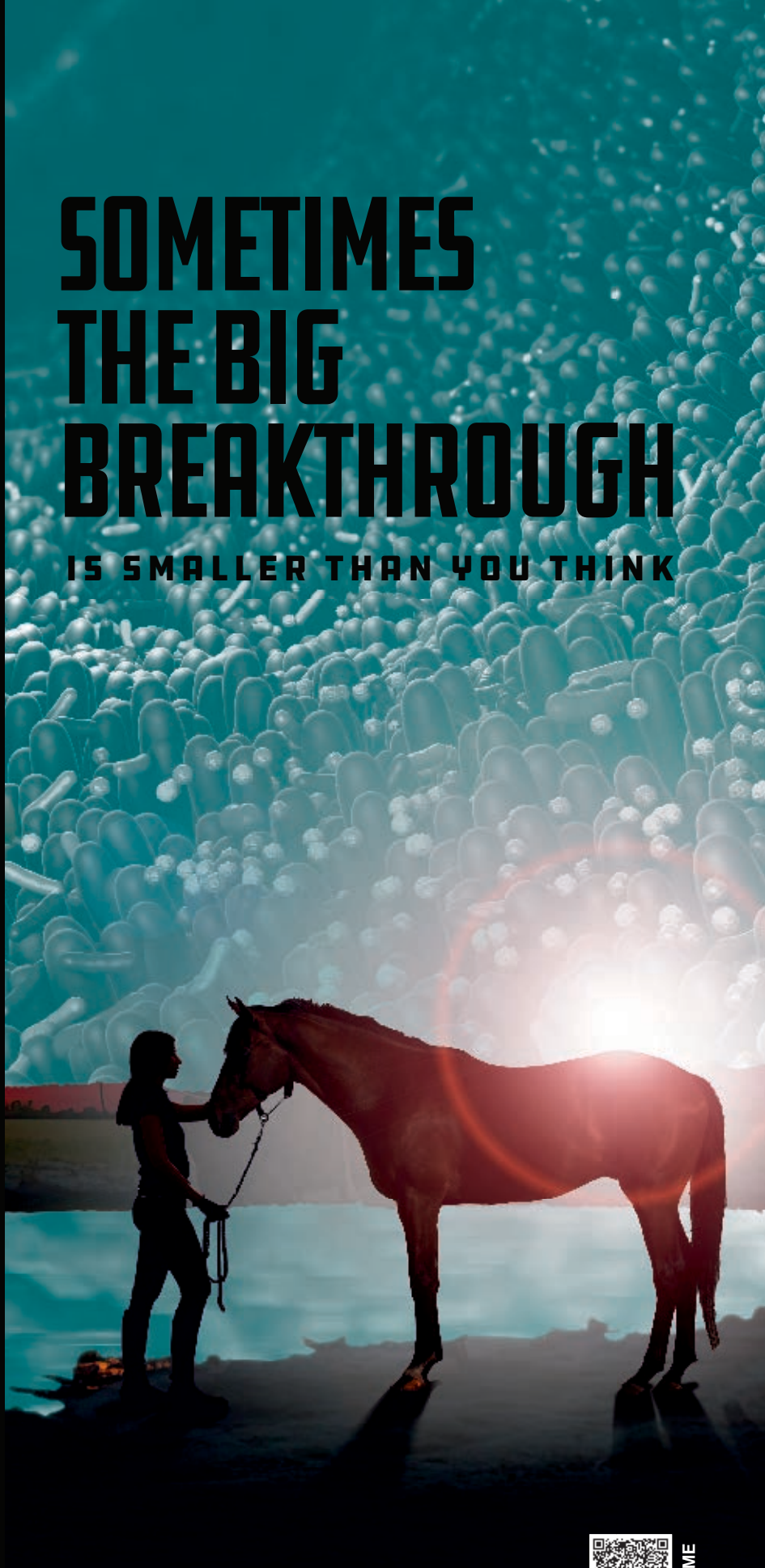

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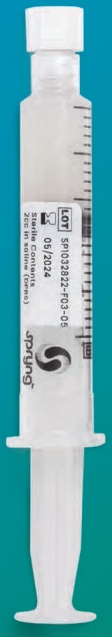

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## SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

**How to cite this article:** Sellon, D.C., Sanz, M.G. & Kopper, J.J. (2024) Perioperative pain management protocols of veterinarians in the United States for horses undergoing routine orchiectomy (castration). *Equine Veterinary Education*, 36, 133–139. Available from: https://doi.org/10.1111/eve.13861

Received: 30 January 2023 | Accepted: 29 July 2023

DOI: 10.1111/eve.13883

Equine Veterinary Education BEVA

CASE IMAGE

# A novel radiographic projection for the detection of a scapula body fracture in a Thoroughbred foal

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## Summary

This case report describes the clinical and diagnostic imaging findings, treatment and outcome of a 3-month-old Thoroughbred foal referred for the evaluation of severe left forelimb lameness and presents the acquisition of a novel radiographic projection of the scapula used to achieve the diagnosis. No abnormalities were found with the standard radiographic examination of the left shoulder. A right dorsal 45° lateral- left ventral lateral oblique radiographic view was performed to evaluate the body of the left scapula. A longitudinal, minimally displaced fracture of the infraspinous fossa was diagnosed with this projection. Ultrasound was used in conjunction to confirm the diagnosis, assess the displacement of the fracture in the sagittal plane and evaluate soft tissue damage. The foal was treated conservatively with anti-inflammatory drugs and stall rest. A radiographic follow-up and clinical reassessment were performed, and the horse appeared sound, no atrophy of the region was appreciated and no radiographic abnormalities were found.

**KEYWORDS**
horse, foal, fracture, radiograph, scapula

## INTRODUCTION

Scapular fractures are uncommon in horses (Adams & Nixon, 2019; Auer & Fürst, 2017; Dyson, 1985). Fractures may involve the supraglenoid tubercle, neck, spine, body and glenoid cavity (Adams & Nixon, 2019; Richardson & Ortved, 2022). These fractures are usually caused by traumatic processes or stress fractures in race breeds (Thoroughbreds and Quarter Horses). The latter may evolve into comminuted catastrophic fractures (Auer & Fürst, 2017).

The distribution of shoulder fractures generally includes horses less than 2 years of age due to the presence of the physis between the supraglenoid tubercle and the neck of the scapula. An overload of tension at the attachment of the biceps brachii and coracobrachialis tendons to the supraglenoid tubercle makes this the most common site for fracture of the shoulder (Auer & Fürst, 2017; Fortier, 2019; Richardson & Ortved, 2022).

Fractures involving the supraglenoid tubercle and the neck of the scapula are identified with routine radiographic projections of the shoulder (Adams & Nixon, 2019; Dyson, 1985). Due to the amount of superimposition of soft tissue and thoracic structures with the proximal thoracic limb, other methods of imaging, such as ultrasonography and nuclear scintigraphy, have been used to diagnose fractures involving the body and spine of the scapula (Richardson & Ortved, 2022; Vallance et al., 2009).

Conservative management of scapular body fractures not involving the glenoid cavity has been described (Dyson, 1985). Kidd et al. (2007) described a longitudinal fracture of the body of the scapula in a 2-year-old Paint horse with successful surgical treatment.

The aim of this case report is (1) to describe a novel radiographic projection, a dorsal 45° lateral- ventral lateral oblique, which is used to image the body of the scapula and (2) to describe the outcome of conservative management of a longitudinal scapular body fracture.

## CASE HISTORY AND CLINICAL FINDINGS

A 3-month-old Thoroughbred filly was referred to the Davidson Surgery Center at the Hagyard Equine Medical Institute in Lexington, Kentucky for a history of left forelimb lameness of 3 days duration. The lameness was first noted by the owner when the filly was brought in from turnout with other mares and foals. There was no evidence of visible trauma and the filly had been reportedly healthy. At the time of admission, the foal appeared bright, heart rate was 60 beats/min, respiratory rate was 24 breaths/min and temperature was 38.0°C. There was no evidence of joint sepsis or other comorbidities on physical examination. The foal was mildly resistant to palpation of the proximal forelimb. The passive range of motion of the proximal forelimb appeared normal. No crepitus, asymmetry or muscle atrophy of the region was appreciated. The foal's lameness was described as a 4/5 left forelimb lameness (AAEP grading scale, AAEP, 2018) with a reduction of the cranial phase of the stride at the walk.

### Imaging findings

The foal was sedated with xylazine (0.5 mg/kg IV) and radiographs of the left scapulohumeral joint were obtained while standing due to high suspicion of fracture in the proximal forelimb, with a low-output radiographic generator with exposure of 80 KV and 3 mAs and direct digital radiography (Enduras Wireless- MinXray, Cuattro Hub-Heska). Standard radiographic projections, including mediolateral (ML) (Figure 1) and cranial 45° medial-caudolateral oblique (Cr45°M-CaLO) views of the left scapulohumeral joint were unremarkable. A right dorsal 45° lateral- left ventral lateral oblique (RD45°-LVO) projection was obtained with the leg in flexion and the cassette placed closest to the affected limb, with the radiographic beam at 45° with respect to the sagittal plane of the horse (Figure 2). This image revealed a longitudinal fracture of the infraspinous fossa, coursing from the caudal border of the scapula, proximal to the neck, through the infraspinous fossa, parallel to the spine and exiting through scapular cartilage (Figure 3). Ultrasound examination of the left scapula, performed with a multi-frequency linear probe (GE Logiq E, 3–10 MHz), identified a defect in the surface of the infraspinous fossa, with no displacement of the fracture in the frontal plane (Figure 4). A similar radiographic evaluation performed 6 months later revealed complete healing of the fracture.

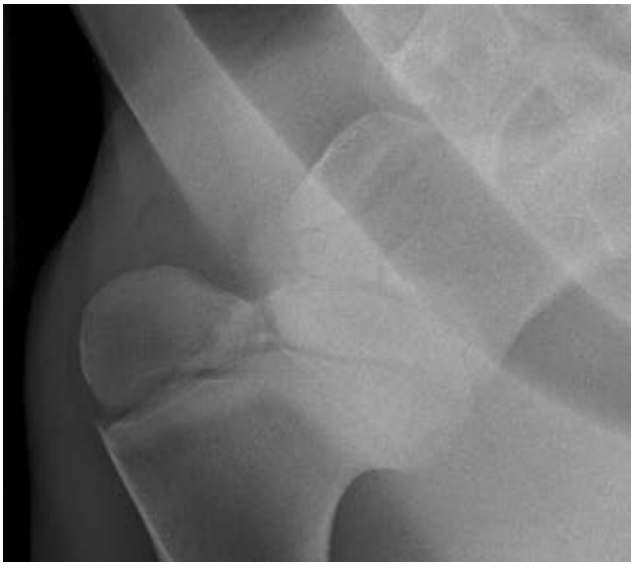

**FIGURE 1** Mediolateral radiograph of the left scapulohumeral joint. The supraglenoid tubercle and cranial glenoid centres of ossification remain cartilaginous.

### Treatment and outcome

The foal was confined to stall rest for 30 days with 5 days of non-steroidal anti-inflammatory therapy (firocoxib 0.1 mg/kg orally [PO] once a day). After 6 months, the filly was considered to be sound at the walk. No atrophy or asymmetry of the shoulder muscles was appreciated during the examination. The filly is expected to enter training the following year.

## DISCUSSION

In small animals, fractures of the scapular body are categorised by articular involvement as well as stability including intra-articular, unstable extra-articular and stable extra-articular (Cook et al., 1997). This categorisation scheme guides the need for stabilisation (surgical or coaptation) as well as prognosis. Surgical intervention is recommended for intra-articular as well as unstable extra-articular fractures of the scapula. The filly in this report fits into the category of a stable extra-articular fracture in which conservative management is an appropriate treatment option. The comfort level of the horse in this case was also a large determinant of management, as the horse was weight-bearing. Potential complications for conservative management of this fracture include mal- or non-union, propagation or displacement of the fracture, development of angular limb deformities or laminitis in the support limb or flexural limb deformity in the affected limb, secondary suprascapular neuropathy and persistence of lameness.

There are several reports of surgical repair of fractures of the supraglenoid tubercle in horses, only one case report describing the repair of a fracture of the scapular body was identified (Auer & Fürst, 2017; Kidd et al., 2007). Kidd et al. (2007) reported a longitudinal scapular fracture of the infraspinous fossa in a 2-year-old horse. The horse was reported to be sound for 10 months (Kidd et al., 2007). Repair of these fractures in a foal could result in an increase in morbidity due to the application of internal fixators to thin, compact bone. The ability to reduce the fracture line would be limited and the goal of repair would be to provide stability, which is inherently provided by the large muscle mass surrounding this region. With a lack of glenoid cavity involvement, the risk of degenerative

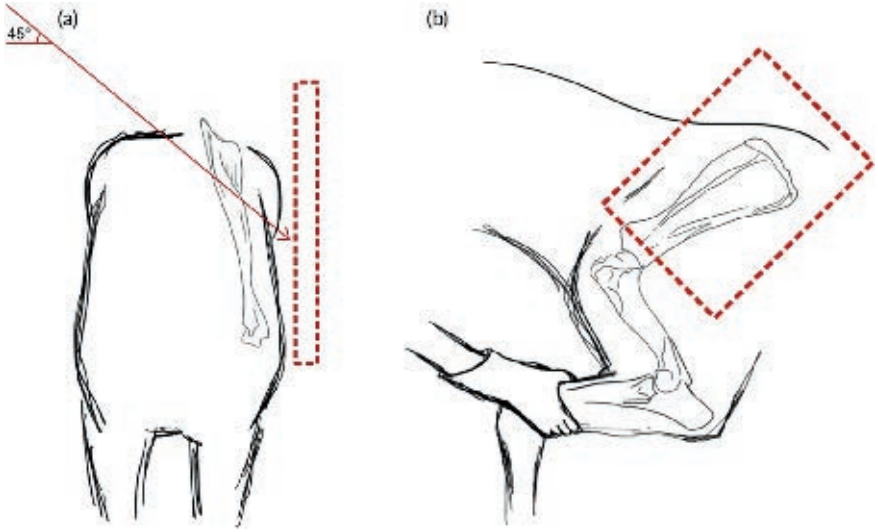


**FIGURE 2** Technique used to obtain the dorsal 45° lateral-ventral lateral oblique projection of the left scapula. The left forelimb is flexed and the cassette is positioned on the side of the affected limb. The beam is directed from the opposite side, at a 45° angle with respect to the cassette.

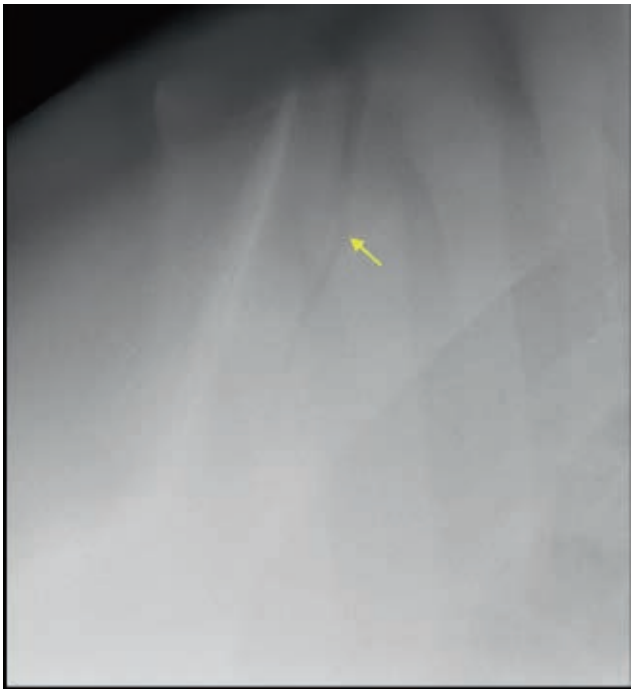

**FIGURE 3** Right dorsal 45° lateral- left ventral lateral oblique radiograph of the left scapula, revealed an incomplete longitudinal fracture of the infraspinous fossa, extending from the proximal aspect of the scapula and through the infraspinous fossa, parallel to the scapular spine (arrow).

joint disease was negligible. In the authors' opinions, the treatment of choice for stable extra-articular fractures of the scapula is conservative management.

The prognosis for return to full function for such fractures with conservative management in small animal medicine is said to be excellent (Cook et al., 1997; Peck, 2012). The prognosis for equine athletes cannot be determined based on the extrapolation of small animal cases, as the definition of full function for a pet animal and an adult horse differ. Recovery of three cases with longitudinal body fractures has been described in the equine veterinary literature. As previously mentioned, a single case report by Kidd et al. (2007) describes one horse returning to soundness after surgical repair. Dyson reports one horse returning to previous use while another was retired. Based on reported cases, of both small animals and horses, it is suggested that scapular fractures without articular involvement carry a better prognosis for future soundness than fractures with articular involvement (Cook et al., 1997; Dyson, 1985; Kidd et al., 2007; Peck, 2012).

With suspected fractures of the equine proximal forelimb, ultrasound has been described as a valuable diagnostic tool (Davidson & Martin, 2004). Ultrasound has the limitation of only being able to highlight the lateral aspect of the scapula and centres of ossification or normal roughening of the bony surface can be confused with a fracture line. While the exact configuration of a fracture cannot be determined by two-dimensional imaging alone, radiography was able to highlight the displacement in the cranial-caudal plane, while ultrasonography was performed as an adjunctive diagnostic tool to evaluate a potential displacement of the fracture in the sagittal plane or bone sequestrum, along with the concurrent soft tissue damage. Nuclear scintigraphy has been reported to have little value in the diagnosis of lameness in foals due to a diffuse increase in radiopharmaceutical uptake of the normal physes (Hunt, 2011).

The gold standard for the determination of the exact configuration of fractures is computed tomographic (CT) examination. Prohibiting factors of such examination include patient size as well as equipment access, owner funds and the associated risks of general anaesthesia. With standard radiographic views of the shoulder, the diagnosis would have not been reached in this case. In cases where the standard radiographic views are non-rewarding and there is an indication of proximal limb lameness such as crepitus, reduced

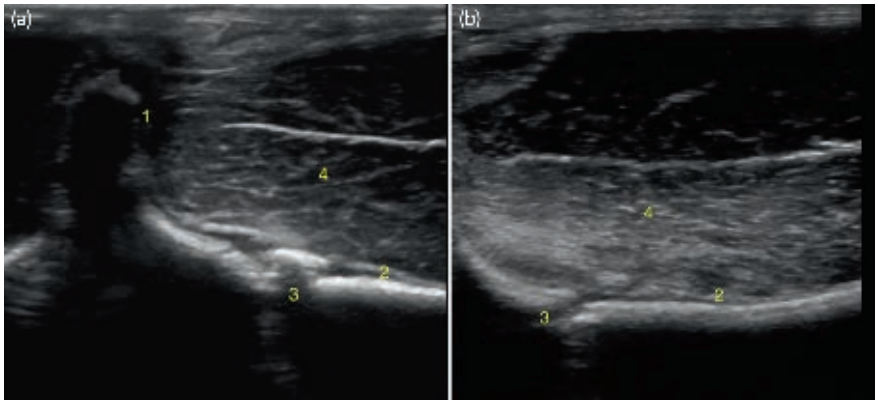


**FIGURE 4** Transverse ultrasound of the left scapula (cranial to the left). The fracture can be appreciated in the infraspinous fossa as discontinuity of the hyperechoic osseous interface. 1: scapular spine, 2: infraspinous fossa, 3: fracture, 4: infraspinatus muscle.

cranial phase of the stride, dropped elbow or pain on palpation, an additional skyline view of the body of the scapula may be indicated. The skyline radiographic view described in this report was sufficient for the diagnosis and development of a treatment plan. To the authors' knowledge, this radiographic projection has not previously been described in the equine literature. This radiograph was captured with a low-output portable generator making the acquisition of this image achievable to field practitioners, who lack access to or possess an insufficient ultrasound machine. Superimposition of the opposing limb and thoracic structures makes the diagnosis of a dorsal scapular fracture difficult with traditional radiographic views. In neonatal foals, a lateral view of the cranial thorax may allow for visualisation of the entire scapula, however, the described oblique view (RD45°-LVO) provides the advantage of anatomic isolation.

When the radiographic examination was repeated after 6 months with a low-output portable generator, the fracture was no longer present. As the horse aged and significantly increased in size, the radiographic image quality decreased, especially in the distal aspect of the scapula. Better radiographic images could likely be obtained with a high-output radiographic generator and a higher exposure. Based on the findings in this case report, the authors recommend using the described radiographic projection in conjunction with ultrasound if there is a suspicion of a fracture of the scapular body. In addition, extra-articular stable fractures, with none or minimal displacement carry on a good prognosis with conservative management.

## CONCLUSION

A dorsal 45° lateral-ventral-lateral oblique is an easily achievable radiographic projection that allows for the diagnosis of scapular body fractures in foals. Multiple diagnostic aids such as radiography paired with ultrasound can provide a better understanding of fracture configuration for areas in which more advanced imaging is not possible. The radiographic projection described in this report is recommended when the suspicion of a scapular body fracture is present. Stable, extra-articular fractures of the scapula can be managed conservatively with a favourable outcome.

### AUTHOR CONTRIBUTIONS

Case management and imaging interpretation were performed by all authors. The manuscript was prepared by V. Santalucia and M. Murphy and approved by R. Hunt. All authors gave their final approval of the manuscript.

### CONFLICT OF INTEREST STATEMENT

No conflicts of interest have been declared.

### FUNDING INFORMATION

No funding received.

### ETHICS STATEMENT

No experimental animals were used in this case report. No submission to an ethics committee was required. The owner gave permission to use the patient data for scientific purposes.

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**How to cite this article:** Santalucia, V., Hunt, R. & Murphy, M. (2024) A novel radiographic projection for the detection of a scapula body fracture in a Thoroughbred foal. *Equine Veterinary Education*, 36, 140–144. Available from: https://doi.org/10.1111/eve.13883

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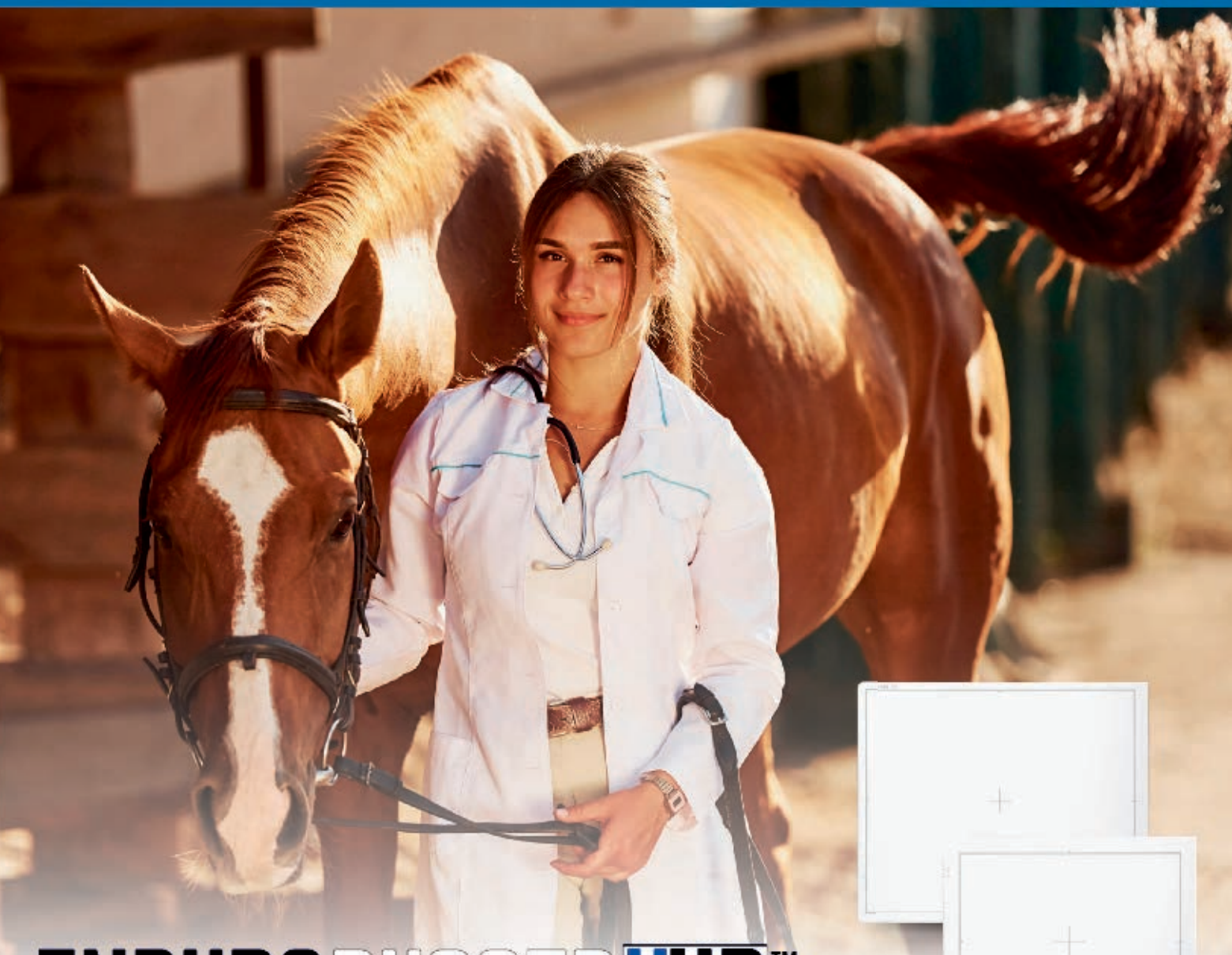

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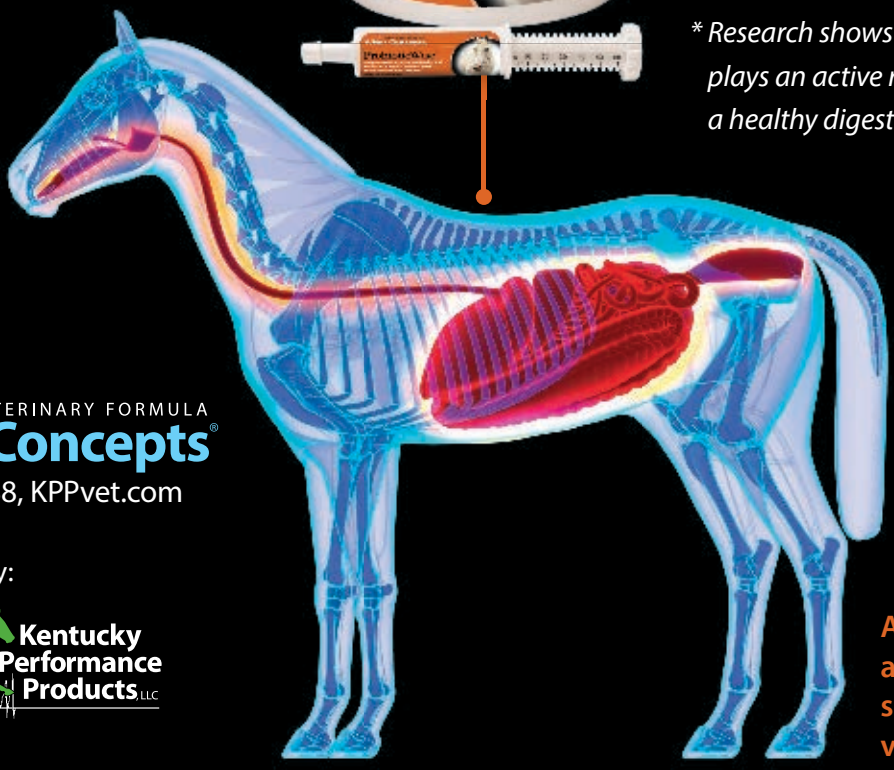

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Received: 6 June 2023 | Accepted: 25 August 2023

DOI: 10.1111/eve.13894

Equine Veterinary Education BEVA

REVIEW ARTICLE

# Bringing equine adipose tissue into focus

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## Summary

Adipose tissue is not only required for energy storage but is an essential endocrine organ with a central role in the pathology of obesity. The understanding of its role, both in human and equine medicine, is continually evolving. With obesity being an ever-growing problem in equine populations, gaining owner compliance is critical when implementing management plans. The aim of this review is to encourage the inclusion of the concept of adiposity in discussions with horse owners on obesity and metabolic syndrome. In doing this, we hope to improve clients' understanding and, therefore, maximise the impact of diagnostic tests, monitoring tools and management.

**KEYWORDS**
adipose tissue, horse, obesity

## INTRODUCTION

Adipose tissue (AT) is a multifaceted organ, essential for health. Whilst its role in lipid storage is well established, understanding the importance of AT as an endocrine organ, with a central role in the pathology of obesity, is evolving. Helping clients to understand the systemic impacts of excess AT can maximise the impact of diagnostic tests, monitoring tools, management and owner compliance.

## ADIPOSE TISSUE AS A STORAGE ORGAN

The amount of AT varies hugely between individual horses and is affected by factors such as age, sex and breed (Wallis & Raffan, 2020). Estimation of body fat content by deuterium oxide dilution found AT to account for 2.7%–35.6% of total body weight in horses (Dugdale et al., 2012). There are two distinct types of AT: white AT and brown AT. The white AT predominates and is specialised for energy storage. The brown AT, named due to its colouration from the high concentration of mitochondria, has a role in thermoregulation and, as a consequence, is abundant in neonates and animals in hibernation (Kiranmayi & Bhargav, 2019). Most adult mammals have very low quantities of brown AT and indeed, this form has not been described at all in the adult horse. There is great interest in brown AT pharmacological activation as a novel therapeutic target in human obesity (Liu et al., 2022); further research into brown AT in horse may, therefore, be of interest.

White AT is primarily an energy storage organ and is composed of adipocytes, in which lipid accumulates, as well as connective tissue, immune cells and blood vessels. AT can store almost 100 times more megajoules of energy than muscle, and this resource allows mammals to cope with changes in energy availability over time. Humans with congenital lipoatrophy (Berardinelli-Seip syndrome), who have a functional failure of their AT, rapidly develop severe insulin resistance and hepatic lipidosis, leading to liver failure and cardiovascular disease (Garg, 2004). This demonstrates that AT is an essential component of energy homeostasis and should not be viewed as always detrimental to health.

Adipocytes are developed from mesenchymal stem cells in a process called adipogenesis; a complex multistep process which includes the formation of pre-adipocytes from undifferentiated stem cells and the formation of mature adipocytes from these pre-adipocytes by accumulation of lipid. Equine adipose mesenchymal stem cells (MSCs) are well understood due to their regular use in the developments of biological therapeutics for the treatment of musculoskeletal disorders (stem cell therapy), which has evolved due to their relative ease of acquisition and robust response to in vitro manipulation (Marycz et al., 2016). The in vitro characteristics of equine AT MSCs are very similar to those of humans, including the

pre-adipocyte response to insulin and glucocorticoid stimulation, which induces lipid accumulation and drives differentiation to mature adipocytes (Bukowska et al., 2021).

Adipocytes within WAT store fat as triglyceride (triacylglycerol) in one large lipid droplet per cell. Triglycerides from dietary fat are absorbed from the gut and transported to the AT in the form of chylomicrons. AT, as well as liver, can also synthesise triglycerides from excess carbohydrates, a process known as de novo lipogenesis (DNL) (Ameer et al., 2014). The starting substrate for DNL varies greatly between species, with horses using acetate, unlike humans who primarily use glucose or pigs who can use either (Suagee et al., 2010). AT, as opposed to the liver, is the primary site of DNL in horses in contrast to humans (Adolph et al., 2019; Suagee et al., 2010), which may explain why fatty liver is less common in horses than other species. Aberrant DNL is associated with insulin resistance and cardiovascular risk in humans and is a potential link between excess carbohydrate intake and these conditions (Ameer et al., 2014); however, very little is known about changes in DNL in equine disease.

In the face of increased caloric intake, AT can expand through hyperplasia (increased adipocyte numbers derived from pre-adipocytes) or by hypertrophy (increase in individual cell size via lipid accumulation). The capacity for hyperplasia varies between AT depots, as discussed later. When required, the stored triglycerides are broken down into glycerol and free fatty acids by lipolysis. Lipolysis is predominantly controlled by insulin in the fed state through actions on adipose triglyceride lipase (ATGL) and hormone-sensitive lipase (HSL). These fatty acids are then available to mitochondria for respiration. The ability and speed with which AT mobilises stored triglycerides is one of the main distinguishing features between AT depots.

## ADIPOSE TISSUE AS AN ENDOCRINE ORGAN

In addition to its primary role in storage, AT is the largest endocrine organ in the body, producing and responding to hormonal signals and critical in the cross-talk between metabolic organs, which govern energy homeostasis (Figure 1). Adipocytes secrete bioactive peptides (adipokines and adipocytokines), which can act locally (autocrine/paracrine) or systemically (endocrine). AT also possesses a complex receptor profile, which allows it to respond to endocrine and nervous system input. Finally, adipocytes are able to exert fine control over endocrine signalling through their enzyme machinery, important in the metabolism of hormones, particularly steroid hormones. Our understanding of the equine adipocyte endocrine profile is more limited than that of humans or rodents, but some data have been published.

### Adipokines

Leptin is a critical regulator of energy storage through appetite/satiety control: when energy stores are adequate, leptin signals to the hypothalamus to reduce appetite drive, thus reducing food intake. As in other species, leptin is secreted from equine adipocytes in proportion to body fat mass. Indeed, animals with increased body fat (without disease) have higher plasma and AT expression of leptin (Buff et al., 2002; Staub et al., 2019).

Adiponectin is produced almost exclusively by adipocytes (Fang & Judd, 2011) and it acts primarily on muscle and liver to increase insulin sensitivity and reduce inflammation. In most species, including horses, there is an inverse relationship between fat mass and plasma adiponectin (Kearns et al., 2006).

Resistin is also an AT-specific protein, whose transcription is induced during differentiation of adipocytes. In rodents, it has been shown to decrease gluconeogenesis in the liver (Banerjee et al., 2004). Numerous human studies have failed to demonstrate a reliable association between resistin levels, obesity and/or insulin dysregulation. In contrast, resistin may be a marker of inflammation (Banerjee & Lazar, 2003), consistent with recent findings in horses (Fuentes-Romero et al., 2021).

### Adipocytokines

Adipocytes, stromovascular elements and inflammatory cells all contribute to adipocytokine secretion. Cytokines released include interleukin 6 (IL-6), tumour necrosis factor alpha (TNFα), monocyte chemoprotectant protein-1 (MCP1) and interleukin 1 beta (IL1β). The actions of the adipocytokines are diverse and are not limited to inflammatory effects such as chemoattraction and immune cell activation. For example, TNFα suppresses free fatty acid and glucose uptake into adipocytes in humans (Ruan et al., 2002). Little is known about the actions of these cytokines in equine AT but expression of several, including *TNFα*, *IL1β*, IL-6 and *MCP1*, has been shown (Basinska et al., 2015; Reynolds et al., 2019). The analysis of these cytokines has been proposed as a potential diagnostic target in equine metabolic syndrome but are not widely available and will require careful interpretation because of their lack of disease specificity.

### Steroid hormones

Adipose tissue is crucial for the action and metabolism of steroid hormones, particularly glucocorticoids and sex steroids. Furthermore, glucocorticoids are essential in the differentiation of mature adipocytes. AT in humans and horses contains high levels of glucocorticoid receptor (GR) and also the metabolising enzyme 11β-hydroxysteroid dehydrogenase type 1 (HSD1), which metabolises inactive cortisone into active cortisol and thus potentiates activation of GR (Morgan et al., 2018). Horses predominantly metabolise endogenous cortisol through the carbonyl reductase 1 pathway, which is very active in AT and produces a metabolite (20-beta dihydrocortisol), which impairs glucose tolerance (Bell et al., 2021; Morgan et al., 2017). Less is known about sex steroid metabolism

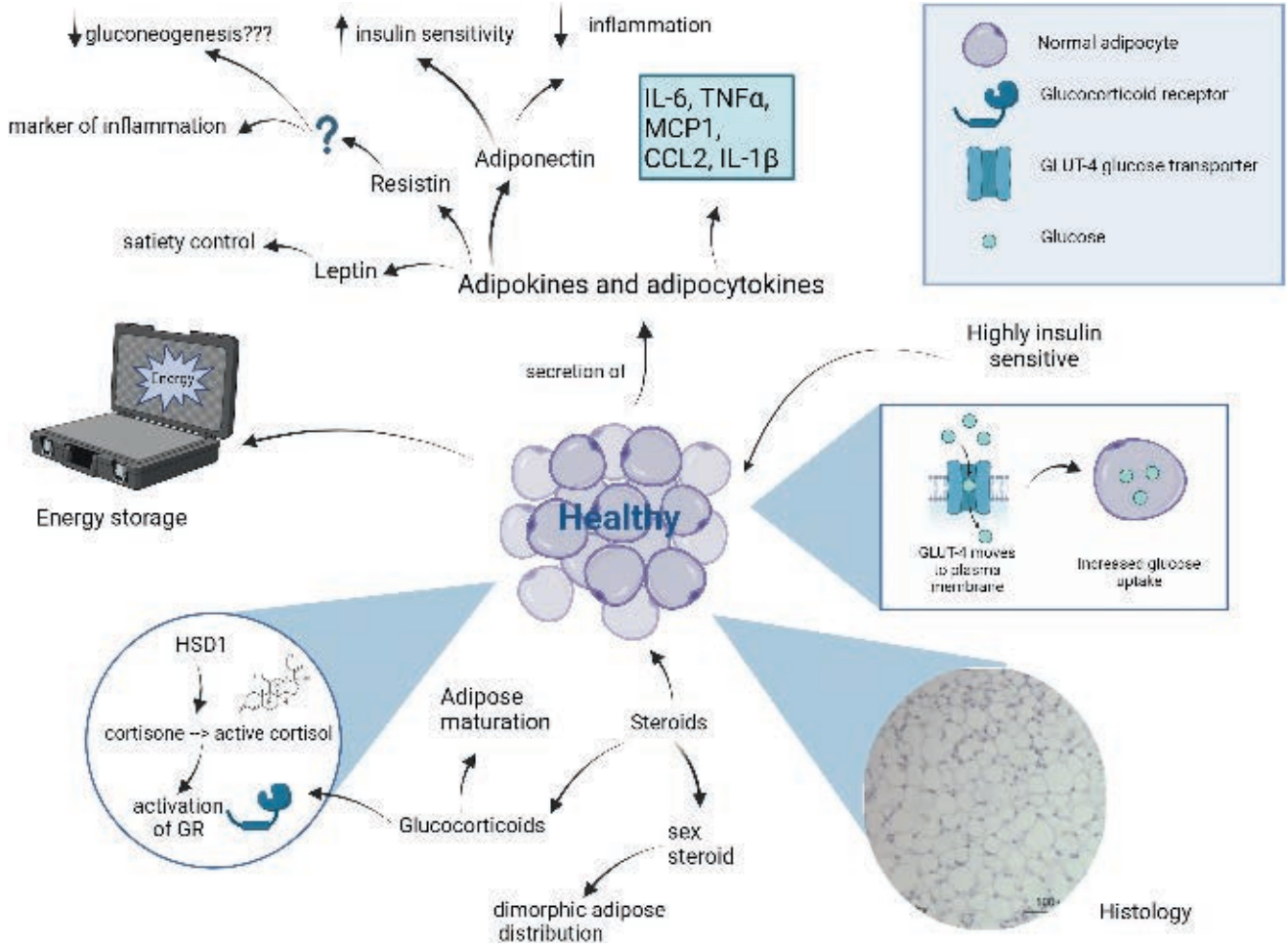


**FIGURE 1** Diagram showing some of the key functions of adipose tissue. Created with BioRender.com.

and receptors in equine AT, though sexually dimorphic patterns of AT distribution are noted (such as the large nuchal crest adipose depots common in stallions) suggesting androgens may impact AT deposition.

### Insulin

Adipocytes are exquisitely sensitive to the effects of insulin. Binding of insulin to its receptor initiates the movement of the insulin-sensitive glucose transporter GLUT4 from cytosolic sequestration to the plasma membrane to allow uptake of glucose into the adipocyte. Adipocyte GLUT4 trafficking not only keeps substrate available for DNL but also seems to mediate the cross-talk of AT with liver and muscle to maintain systemic glucose homeostasis (Abel et al., 2001). GLUT4 trafficking is evident in equine adipocytes, but there is a suggestion that more complex and additional insulin signalling pathways may also contribute to glucose uptake (Warnken et al., 2017). Insulin also suppresses lipolysis in equine and human adipocytes (Duncan et al., 2007; Warnken et al., 2017) and this acts to regulate the availability of free fatty acids and glycerol for hepatic gluconeogenesis.

## HEALTHY ADIPOSE TISSUE

### Not all adipose tissue is created equal

The anatomical location of AT appears to impart several important characteristics, most likely due to the slightly different role AT plays in each different site. The simplest distinction is that of subcutaneous AT (SAT), which lies immediately underneath the skin and visceral AT (VAT) which surrounds organs. In addition to these two major categories, AT within the bone marrow, around the heart (epicardial) and blood vessels and in/around skeletal muscle have all been found to have distinct phenotypes in humans and rodents. Very little is known about these tissue-specific depots in the horse, so this review will focus on what is known about SAT and VAT.

#### Subcutaneous versus visceral adipose tissue

The distinction between SAT and VAT is important because of the independent association of VAT with increased metabolic risk. People who carry their weight around their femorogluteal region (SAT),

which is the predominant pattern seen in women, are relatively protected from metabolic risk in obesity (Booth et al., 2014). In contrast, those who predominantly lay down VAT are at increased risk (Kwon et al., 2017). The reason for this different impact on metabolic health is not clear, but several features of SAT and VAT have been implicated in the sparse data available:

- SAT has a greater capacity for expansion by hyperplasia, which results in more abundant but smaller and 'healthier' adipocytes. VAT, however, favours hypertrophy for expansion, which results in large and less healthy adipocytes, which can more readily induce lipolysis and release fatty acids into the circulation or directly to the liver via the portal circulation (Bergman et al., 2006). A similar pattern of adipocyte size is found in horses, with peri-renal and retroperitoneal adipocytes having a significantly larger cross-sectional area than nuchal adipocytes (Bruynsteen et al., 2013).
- In humans, VAT has a more inflammatory phenotype when compared with SAT. This is the case both in 'normal' individuals and those in the face of persistent caloric excess (Ibrahim, 2010). In horses, there was increased mRNA levels of inflammatory cytokines in visceral depots compared with the nuchal depot (Bruynsteen et al., 2013).
- Leptin secretion is greater from SAT relative to VAT in humans and horses (Bruynsteen et al., 2013). In humans, adiponectin expression is higher in SAT than VAT (Fain et al., 2004) but the opposite is found in horses (Bruynsteen et al., 2013). There is also some evidence in humans and horses (Warnken et al., 2017) that SAT is more insulin sensitive than VAT, potentially contributing to 'safer' storage of lipids.

This distinction between SAT and VAT is relevant in horses because clinical measures of AT depots (body condition scoring, weigh tapes) almost exclusively measure SAT. It is important to recognise this as a limitation and remember that VAT may be more important in predicting disease risk. Body condition score is only strongly correlated with total body fat as determined by deuterium oxide dilution (eTBF%) (Dugdale et al., 2011) in lean or nonobese horses. As BCS increases, the predictive ability of BCS for adiposity reduces significantly (Dugdale et al., 2012). In addition, reliance on BCS may lead one to miss horses with body fat carried almost exclusively around the viscera, a state referred to as Thin on the Outside Fat on the Inside (TOFI) in human medicine (Thomas et al., 2012). This accumulation of VAT, which remains undetected by standard assessment techniques, could explain why outwardly nonobese horses and ponies may demonstrate an insulin-dysregulated phenotype. This explanation may enable owners to begin to understand why their horse is at risk of diseases associated with increased adiposity without appearing grossly overweight. Equally, some horses and ponies that are outwardly obese, do not demonstrate an insulin-dysregulated phenotype. Whilst we cannot yet use cross-sectional imaging techniques that are employed in human medicine, such as MRI, to identify and characterise visceral adiposity, it is worth considering abdominal ultrasound in horses if a TOFI phenotype is suspected.

## Adipose tissue in obesity

Obesity is defined by the World Health Organisation (WHO) as 'abnormal or excessive fat accumulation that may impair health.' In horses, we have yet to define this parameter in a similar manner, so our definition of obesity is based solely on measures of subcutaneous fat. We do know that in obese horses, as defined by BCS, AT can account for up to 35% of body mass (Dugdale et al., 2012).

Obesity-associated AT, particularly VAT, is markedly dysfunctional in humans and directly related to the development of obesity-associated morbidities, including insulin resistance and cardiovascular risk (Santillana et al., 2023). In horses, dysfunction of obese AT is also clear (Figure 2). Reynolds et al. (2019) found marked hypertrophy of VAT adipocytes, a hallmark of dysfunctional AT in humans. Fibrosis, a key feature of human dysregulated AT, was not found in the AT of these horses but has been reported by others (Basinska et al., 2015); this discrepancy perhaps due to the chronicity of disease in the animals studied. Dysfunctional AT has also been shown to display increased expression of leptin (Reynolds et al., 2019). There is conflicting data regarding adipocytokine expression in obese AT of horses, as there is in the human literature. Whilst Burns et al. (2010) showed no change in TNF$\alpha$ and IL1$\beta$ in obese AT, several groups have since shown increases associated with obesity and insulin dysregulation in VAT (Jayathilake et al., 2022; Reynolds et al., 2019) and SAT (Basinska et al., 2015). Basinska et al. (2015) also reported macrophage infiltration. It should be noted that the populations studied are invariably diverse in terms of disease state. In humans, obese AT expansion by hypertrophy fails to stimulate angiogenesis which is typically driven by increased cell number (hyperplasia); therefore, the blood supply eventually becomes limited and hypoxia occurs, contributing to inflammation (Hammarstedt et al., 2018). Although this has not yet been investigated, the same process may occur in the horse. The authors have found the use of the term 'unhealthy' AT useful in discussions with owners on equine obesity.

A key question in understanding how obesity results in systemic insulin resistance is whether obese AT is itself insulin resistant. There is currently not a clear answer to this question in humans, as it appears to be the case in some individuals but not in others. This alludes to the increasing acceptance that many sub-types of insulin resistance syndromes exist (Imi et al., 2023). Very little is known about insulin sensitivity of obese AT in horses. A crude measure of insulin signalling components in AT showed no differences between lean horses and those with equine metabolic syndrome (Reynolds et al., 2019); however, insulin signalling is dynamic and relies on altered phosphorylation state, so much more work is required in this area.

The next question is whether dysfunctional obese AT contributes to whole-body insulin dysregulation. The answer is invariably yes in human and rodent models, although there is still debate over the temporal nature of this relationship (Blüher, 2016; Kahn & Flier, 2000; Kahn et al., 2006). In general, it is thought that factors released in greater or lesser quantities from obese AT perturb insulin signalling; whether that is a function of the insulin receptor

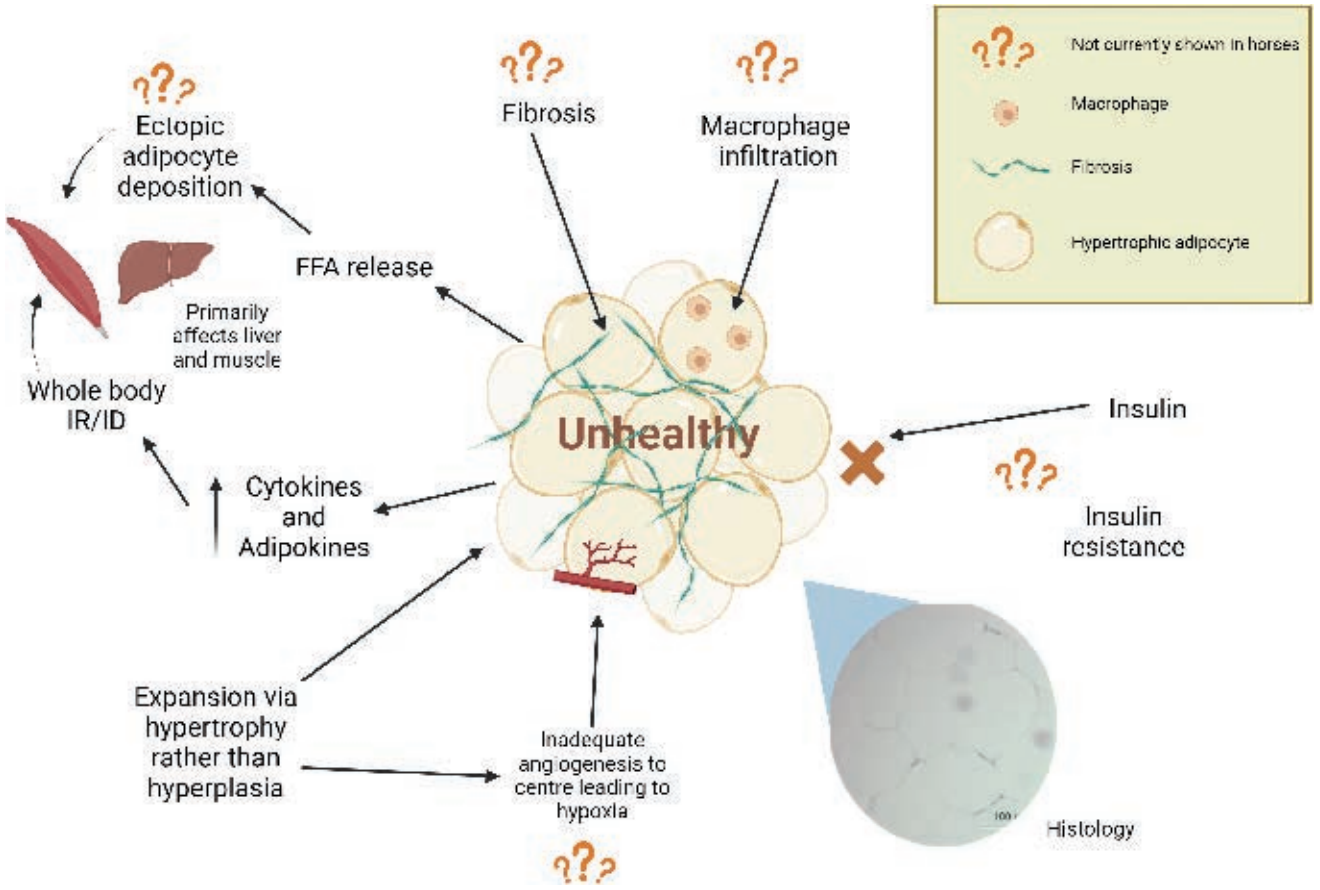


**FIGURE 2** Diagram showing some of the known and proposed effects of unhealthy adipose tissue seen in overweight/obese cases. Created with BioRender.com.

itself or the downstream signalling cascade, particularly in liver and muscle but also in AT, is unclear. These downstream factors include adipocytokines (Blüher, 2016), leptin and adiponectin (Yadav et al., 2013). Overloaded adipocytes, especially in VAT, can release free fatty acids into the portal circulation and overwhelm hepatic gluconeogenesis, thus impacting insulin sensitivity (Longo et al., 2019). If adipocytes have reached capacity for safe storage, then lipid will be deposited ectopically in other sites (such as muscle and liver), which is detrimental to insulin sensitivity in those tissues (Longo et al., 2019).

## CONCLUSION

The aim of this review was to demonstrate our growing understanding of the complexities of AT and its essential role in the morbidity of equine obesity. It is vital that knowledge gleaned from this work should be translated into practical benefits in terms of equine welfare; helping owners and veterinary surgeons to mitigate the impact of obesity on the domestic equine population. The authors have found that it is useful to include discussion of 'unhealthy' AT when communicating with owners about equine obesity. In doing this, the owner should become aware that the horse is not just fat, but that this fat is producing factors, which negatively impact most other organs in the body. By encouraging the practitioner to consider obesity as a disease in its own right, this review hopes to aid discussions with owners and, therefore, improve compliance.

### AUTHOR CONTRIBUTIONS

S. McCullagh is the primary author of this review and was supervised by R. Morgan, who helped with the preparation of the manuscript. J. Keen and M. Dosi reviewed the manuscript and helped finalise it.

### CONFLICT OF INTEREST STATEMENT

No conflicts of interest have been declared.

### ETHICS STATEMENT

No ethical review or approval is required for this review article.

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**How to cite this article:** McCullagh, S., Keen, J., Dosi, M. & Morgan, R. (2024) Bringing equine adipose tissue into focus. *Equine Veterinary Education*, 36, 145–151. Available from: https://doi.org/10.1111/eve.13894

Received: 11 July 2023 | Accepted: 30 October 2023

DOI: 10.1111/eve.13919

Equine Veterinary Education BEVA

REVIEW ARTICLE

# Review of the role of biofilms in equine wounds: Clinical indications and treatment strategies

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## Summary

Recognition of the role that biofilms play in the persistence of wounds and lack of response to therapy in horses is increasing. Prevention of biofilm development in the early stages of wound care involves three primary strategies: wound debridement and cleansing to reduce bacterial counts in the wound bed, appropriate use of advanced wound dressings and implementation of topical antimicrobial agents. Once formed, eradication of biofilms requires elimination to improve the wound environment for contraction and epithelialisation while not further harming the native cells integral to the healing process, which is achieved predominantly through repeated lavage and debridement combined with topical antimicrobial therapy. *Pseudomonas*, *Enterococcus* and *Staphylococcus* bacterial species have been most commonly identified in equine wounds with delayed healing. This review will establish why and how biofilms form, how to recognise clinical indications that biofilms have formed in equine wounds and review current diagnostic options and biofilm-based wound care strategies to eradicate biofilms. Clinical scenarios for cases in which biofilms developed and were successfully treated will be presented. This review will advance practitioners' understanding of the presence and role of biofilms in equine wounds and provide an updated summary of recommended treatment strategies.

**KEYWORDS**
horse, biofilms, wounds

## INTRODUCTION

Bacterial biofilms are organised communities of bacteria attached to a surface and enveloped in a three-dimensional extracellular matrix. A recent Delphi process survey of key opinion leaders in infection was undertaken with the goal to achieve international consensus regarding clinical indicators for chronic wounds, wound infection and biofilm presence to inform the development of international clinical guidance to assess and manage wound infection (Haesler et al., 2019). A chronic wound was agreed upon as one that has a slow progression through the healing phases or delayed, interrupted or stalled healing due to intrinsic and extrinsic factors impacting the individual and their wound, while the actual duration of the wound and original aetiology were not considered necessarily relevant (Haesler et al., 2019). A chronic nonhealing wound was further considered suggestive of biofilm infection, providing that other underlying pathologies such as ischaemia had been excluded (Haesler et al., 2019). While the precise definition of a biofilm has been extensively discussed in the literature (Hurlow et al., 2015; Metcalf & Bowler, 2013; Metcalf et al., 2014), in this consensus, biofilms were agreed upon to be defined as 'a structured community of microbes with genetic diversity and variable gene expression (phenotype) that create behaviours and defences used to produce unique infections (chronic infections)' (Haesler et al., 2019). Relevant to their clinical

management, biofilms were further considered to be characterised by significant tolerance to antibiotics and biocides while remaining protected from host immunity (Haesler et al., 2019).

A systematic review and meta-analysis of wound care literature in humans reported the prevalence of biofilms in wounds with delayed healing to be 78.2% (Malone, Bjarnsholt, et al., 2017; Malone, Johani, & Jensen, 2017). Consensus guidelines for the identification and treatment of biofilms have further stated that biofilms should be assumed to be involved in most, if not all, nonhealing wounds (Schultz et al., 2017) and the majority of medical device-related infections (Khatoon et al., 2018). In addition, multiple studies to date have documented the evidence of biofilms in nonhealing wounds of horses specifically (Freeman et al., 2009; Westgate et al., 2011). Furthermore, there has been a progressive development of antimicrobial resistance in microorganisms across human and veterinary healthcare sectors, which has been considered by the World Health Organization to be one of the leading causes of illness and mortality in the 21st century (Rippon et al., 2023). The high prevalence of biofilms in wounds with delayed healing, the frequency with which equine practitioners treat wounds in daily practice and the increasing reported incidence of multi-drug resistant bacterial strains in equine practice in general (Herdan et al., 2012; Loncaric et al., 2014; Theelen et al., 2014; van den Eede et al., 2012) highlight the need for implementation of more advanced training in wound care strategies to address these clinical scenarios.

The purpose of this review is to summarise the current literature describing problems caused by bacterial biofilms in wounds, clinical indications that biofilms are involved, laboratory testing to improve biofilm detection and biofilm-based wound-care (BBWC) strategies to provide clinicians in general and referral practice with practical guidelines for case management where biofilms are suspected. Recommendations for antimicrobial duration in veterinary practice further support administration for the shortest effective duration to reduce risk of development of resistant pathogens (Gandini et al., 2022; Hansen et al., 2014); therefore, local surgical and topical techniques to address biofilm formation will be emphasised to minimise unnecessary systemic antimicrobial administration. Early recognition of the presence of biofilms in nonhealing wounds and targeted treatments are key to the successful management of biofilms in equine practice (Pezzanite et al., 2021).

## UNDERSTANDING THE ROLE OF BIOFILMS IN WOUNDS

Biofilm formation is divided into three main stages: bacterial attachment, growth and detachment (Lappin-Scott & Bass, 2001). In stage 1, planktonic (free-floating) bacteria adhere to surfaces within several minutes (Parsek & Greenberg, 2005). In stage 2, individual attached bacteria (i.e., 'sessile') secrete a three-dimensional extracellular matrix (also known as extracellular polymeric substance [EPS]) that includes water, proteins, glycolipids, polysaccharides, bacterial DNA and potentially other microbes benefiting from the protected environment which makes up 90% of the biomass of the biofilm itself (Clutterbuck et al., 2007; Flemming & Wingender, 2010; Jacques et al., 2010; Percival, McCarty, & Lipsky, 2015; Percival, Vuotto, et al., 2015; Wolcott et al., 2008). This occurs within 6–12 h of attachment, and the biofilm continues to grow based on coordinated cell-to-cell signalling known as 'quorum-sensing' (McCarty et al., 2012; Parsek & Greenberg, 2005; Preda & Săndulescu, 2019). In stage 3, biofilms reach maturity within 2–4 days and shed free-floating planktonic cells, which disperse and attach to other areas of the wound bed (Kostakioti et al., 2013). This cell distribution activates the host immune response, which further stimulates production of exudates that provide nutrients and promote survival of the biofilm (Dart et al., 2017a; Orsini et al., 2017; Stewart & Richardson, 2017), and may lead to additional complications for the host animal, including bacteraemia or bacterial colonisation of distant anatomical sites (Bjarnsholt et al., 2013).

Predisposing factors to wound infection and biofilm formation include the presence of foreign bodies, sequestra and surgical implants, reduced vascular perfusion to the anatomical region, inappropriate antimicrobial sensitivity, radiation or chemotherapy, and the immune status of the patient (age, sepsis, malnutrition or obesity, antibody deficiency, chronic stress, corticosteroid administration or underlying diseases) (e.g., pituitary pars intermedia dysfunction [PPID] or Cushing's disease) (Orsini et al., 2017; Swanson et al., 2022). Strategies to prevent biofilm development in acute wounds include wound debridement and cleansing to reduce bacterial counts and appropriate use of advanced dressings and topical antimicrobial agents. Addressing systemic conditions (e.g., Cushing's disease in horses) may promote more rapid bacterial clearance and healing in immune-incompetent cases as well. Furthermore, the ability of the host's immune response to effectively control microbes decreases as the biofilm matures. As a consequence, infections involving biofilms frequently recur following discontinuation of antimicrobials (Dart et al., 2017b), emphasising that early recognition of treatment of both the wound and the animal's systemic health status is key to successful management.

Wounds with biofilms may not necessarily exhibit signs typically associated with infection (e.g., heat, swelling, pain and redness) besides a deviation from the expected trajectory of healing with prolonged and impaired recovery (Dart et al., 2017a). Specific bacterial species may integrate chromosomal β-lactamase encoding genes, efflux pumps and mutations in target antibiotic molecules to evade host defences. The presence of biofilms has been demonstrated to delay epithelialisation and induce a nonhealing inflammatory state (Schierle et al., 2009; Wolcott et al., 2008). However, it is important to note that polymicrobial biofilms, which are considered more pathogenic than monobacterial colonies, have been reported in multiple types of equine wounds, regardless of timing since initial wounding or origin of the wound itself (surgical vs. traumatic) (Freeman et al., 2009; Pastar et al., 2013; Westgate et al., 2011). Metabolically active, nondividing persister cells, which are tolerant to antimicrobials, are integral to re-establishing biofilms following topical treatments (Kostakioti et al., 2013). Finally, extracellular DNA (eDNA) present in bacterial biofilms promotes acid–base interactions between bacterial cells and surfaces, therefore playing an essential structural role in both establishing biofilms and protecting cells

within the biofilm from environmental challenges (Lewenza, 2013; Thomann et al., 2016).

Locally, polymicrobial infections delay wound closure through alteration of cytokine levels and receptors (Pastar et al., 2013). For example, *Staphylococcus aureus* and *Pseudomonas aeruginosa* are known to downregulate keratinocyte growth factor 1 expression of fibroblasts, resulting in delayed re-epithelialisation through reduction in keratinocyte migration and proliferation (Pastar et al., 2013). Bacteria in biofilms secrete enzymes (e.g., proteases, elastase and phospholipase) to degrade local host tissues to provide nutrients and protect bacteria within the biofilm from host immune cells (Flemming & Wingender, 2010; Michałkiewicz et al., 1999). For example, the proteases secreted by *P. aeruginosa* degrade and inactivate interferon gamma, which suppresses innate immune recruitment and reduces elimination of biofilm bacteria (Michałkiewicz et al., 1999). Continuous exudate production is detrimental to wound healing as the inflammatory process continuously breaks down the extracellular matrix (McCarty et al., 2012) and may degrade growth factors associated with normal wound healing processes (Percival, McCarty, & Lipsky, 2015; Percival, Vuotto, et al., 2015). Various cell types, including keratinocytes, fibroblasts, endothelial cells and inflammatory cells (e.g., monocytes, lymphocytes and macrophages), express matrix metalloproteinases (MMPs) involved in epithelial repair, wound contraction and degradation of damaged ECM within the skin (Caley et al., 2015), which is upregulated in wound edge keratinocytes to allow epidermal cell migration across wound beds (McCarty et al., 2012). However, in wounds associated with biofilms, the presence of devitalised tissue and abnormal immune cell activity results in excessive production of MMPs, which perpetuates ECM destruction, propagating the inflammatory response and wound chronicity (Caley et al., 2015; Kandhwal et al., 2022; Parnham & Bousfield, 2018). Approaches to restore normal wound healing involve techniques directed towards inhibition of these biofilm virulence factors through effective, sustained debridement of devitalised tissues (Parnharm & Bousfield 2018; Schierle et al., 2009).

Development of infection involving biofilms has important implications in wound management, as they present unique challenges in diagnosis, and are more resistant to typical treatment methods (Dart et al., 2017a). Bacteria that produce biofilms can survive and grow at slower metabolic rates in environments depleted of nutrients and oxygen, termed phenotypic heterogeneity (Clutterbuck et al., 2007; Donlan, 2001a, 2001b; Donlan et al., 2001). Mature biofilms secrete protective enzymes, shielding themselves from host defences and exterior physiological changes that may be detrimental to bacterial health (Percival, McCarty, & Lipsky, 2015; Percival, Vuotto, et al., 2015). Once formed, bacteria in biofilms differentiate into complex communities with enhanced resistance to environmental challenges (e.g., cells of the innate immune system and desiccation), biocides and antibiotics (Costerton et al., 1999; Fux et al., 2005) and variable morphology depending on nutrient availability (Flemming & Wingender, 2010; Klausen et al., 2003). As a result, bacteria within biofilms are more tolerant to the host immune response, antimicrobial therapy administered systemically (antibiotics) or topically (antiseptics), including hydrogen peroxide, alcohols, bleach, oxygen radical generators and acids (unless administered at concentrations toxic to the animal's cells) (Clutterbuck et al., 2007). For example, *S. aureus* has been shown to be up to 100 times more resistant to antimicrobials when in biofilm versus planktonic form (Leid et al., 2002). These challenges in addressing bacteria in biofilms may only be overcome if antimicrobials to which the bacteria are sensitive can be delivered at adequate concentrations for a sufficient time to achieve bactericidal activity (Stewart & Richardson, 2017).

## DIAGNOSING BIOFILMS—LABORATORY TESTING AND CLINICAL INDICATIONS

Traditional bacterial culturing techniques are generally considered inadequate to comprehensively identify bacterial species associated with biofilms (Kirketerp-Moller et al., 2008). Classic antibiotic susceptibility tests that provide the minimum inhibitory concentration (MIC) used to define susceptibility breakpoints that predict therapeutic success are performed with planktonic growing bacteria and the results cannot be considered predictive for biofilm infections (Doring et al., 2012). Methods described to detect biofilm production include tissue culture plate (TCP), tube method (TM), Congo red agar method (CRA), bioluminescent assays, piezoelectric sensors and fluorescent, confocal or electronic microscopy examination (considered the gold standard), which may not be available to clinicians (Aparna & Yadav, 2008; Christensen et al., 1982, 1995; Dart et al., 2017a, 2017b; Donlan, 2001a, 2001b; Donlan et al., 2001; Freeman et al., 1989; Hassan et al., 2011; Hurlow et al., 2015; Percival, McCarty, & Lipsky, 2015; Percival, Vuotto, et al., 2015; Schultz et al., 2017; Wolcott & Rhoads, 2008; Zufferey et al., 1988). In one study, three methods (TCP, TM and CRA) were compared and concluded that TCP was more quantitative and reliable for the detection of biofilm-forming microorganisms; however, all three methods may be performed by clinicians with laboratory facilities to perform bacterial culture, the techniques of which have been described in detail elsewhere (Hassan et al., 2011). More recently, a 'wound blotting' method that involves attaching a piece of nitrocellulose membrane to the wound surface followed by staining with Alcian blue, a dye specific to mucopolysaccharides found in biofilms, has been described as a noninvasive point-of-care test to identify biofilms in human wounds and further guide debridement strategies, but is not commercially available currently (Minematsu et al., 2013).

Recent studies have demonstrated that biofilms associated with wounds are most commonly polymicrobial communities, with an average number of $3.02 \pm 1.65$ species identified (range, 0–8) (Freeman et al., 2009; Westgate et al., 2011). Both Gram-positive and Gram-negative bacteria have the capability to form biofilms; in human wounds, bacteria commonly involved include *Enterococcus faecalis*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus viridans*, *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis* and *Pseudomonas aeruginosa* (Donlan, 2001a, 2001b; Donlan et al., 2001). Genera identified in equine wounds are similar to those found in human infections, with *Pseudomonas*, *Enterococcus* and *Staphylococcus* species being most common (Darvishi et al., 2022; Dowd et al., 2008;

James et al., 2008; Wolcott & Rhoads, 2008). However, molecular analyses of samples from wounds with delayed healing have revealed far more diverse polymicrobial communities with up to 17 genera per wound, including anaerobic species not identified by routine culturing, and further highlighting the challenges faced by clinicians in accurately identifying and treating bacterial species contained within biofilms (Han et al., 2011; James et al., 2008).

Standard methods to assess bacterial burden in wounds include qualitative and quantitative techniques (Hendrickson, 2019). Qualitative assessment determines the genera of bacteria found in wounds and is coupled with antimicrobial sensitivity testing to provide clinicians a basis for antibiotic choices in treatment. In polymicrobial infections, as is most typical of those involving biofilms, multiple microorganisms act synergistically to result in greater virulence compared with an infection caused by either species alone (Serra et al., 2015). Quantitative bacteriology methods are less commonly performed in veterinary medicine but should be considered in cases when wound healing is not progressing as anticipated or following skin graft failure. The current gold standard to confirm presence of biofilm in human practice includes tissue biopsy and subsequent microscopic examination (Costerton et al., 2003). The best diagnostic method currently available to clinicians in equine practice when biofilms are suspected is submission of a deep tissue biopsy or swab of the deepest tissues available (or both) for bacterial culture and sensitivity to guide future treatment practices (Dart et al., 2017a, 2017b). Sample submission of swabs versus biopsies for chronic nonhealing wounds has been discussed in both human and veterinary medicine, with conflicting results as to the relative benefit of one technique over another (Concannon et al., 2020; Esposito et al., 2017; Haalboom et al., 2018; Huang et al., 2016; Reddy et al., 2012; Van Hecke et al., 2017). However, several publications have supported that tissue samples, while being more invasive to collect, are more likely to yield reliable culture results compared with swabs and to detect antimicrobial-resistant bacteria in wounds (Esposito et al., 2017; Freeman et al., 2009; Haalboom et al., 2018; Huang et al., 2016; Reddy et al., 2012; Westgate et al., 2011). Specific to horses, wound surface swabs (Levine technique) were recently compared with tissue biopsies to detect methicillin-resistant *Staphylococcus aureus* (MRSA) in experimental equine wounds, concluding that bacterial load and diversity did not differ between techniques, but MRSA was more frequently detected from cultures of tissue biopsies versus swabs (Brock et al., 2022). Ideally, submission of tissue samples should be performed prior to beginning or altering antimicrobial protocols; however, if considered necessary to collect samples while horses are currently receiving antimicrobials, it is recommended to notify the receiving laboratory of the horse's current regimen and when the most recent dose was received in relation to sample collection (Orsini et al., 2017). Following superficial wound debridement, tissue samples should be collected from within the deepest regions of the wound (e.g., fissures or pockets in the wound bed) and from multiple sites if possible, to avoid false-positive results (Rhoads et al., 2012; Sen et al., 2021). If tissue swabs are collected, the swab should be drawn across the wound surface with sufficient pressure to collect the biofilm itself while avoiding drawing blood which contains antimicrobial elements that may affect culture results. Positive culture results should be interpreted with the assumption that the full microbial spectrum is likely underrepresented with currently available techniques.

In the absence of a diagnostic test to confirm presence of biofilm, in lieu of obtaining a positive culture result or if submission is not an option due to financial or other case-related considerations, diagnosis of biofilms in wounds may be based on clinical indicators during wound care assessment (Table 1) (Haesler et al., 2019). Clinical findings consistent with biofilm presence may include one or more indicators such as inflammation (heat, swelling, pain and redness), persistent or recurrent infection despite administration of antimicrobial therapy or recurrence following antibiotic discontinuation, excessive wound moisture/exudate, poor-quality granulation tissue, history of negative culture findings despite clinical suspicion of infection or, in general, a wound that remains in a chronic and recalcitrant inflammatory state despite standard treatment and evaluation of the patient for comorbidities (e.g., immunosuppression).

Examples of images of wounds diagnosed with biofilm infection are provided in Figure 1. If any one of these situations is noted, specifically lack of progression according to the anticipated trajectory of healing in lieu of other clinical findings of inflammation, it is recommended that biofilm involvement within the wound be suspected and treated as such. Whether biofilms can be visualised as a shiny or slimy surface on wounds has been debated in the literature (Hurlow et al., 2016; Lenselink & Andriessen, 2011; Metcalf & Bowler, 2013). Given the physical location of biofilms deep beneath the wound surface and the gold standard for detection of biofilms considered to be microscopy (fluorescence, confocal or electronic), macroscopic visualisation of the wound surface substance should be considered a less sensitive determination of biofilm presence (Fazli et al., 2009; Haesler et al., 2019; International Wound Infection Institute, 2016; Kirketerp-Moller et al., 2008; Metcalf & Bowler, 2013). However, culture findings to diagnose biofilms are unreliable and observation of clinical indicators that biofilms are present in the wound bed should prompt practitioners to implement wound care strategies directed specifically at addressing and reducing biofilm formation in wounds.

## BIOFILM-BASED WOUND-CARE TREATMENT STRATEGIES

### Biofilm-based wound-care guidelines

Recent consensus documents in human wound care have described biofilm-based wound-care (BBWC) strategies to provide practical guidelines for case management in which biofilms are suspected (Bianchi et al., 2016; Metcalf et al., 2014; Schultz et al., 2017; Wolcott & Rhoads, 2008). Biofilm treatment is recommended in three stages: (1) physical debridement of the biofilm, (2) topical treatment to delay or prevent reformation and (3) repeated therapy until full resolution

**TABLE 1** Indirect clinical indicators of wound biofilm (Metcalf et al., 2014; Wolcott et al., 2008).

| Clinical observation | Biofilm explanations for clinical observation |
|---|---|
| Excessive moisture associated with wound | Bacteria in biofilms secrete extracellular matrix and biofilm presence promotes inflammation, resulting in increased exudate |
| Autograft or allograft fails on wounds | Applying tissue grafts over biofilms provides a second growth surface and food source, leading to devitalisation of graft tissue and increased exudate and inflammation |
| Poor-quality granulation tissue (e.g., hypergranular, friable) | Biofilm presence contributes to delayed epithelialisation and is frequently associated with poor-quality granulation tissue |
| Indications of local infection (swelling, sensitivity, redness and heat) | Biofilms promote inflammation and may be a precursor to other clinical indications of infection |
| History of persistent or recurrent infection despite antimicrobial therapy | Biofilm bacterial phenotypes adapt rapidly and may only demonstrate 1 to 2 log reduction with antibiotic therapy at 50 to 1000× MIC. Biofilms contain persister cells that remain once antibiotic therapy is discontinued, seeding and contributing to subsequent biofilm reformation |
| Negative culture results despite clinical suspicion of infection or signs of bacterial colonisation | Biofilm bacteria metabolise more slowly and are phenotypically different from planktonic bacteria. Standard microbiological culture techniques are not capable of identifying all species present, making bacteria in biofilms difficult or impossible to identify by culture |
| Wound remains in chronic inflammatory state and recalcitrant to therapy despite addressing comorbidities | Biofilms are resistant to host inflammatory responses and actually feed off exudate produced by inflammation, further promoting inflammation |

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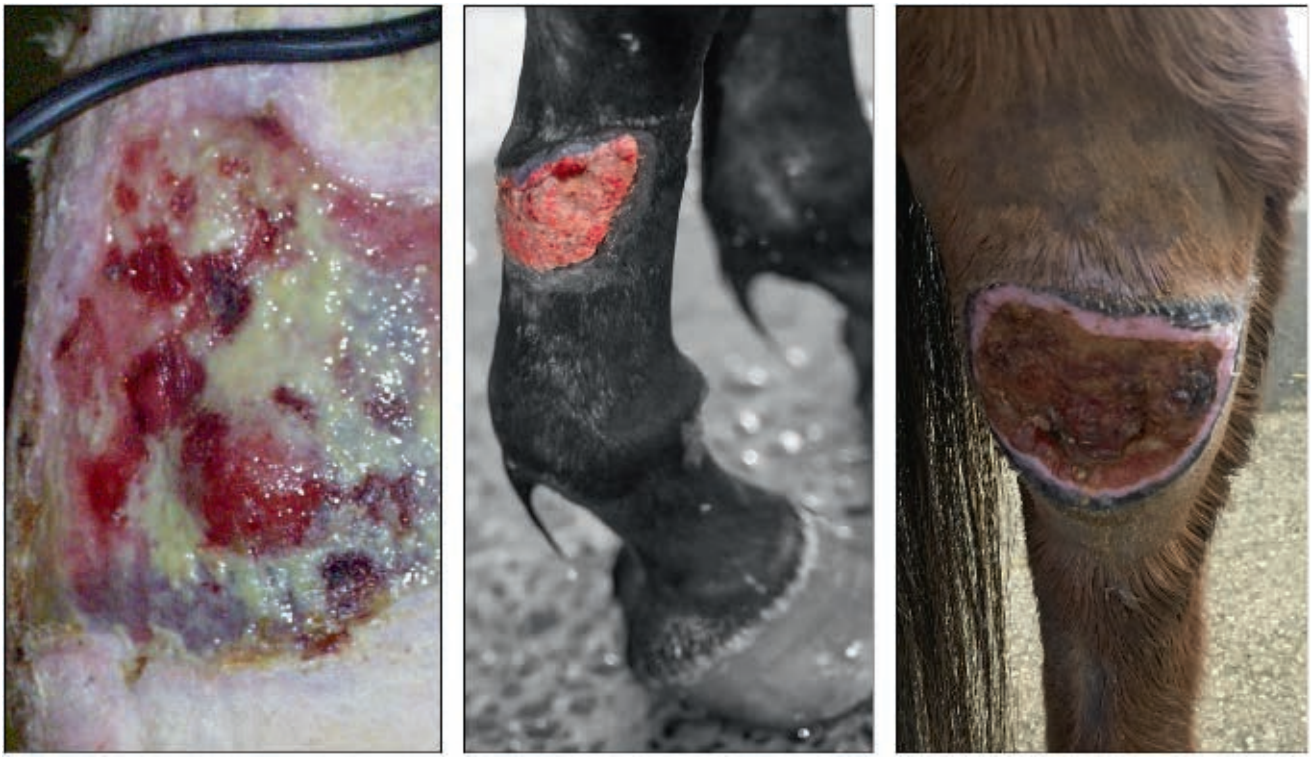

**FIGURE 1** Clinical indicators of wound biofilm. Examples illustrate clinical indicators of wound biofilm including, from left to right, excessive moisture associated with the wound and indications of local infection (swelling, redness and heat), poor-quality granulation tissue (e.g., hypergranular and friable), and a wound remaining in a chronic inflammatory state recalcitrant to therapy despite standard treatments.

is achieved (Orsini et al., 2017). These strategies emphasise that repeated debridement to physically disturb the biofilm structure is necessary to disrupt the matrix and remove devitalised tissues that serve as nutrients to the microbes involved and allow increased susceptibility to antimicrobial therapies for a period of time to prevent bacterial reattachment as immature biofilms are more susceptible to antimicrobials, in addition to physically removing bacteria to reduce the overall bioburden (Dart et al., 2017a, 2017b). Implementation of a multimodal therapeutic strategy to address biofilms has a reportedly higher success rate compared with antimicrobials alone (Wu et al., 2022).

## Debridement principles

The overall objective of debridement is to remove as much devitalised tissue, biofilm and associated extracellular matrix as possible to expose the remaining bacteria to antimicrobial agents. The organisation and complex physiology of mature biofilms increases their resistance to antibiotics, resulting in colonised bacteria being up to 1000-fold times more resistant to antimicrobials than planktonic cells (Høiby et al., 2010). Debridement removes ECM and eDNA to prevent recurrence of biofilms in the wound by removing the basis for nutrition and protection of the bacterial component of the biofilm (Hajská et al., 2014). The immature biofilms that begin to reform following debridement are subsequently more susceptible to topical therapies. General principles described by Wolcott et al. (2010) in addressing wounds infected with biofilms include debridement with the goal to alter the wound bed anatomy by removing any devitalised or discoloured tissue and all tissue surfaces that touch one another until normal bleeding tissue is encountered. Application of topical treatments is then recommended within 4 h following debridement prior to biofilm reformation (Hajská et al., 2014; Roche et al., 2012). An example of how biofilms may be successfully treated and how rapidly they reform in the absence of consistent treatment is daily removal of dental enamel plaque by regular tooth brushing or treatment of clinical periodontitis in humans (i.e., debridement) performed in combination with topical antiseptic mouthwashes, which are of minimal benefit without prior flossing and tooth brushing (Jepsen & Jepsen, 2016; Orsini et al., 2017).

Biofilm debridement may be performed as with any wound, either sharply, mechanically, chemically or enzymatically, biologically, via autolysis or using low-frequency ultrasonic debridement (Crone et al., 2015; Hendrickson, 2019). Sharp debridement (e.g., scalpel blade) is the least traumatic and most cost-effective method available to equine clinicians and should be used whenever possible in the initial wound treatment stages with other methods playing a more secondary role. Mechanical debridement is more traumatic and currently likely the most commonly performed in equine practice via dressings (e.g., gauze across the wound bed, wet-to-wet or wet-to-dry dressings) or lavage using noncytotoxic cleansing solutions delivered at a pressure between 10 and 15 pounds per square inch. One method to achieve this pressure in equine practice is using a 19-gauge needle or catheter to a 35-mL syringe although other methods are available (Rodeheaver, 2001). Chemical debridement with acetic acid (i.e., to treat *Pseudomonas aeruginosa*) or hypertonic saline are nonselective methods and should be reserved for contaminated wounds in the early stages of wound healing and typically following sharp debridement. However, they are nonselective and must be monitored carefully to minimise local cytotoxicity with dressings changed every 24–48 h. Hypertonic saline can be made in practice by mixing 200 g (0.5 cup) of salt to 1 L of hot water. Enzymatic debridement by placing enzymes directly onto the wound bed may be more effective than sharp debridement as less host tissue is removed and has been described to treat human diabetic foot ulcers (Tallis et al., 2013). Commonly used enzymes include collagenase, DNase/fibrinolysin, papain/urea, trypsin and streptokinase/streptodornase (Hendrickson, 2019).

Biological debridement using *Lucilia sericata* (greenbottle fly) larvae provides rapid and relatively selective debridement as larvae digest only the necrotic tissue and pathogenic bacteria, although clients are often reluctant to allow larvae to inhabit the wound (Hendrickson, 2019). There is some evidence that larval secretions may be used for debridement for their antibacterial effects and to promote angiogenesis (Bexfield et al., 2010; Valachova et al., 2014). Finally, autolytic debridement can be achieved by leaving wound fluid containing leukocytes and enzymes from dead leukocytes in contact with the wound bed, which can only occur in moist wounds. It is recommended that sharp debridement be performed before using autolytic debridement as the latter is ineffective in the presence of significant amounts of necrotic material (Hendrickson, 2019). A combination of identification of biofilms using 'wound blotting' techniques with Alcian blue dyes followed by sharp and ultrasonic debridement to promote wound healing was recently described in human practice and demonstrated to be a promising therapeutic strategy to specifically identify and selectively debride biofilms in wounds not accompanied by necrotic tissue (Mori et al., 2019).

In terms of practical considerations to perform debridement in equine clinical practice, it is recommended that horses be sedated, and if necessary, the region desensitised with local or regional anaesthesia to facilitate procedures and reduce discomfort to the case, although this may not be necessary in all cases (e.g., simple granulation tissue debridement). In some cases, the initial debridement may be performed under general anaesthesia in the field or hospital setting if the wound is extensive or inaccessible or if dictated by the case's temperament. When working with multi-drug resistant organisms or particularly when using pulsed water-jet irrigation, face protection or use of surgical masks during the debridement stage is recommended to protect against aerosolised organisms. Debridement and efforts to reduce biofilm reconstitution should be repeated daily to at least every other day for as long as necessary to resolve infection. Mature biofilms reform as rapidly as every 24–72 h after debridement, resulting in a window of opportunity to impede regrowth in which topical therapies, and bactericidal drugs can exert an enhanced effect. If improvement is not observed within 3–4 days of initiation of the multimodal therapeutic approach outlined or if response to therapy is less than anticipated, review of all aspects of the case is indicated. These may include repeated physical examination, haematology, evaluation of antibiotic suitability with repeated bacterial culture and sensitivity, and further debridement and exploration of the wound and potentially additional diagnostic imaging to evaluate for alternate reasons for delayed healing (e.g., foreign material).

## Antimicrobial guidelines

Antimicrobials refer to agents that reduce the possibility of infection and sepsis, including antibiotics, antiseptics, disinfectants and

antifungals. Antibiotics, made synthetically or derived from moulds, may be administered systemically or topically and are absorbed in the body with the goal to kill bacteria (bactericidal) or limit their replication (bacteriostatic). Antiseptics are substances applied topically that may reduce the possibility of infection but are not significantly absorbed systemically. Less important in the context of biofilm treatment, disinfectants kill microorganisms on nonliving objects and antifungals are drugs that kill or inhibit growth of fungi, including yeasts.

Biofilms are recalcitrant to antibiotic treatment due to multiple tolerance mechanisms (phenotypic resistance) causing persistence of biofilm infections despite exposure, which predisposes them to the development of antibiotic resistance (genetic resistance) (Ciofu et al., 2017). Factors associated with biofilm tolerance to antibiotics include restricted penetration of antibiotics, restricted growth at low-oxygen tension, expression of biofilm-specific genes and presence of persisters (Ciofu et al., 2017). Acquisition of certain chromosomal mutations can also compromise antimicrobial susceptibility of bacterial biofilms (Ciofu et al., 2017). Antimicrobial drug resistance is a critical issue in veterinary and human medicine, with a body of emerging data supporting shorter duration of antimicrobial prophylaxis and greater consideration for the development of antimicrobial stewardship programmes (Southwood, 2023). In general, BBWC strategies to treat wounds with delayed healing emphasise the importance of local debridement and topical antimicrobial (i.e., antibiotics or antiseptics) and surfactant administration versus systemically administered antibiotics. Topical administration of antibiotics provides high local concentrations directly to the site of infection with low to undetectable serum concentrations, thereby avoiding systemic side effects (Ciofu et al., 2017). Furthermore, this route of administration has been suggested to decrease the risk of developing resistance and may effectively reduce bacterial counts normally considered resistant as antibiotic concentrations remain well above MIC (Ciofu et al., 2017). Topical antibiotic agents can also have minimal negative side effects on wound healing depending on the vehicle and dose used and provide efficacy against bacteria in the wound bed when administered following debridement and based on results of culture and sensitivity. However, dose-dependent cytotoxicity has been demonstrated following co-culture of antibiotics with multiple equine cell lines (Pezzanite et al., 2021), indicating that antibiotic selection and evidence-based dosing may minimise local collateral tissue damage if used topically in equine wounds.

While appropriate to treat planktonic infections in acute wounds and/or immediately following debridement, the benefit of systemic antibiotics to treat biofilms remains controversial. In treatment of human periodontitis as an example of clinical biofilm formation, a recent meta-analysis did however support that the use of adjunctive systemic antibiotics could be clinically relevant in specific situations, such as patients with deep periodontal pockets, progressive 'active' disease or specific profiles (Herrera et al., 2002). A tendency for improved clinical outcomes was furthermore found in studies in which systemic antibiotics were used as an adjunctive therapy with scaling, but due to the high level of heterogeneity, authors could not establish definitive guidelines regarding the use of systemic antimicrobials (Herrera et al., 2002). A summary of randomised clinical trials and systematic reviews in human periodontitis further support that adjunctive systemic antibiotics, when combined with mechanical debridement, offer clinical improvements additional to those obtained with scaling and root planning alone (i.e., local debridement), which were more pronounced in aggressive versus chronic disease and those with deep tissue pockets (Jepsen & Jepsen, 2016). This review further supported the well-documented positive effects of local antimicrobial administration (Jepsen & Jepsen, 2016). If systemic antibiotics are instituted in the treatment of wound biofilms, contaminated wounds are more appropriately treated with bactericidal versus bacteriostatic antibiotics. Although ideally dictated by culture and sensitivity findings, broad-spectrum therapy is generally instituted initially with agents such as penicillins, cephalosporins and aminoglycosides (Orsini et al., 2017). Commonly used initial combinations include penicillin G (crystalline or procaine penicillin) or a cephalosporin (cefazolin or ceftiofur) and an aminoglycoside such as gentamicin. Collection of a separate sample to perform in-house point-of-care gram staining may help to guide interim therapy in lieu of culture and sensitivity findings.

In treatment of distal limb wounds, antibiotics can also be delivered via regional limb perfusion (RLP). Despite the frequency of use and large number of studies investigating drug pharmacokinetics and side effects associated with RLPs of antibiotics, there remains a lack of evidence supporting whether RLP makes a difference in prophylaxis or treatment of equine orthopaedic infections (Rubio-Martinnez, 2021). The clinical benefits of RLP are difficult to confirm or compare between studies due to the lack of standardisation or determination of optimal methods of technique in terms of drug selection, perfusate volume and concentration, duration of tourniquet application and whether the procedure is performed under general anaesthesia or standing sedation (Biasutti et al., 2021). However, greater evidence for efficacy is reported in human orthopaedic surgery in clinical scenarios where biofilm formation would be suspected including chronic osteomyelitis (Finsterbush & Weinberg, 1972), infected total knee arthroplasty (Lazzarini et al., 2003) and ischaemic diabetic nonhealing pedal ulcers (Agarwal et al., 2005; Seidel et al., 1994). When examining variables associated with RLP administration in equine practice in a recent meta-analysis utilising multivariable meta-regression, the use of wide rubber tourniquets and concurrent local analgesia were identified as factors that resulted in significantly greater local concentrations of antibiotic (Redding et al., 2022). On univariable meta-regression, high perfusate volume (100–120 mL) was further demonstrated to result in significantly higher concentrations than low volume (30 mL or less) (Redding et al., 2022). Therapeutic concentrations achieved across a variety of techniques ($C_{max}$: MIC ≥ 10) would be considered adequate for susceptible but not resistant pathogens, as may be the case in many wounds containing biofilms (Redding et al., 2022). However, if implemented, antimicrobial concentrations delivered locally via RLP are greatest immediately following biofilm degradation, so timing of perfusion

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to directly follow debridement may improve outcomes although further investigation is indicated. Additional studies standardising techniques and investigating the use of RLP specifically in the context of wounds with delayed healing in horses are warranted.

## Topical therapies to prevent biofilm reformation

Reduction or prevention of biofilm reformation following debridement may be achieved in multiple ways. Topical antiseptic agents do not penetrate necrotic debris and have minimal effect to reduce bacterial populations deep in the wound bed or without debridement; therefore, they should generally be reserved for use on intact skin and in wound beds (Alves et al., 2021). Examples of antiseptic agents that may be contraindicated for use in biofilm-associated wounds due to described local cytotoxicity include alcohols, hydrogen peroxide, chlorhexidine, aluminium salts, boric acid, formaldehyde, hexachlorophene, hypochlorite, merthiolate or permanganate, although this warrants further investigation. A recent article discussed the role of antiseptics in management of chronic wounds and biofilm, further supporting a time-dependent 'window of opportunity' after wound debridement during which biofilms are increasingly susceptible to treatment, particularly topical antiseptics (Alves et al., 2021). The mode of action and clinical significance of currently available antibiofilm products, including surfactants, dressings and others have been recently reviewed and are summarised below (Weigelt et al., 2021).

### Polyhexamethylene biguanide

Polyhexamethylene biguanide (PHMB) is a cationic polymer that can be used as an adjunctive therapy in the early post-debridement period to reduce biofilm surface tension to increase their permeability (Hendrickson, 2019; Palumbo et al., 2016; Percival et al., 2019; Rippon et al., 2023; Weigelt et al., 2021). PHMB is a broad-spectrum biocide that kills Gram-positive and Gram-negative bacteria, fungi, parasites and certain viruses with a high therapeutic index, and has further been used for years clinically and in industry without development of resistance (Rippon et al., 2023; Weigelt et al., 2021). PHMB is a strong base, which is positively charged at physiological pH (Fjeld & Lingaas, 2016). Furthermore, it has been shown to be noncytotoxic nor damaging to epithelialising tissue in healing wounds, with substantial evidence reviewed elsewhere for its application in wound management and importantly, its alignment with recently described antimicrobial stewardship policies to prevent and treat wound infection (Rippon et al., 2023). PHMB is the most used antiseptic to treat critically colonised and locally infected wounds (Alves et al., 2021; Dissemond et al., 2010) and is available in equine practice bound to roll gauze, sponges or other types of dressings (Hendrickson, 2019). Although its antibacterial properties are well-documented, further investigation of its use as an antibiofilm agent in higher powered clinical studies focusing on direct measurement of biofilm burden is warranted (Weigelt et al., 2021).

### Poloxamer-based surfactants

Surfactants are amphiphilic compounds that reduce surface tension between substances and are commonly used as cleaning agents to remove debris from clothes, skin or other materials (Weigelt et al., 2021). In the context of biofilm formation, they interfere with the ability of microorganisms to adhere to surfaces and each other. Polaxamers are nonionic surfactants with a hydrophobic core and two hydrophilic ends. In one study, surfactant-based dressings reduced biofilm, resulting in reduction of IL-6 and TNF-a, and enhanced cellular viability and migration (Salisbury et al., 2018).

### Silver-containing products

Silver-containing products (e.g., silver salts, nanoparticles and colloidal silver) exhibit antimicrobial activity against Gram-negative and Gram-positive bacteria and may also target fungi and viruses through release of positively charged silver ions (Jung et al., 2008; Kang et al., 2019; Kramer et al., 2018; Liao et al., 2019; Mallmann et al., 2015; Pavlik et al., 2019; Percival et al., 2016; Vila Dominguez et al., 2020; Xiang et al., 2011). However, bacterial resistance to silver has been documented, which has been suggested to be encoded on plasmids (Finley et al., 2015; Panacek et al., 2018; Sutterlin et al., 2012; Xiang et al., 2011). Topical dressings such as silver sulfadiazine (1%) or other silver-impregnated wound dressings may be used as early first-line options in the post-debridement stage to reduce biofilm reformation, particularly if bacterial culture and sensitivities to guide topical antimicrobial treatments are not available (Fey, 2010; Gunasekaran et al., 2012; International Wound Infection Institute, 2016; Morones et al., 2005; World Union of Wound Healing Societies, 2016). Silver works through interacting with ribosomes to suppress enzymatic expression and protein formation essential for ATP production (Gunasekaran et al., 2012; Yamanaka et al., 2005). In addition, silver enhances re-epithelialisation, angiogenesis, deposition of collagen fibres and myofibroblast distinction from fibroblasts prompting wound contraction (Toczek et al., 2022).

### Iodine

Povidone iodine is an iodine-releasing agent or iodophor with a neutral polymer base and a complex of iodine that acts as a reservoir of free active iodine (Alves et al., 2021). Iodine has a rapid onset of activity with antimicrobial efficacy against Gram-positive and Gram-negative bacteria, fungi, protozoa, viruses, amoeba and bacterial spores after a contact time of 1 min (Deshmukh et al., 1998; Koburger et al., 2010). No reports of bacterial resistance to iodine

exist despite more than 150 years of use, perhaps due to multiple mechanisms of action exhibited by free iodine (Cooper, 2007; Durani & Leaper, 2008; Eggers, 2019; Sibbald et al., 2011). A recent review article comparing the potential application of silver, polyhexanide (PHMB) and povidone-iodine (PVP-I) as topical antiseptics to treat biofilms concluded that PVP-I had a broader spectrum of antimicrobial activity, no acquired bacterial resistance or cross-resistance, and potent antibiofilm efficacy, as well as relatively low cytotoxicity and good tolerability (Alves et al., 2021).

Cadexamer iodine (CDI) is a more recently described spherical starch bed lattice containing 0.9% iodine by weight that slowly releases iodine as the beads come into contact with exudate (Akiyama et al., 2004). CDI has demonstrated efficacy to disrupt mature biofilms in vivo and in murine and porcine models and human patients with diabetic foot ulcers (Akiyama et al., 2004; Malone, Bjarnsholt, et al., 2017; Malone, Johani, & Jensen, 2017; Roche et al., 2019). However, a recent Cochrane review evaluating four randomised controlled clinical trials using CDI when compared to silver, dextanomer, paraffin gauze or hydrocolloid dressings, which warrants further investigation (O'Meara et al., 2014).

### Manuka honey

Manuka honey has been used in wound healing for centuries and exhibits antimicrobial properties due to its high osmolarity, low pH (3.2–4.5) and high methylglyoxal and leptosperin content and may be used as an adjunctive topical antimicrobial therapy against a variety of bacterial species with minimal host cytotoxicity (Hendrickson, 2019; Liu et al., 2017; Molan & Rhodes, 2015; Weigelt et al., 2021). Peroxide produced during the breakdown of glucose further exerts antimicrobial effects and promotes angiogenesis (Hixon et al., 2019). Honey has been extensively reviewed for its anti-inflammatory and debridement effects elsewhere (Scepankova et al., 2021; Yilmaz & Aygin, 2020) and further demonstrated to inhibit biofilm formation and eradicate mature *Pseudomonas aeruginosa* biofilms (Lu et al., 2019). Its demonstrated spectrum of activity against *P. aeruginosa*, *S. aureus*, *E. coli*, *Acinetobacter*, as well as some antibacterial-resistant strains including methicillin-resistant *S. aureus* and vancomycin-resistant *Enterococcus* are further relevant to those identified in biofilms (Yilmaz & Aygin, 2020).

### Hypochlorous acid

Hypochlorous acid (HOCL) is an antiseptic agent with broad activity against bacteria, fungi and viruses through disruption of chemical linkages (e.g., sulfhydryl enzyme oxidation or ring chlorination of amino acids) (Sakarya et al., 2014; Weigelt et al., 2021). While HOCL has been reported to disrupt biofilm structures at high concentrations in vitro, no studies have evaluated biofilm-dispersing abilities in vivo or clinical settings to the authors' knowledge. A recent expert consensus panel recently found HOCL to be of low evidence for chronic wounds and those of mixed aetiologies (Armstrong et al., 2015).

### Antimicrobial wound gel

Recently, a high osmolarity surfactant antimicrobial wound gel (BlastX™, AWG, Next Science) has been introduced as a biofilm intervention and approved by the US Food and Drug Administration as a medical device (Weigelt et al., 2021). The gel is a combination of benzalkonium chloride 0.13% as a cationic surfactant, polyethylene glycol hydrogel base to maintain a moist wound environment to support granulation, epithelialisation and reduction in tissue necrosis, sodium citrate, citric acid and water. Of note, the gel should not be used in combination with calcium alginate dressings as they destabilise biofilm matrix through chelation of calcium, and in consequence, would neutralise each other's effects (Kim et al., 2018; Wolcott, 2015).

### Plasma

Finally, topical application of plasma (autologous natural plasma or hyperimmune plasma to target specific organisms) may provide additional benefit as a topical therapy as plasma inhibits bacterial adhesion and growth (Bauer et al., 2004; Felts et al., 2000; Lopez et al., 2014).

## Other treatment modalities

Recent data suggest adjunctive approaches to debridement and topical therapies may represent innovative approaches to combat biofilm infection, such as low-frequency ultrasound, phototherapy and lasers, and energy-based technologies, summarised here (Weigelt et al., 2021).

### Low-frequency ultrasound

Low-frequency ultrasound (20–50 kHz) has been used as a debridement modality for chronic wounds through emulsification of necrotic tissues via microstreaming and cavitational effects with gas microbubbles (Chang et al., 2017). Evidence supporting low-frequency ultrasound as an alternative or adjunctive treatment to sharp debridement in nonhealing wounds has been recently reviewed and appears promising although further exploration in randomised controlled trials with larger sample sizes is indicated (Chang et al., 2017).

### Phototherapy/lasers

Blue light lasers demonstrate antimicrobial and antibiofilm effects due to induction of oxidative stress or impairment of polysaccharide

production in fungi (Rupel et al., 2019). Blue laser light has been demonstrated to destroy *P. aeruginosa* biofilms in vitro and in vivo with minimal local cytotoxicity as well as to prevent in vitro *Candida albicans* biofilm formation (da Silveira et al., 2019; Rupel et al., 2019).

### Energy-based technologies ('electroceuticals')

Energy-based technologies generate electrical currents or fields (Banerjee et al., 2015). A recent review reported covering 14 randomised controlled trials demonstrated that use of electrical stimulation was associated with greater rates of wound closure and area reduction in diabetic foot ulcers, venous leg ulcers, pressure and mixed ulcer types (Thakral et al., 2013). In the context of biofilm treatment, the observed effect of electroceuticals may be through disruption of the weak electrostatic forces necessary for bacterial adhesion, production of superoxide and disruption of bacterial membrane enzymes (Banerjee et al., 2015; Barki et al., 2019). Wireless bioelectric and electroceutical dressings have been evaluated to destroy biofilms in vitro, in vivo in porcine models, and Cases with chronic wounds of various aetiologies demonstrating ability to destroy biofilms, faster time to closure and higher wound volume reduction per day compared with standard of care (Kim et al., 2016; Thakral et al., 2013; Whitcomb et al., 2012). Further exploration of energy-based technologies in prospective randomised trials is indicated.

## Other considerations

Limitations of current laboratory testing and definitive clinical signs indicating biofilm presence make it impossible to objectively determine whether biofilms have been eradicated from a wound. Further investigation of point-of-care testing techniques to selectively identify biofilm presence may enhance monitoring techniques in the future; however, currently, monitoring of clinical progression with reduced exudate and slough remains the most effective method to determine response to treatment and biofilm resolution (Leaper et al., 2012). However, despite appropriate treatment, biofilms associated with orthopaedic implants or other foreign devices frequently necessitate removal for resolution (Richardson & Stewart, 2019). In some cases, infection can be controlled temporarily through a combination of systemic and local antimicrobial therapy until fracture or arthrodesis consolidation has occurred (Wu et al., 2014). If cases with both infection and instability, implants may be removed and replaced, or cleaned, sonicated and reimplanted using new orthopaedic screws when financially feasible. Alternatively, internal implants may be removed,

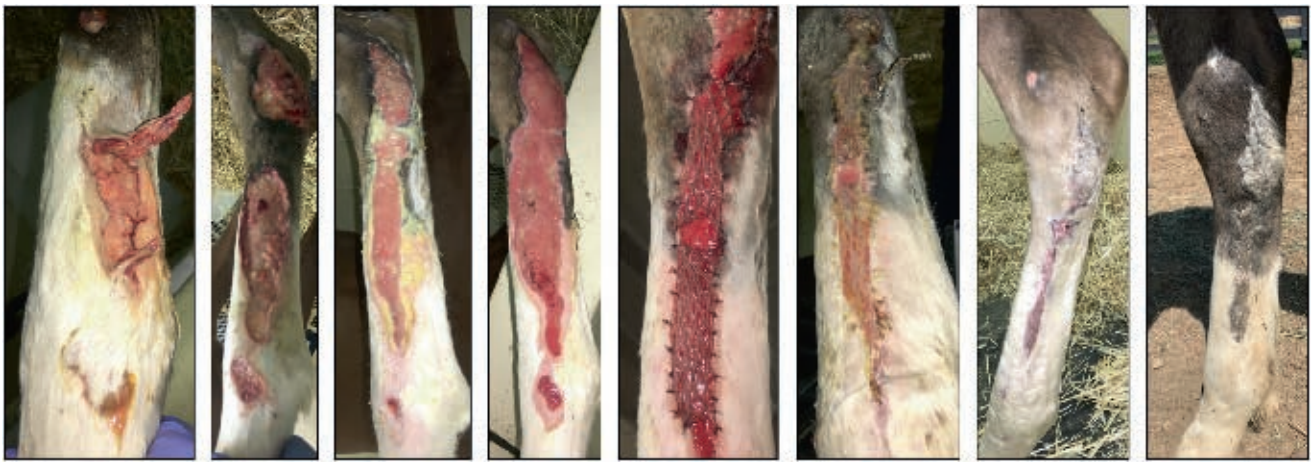

**FIGURE 2** Case example 1. A 7-day-old Warmblood foal was presented for acute swelling of the right tarsus. An approximately 2 cm × 2 cm abrasion was noted over the lateral aspect of the tarsus with moderate periarticular oedema was appreciated. Radiographs of the tarsus revealed no significant abnormal findings and fluid analysis of synovial fluid from the tibiotarsal joint was within normal limits. Ultrasound of the umbilicus, abdomen and thorax revealed no significant abnormal findings. Within 3 days following presentation, the colt developed marked cellulitis of the right hindlimb which was initially with a compression/sweat bandage, and intravenous antibiotic therapy (amikacin, potassium penicillin) and anti-inflammatories (flunixin meglumine). Ultrasound of the limb revealed a suspected extra-articular subcutaneous abscess forming near the level of the distal intertarsal and tarsometatarsal joints. Five days after the initial presentation, strike-through was noted diffusely throughout the bandaged limb, and when the bandage was removed, a generalised necrotic open wound extending from the level of the tarsus to the fetlock was appreciated. Culture and sensitivity yielded *Citrobacter*, *Enterobacter* and *Staphylococcus aureus*. Based on sensitivity results, antibiotics were transitioned to ceftiofur sodium. Repeated daily to every other day topical debridement, lavage and wound care with polyhexamethylene biguanide surfactant-impregnated gauze dressings were performed. Approximately 3 weeks after the initial presentation, the wound bed was deemed healthy enough for skin grafting. Full thickness mesh graft obtained from the ventral abdomen was performed under general anaesthesia. Two weeks later, he received a second mesh graft to cover the remaining area of the wound, obtained from his pectoral region. The tissue here obtained from the ventral abdomen was a different colour but overall, the result was considered a good cosmetic outcome. The foal was dismissed to the care of his owners after approximately 5 weeks hospitalisation and continued to do well at home with approximately 4-year follow-up to date.

and cases managed with a transfixation pin cast or external fixator. The fracture site and surrounding tissues should be debrided and lavaged, and previous screw holes and the surrounding region may be treated locally with antibiotic-eluting materials. In general, when communicating with clients about the cost of care in biofilm-associated wounds, it is recommended to emphasise that the greater expense incurred in the earlier stages of wound management typically reduces duration of therapy and costs overall in treatment long-term (Orsini et al., 2017). Clinical case examples where wounds with biofilms were successfully treated are provided and summarised in Figures 2 and 3.

## FUTURE DIRECTIONS IN DIAGNOSTIC TECHNIQUES AND TREATMENT STRATEGIES FOR BIOFILMS

Novel techniques to prevent or reduce infection burden associated with biofilms are currently being investigated and further developed. Methods described include further investigation of surfactant-based agents, cellular therapeutic options (e.g., platelet rich plasma lysates and mesenchymal stromal cells), quorum-sensing inhibitors (RNAIII inhibiting peptide), hydrophobic polycationic or sol-gel coatings, bacteriophage therapies (antibacterial viruses), antimicrobial peptides, ultraviolet light, low-voltage pulsed electrical fields, acetylsalicylic acid, xylitol, dispersin B, gallium or antimicrobial tethering (Balaban et al., 2005; Barsotti et al., 2013; Bussche et al., 2015; Gilbertie et al., 2020; Gordon et al., 2021; Grassi et al., 2017; Han et al., 2019; Levy et al., 2004; Mohamed et al., 2016; Nablo et al., 2005; Orsini et al., 2017; Schaer et al., 2012; Spaas et al., 2013; Stewart et al., 2012; Tiller et al., 2001; Williams & Hare, 2011). In addition, future diagnostic tests may be more effective at definitively identifying the presence and location of biofilms within a wound bed, with the goal to guide more patient-specific treatment strategies. For example, protease activity correlates to the amount of viable or active biofilm in a wound and a patient-side point-of-care test to quantify wound bed protease activity could be one method to indirectly

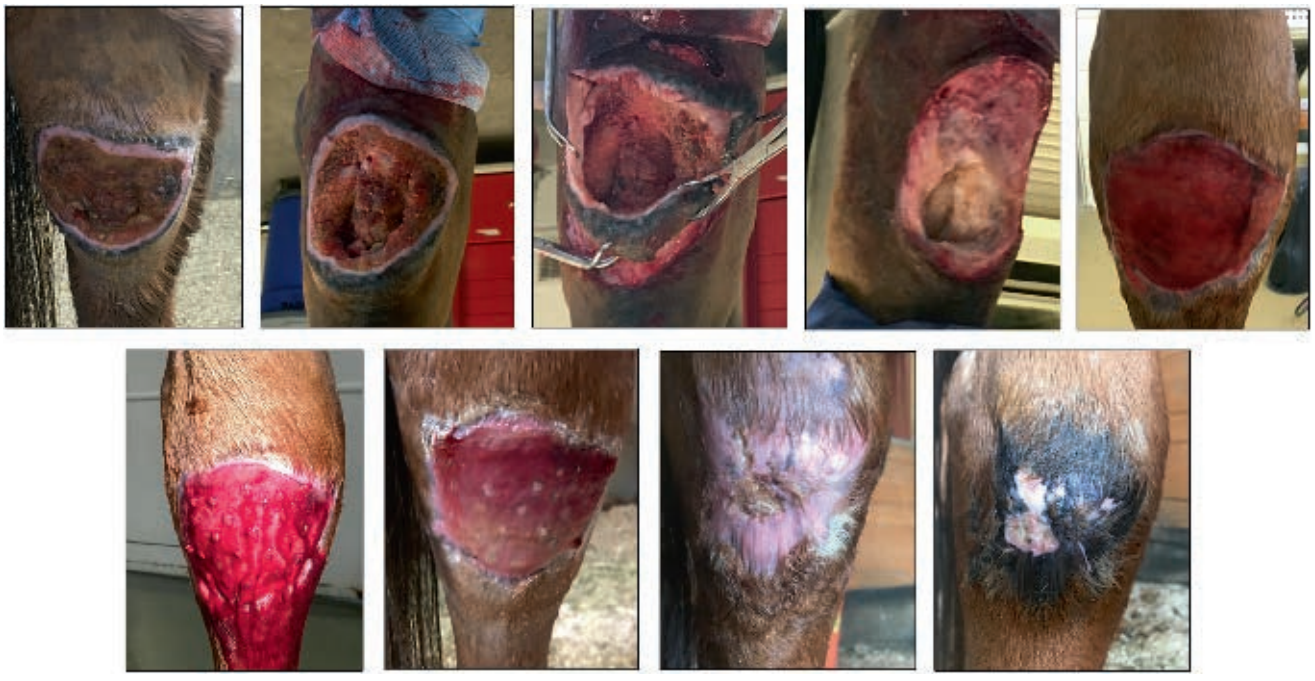

**FIGURE 3** Case example 2. A 14-year-old Quarter Horse gelding was presented for evaluation of a 5-month-old wound on the dorsal aspect of the tarsus, sustained the previous summer on barbed wire fence. He had been initially treated with trimethoprim-sulfamethoxazole antibiotics. Radiographs of the tarsus were performed which revealed no significant abnormal findings and he was turned out on pasture. When the wound continued not to heal, he was brought to Colorado State University Veterinary Teaching Hospital for further evaluation. Radiographs at that time revealed no sequestrum or osseous involvement. The gelding was induced under general anaesthesia and the wound was sharply debrided and lavaged. He was maintained on antibiotics with bandaging in kerlix AMD and splinting to minimise motion through the tibiotarsal joint. A second debridement was performed approximately 3 weeks following the first and samples collected for culture. He was transitioned from trimethoprim-sulfamethoxazole antibiotics to enrofloxacin which was the only appropriate antibiotics based on culture and sensitivity results. Additionally, he was enrolled as a pilot case in a study evaluating the effect of mesenchymal stromal cell therapy to improve wound healing. The lateral half of the wound bed was treated with three doses of intralesional doses of 30 million stem cells. At approximately 3 months following the initial presentation, the wound bed was considered healthy enough to support a graft. A pinch skin graft was performed from skin obtained from the left ventral abdomen. Interestingly, although the wound did fully epithelialise by 10 months following the initial presentation, the lateral aspect of the wound that had been injected with stromal cells did so more rapidly, indicating that the antimicrobial and immunomodulatory properties of MSC may help to accelerate healing and warrants further investigation. AMD, antimicrobial dressing; MSC, mesenchymal stromal cells; ECM, extracellular matrix.

quantify and longitudinally evaluate the amount of residual biofilm in a wound (Leid et al., 2002). Wound blotting techniques using nitrocellulose membrane attached to the wound surface, followed by Alcian blue staining specific to mucopolysaccharides have been used to selectively visualise biofilms in human practice and warrant further investigation towards improved selectively in debridement of wounds, particularly those without apparent necrotic tissue (Minematsu et al., 2013). While prevention of biofilms on medical devices such as central venous catheters (CVC) with topical agents has been described, prevention of biofilm formation with earlier invention in wounds requires further examination. Evaluation of methods to improve detection of biofilms and monitor treatment efficacy of biofilms in case-controlled studies and randomised controlled clinical trials is warranted.

## CONCLUSIONS

The recognition that most nonhealing wounds in equine practice involve pathogenic bacterial biofilms is imperative to successful treatment. Key principles related to BBWC are summarised in Table 2. Clinical indicators that biofilms are present in the wound bed include persistence in a chronic inflammatory state recalcitrant to standard therapies, excessive exudate/moisture, poor quality local granulation tissue, heat, swelling, pain, redness and/or negative bacterial culture results despite clinical suspicion of infection. Biofilm-based wound care strategies emphasise repeated debridement and lavage, combined with topical surfactants or antimicrobials applied within 4 h of debridement. Finally, improved diagnostic tools including noninvasive techniques to detect and quantify biofilms will facilitate research about biofilms in wounds and ease to monitor response to treatment towards improved future outcomes.

**TABLE 2** Key statements on biofilms in wounds (Schultz et al., 2017).

- Biofilms are present in most chronic wounds and are likely to be present on both the surface and deeper wound layers but may not be uniform across or within the wound
- Biofilms are difficult to visualise macroscopically, and exudate, slough or debris may be mistaken for biofilms
- Wounds that contain biofilms may not be identified, leading to ineffective treatment and delayed healing
- Important clinical indications that a wound likely contains a biofilm include a lack of response to treatment with antibiotics or antiseptics
- Debridement is one of the most important treatment strategies against biofilms; however, biofilms reform rapidly, so debridement should be used in conjunction with topical antiseptics and surfactants

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### AUTHOR CONTRIBUTIONS

All authors contributed to the conception and design, acquisition of data, and drafting and final approval of the manuscript.

### CONFLICT OF INTEREST STATEMENT

No conflicts of interest have been declared.

### FUNDING INFORMATION

None.

### ETHICS STATEMENT

Review of current literature; not applicable.

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# HEALTHY Practice MEMBER ASSISTANCE PROGRAM

Services provided by MYgroup McLAUGHLIN YOUNG

The AAEP is excited to announce the expansion of the Healthy Practice Member Assistance Program to allow AAEP members in U.S. and Canada to offer the program's counseling and consultation services to all practice employees at the greatly reduced AAEP program rate. This is an amazing benefit and one that greatly improves your practice culture and your team's well-being.

## Program Pricing & Benefits

The cost for providing Member Assistance Program (MAP) benefits to all practice employees is 22 cents per month per practice employee. We are able to offer the program at this incredibly low rate due to the AAEP's large membership. All current AAEP members in your practice will continue to have their MAP participation covered as a benefit of AAEP membership, and so AAEP members will not be included in the monthly cost for your practice.

Ex: Top Flight Equine Veterinary Services employs 10 individuals, 4 of whom are AAEP members. The cost for Top Flight to provide MAP benefits to the 6 non-AAEP members in the practice will be 6 x .22 for a total cost of $1.32 each month. The yearly cost for coverage for the 6 non-AAEP members will be $15.84.

The monthly rate includes an array of services for employees and immediate family members, such as:

- 24/7 telephonic support
- MAP consultation
- Unlimited number of issues per year
- Face-to-face, telephonic, and virtual counseling
- Extension of services
- Work-life services
- Legal services
- Financial services
- ID theft recovery
- Online seminars
- Account management
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- Promotional materials

## Improve Your Team's Well-Being Today!

**To Learn More or Join the Program, Contact:**

**Gregg Hunter, LPC, M.Ed.**
**Director of Client Development**
**McLaughlin Young Group**
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*The Member Assistance Program is offered to U.S. and Canadian members only at this time due to variances in available providers outside of North America.*

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**How to cite this article:** Marchant, K., Hendrickson, D.A. & Pezzanite, L.M. (2024) Review of the role of biofilms in equine wounds: Clinical indications and treatment strategies. *Equine Veterinary Education*, 36, 152–168. Available from: https://doi.org/10.1111/eve.13919

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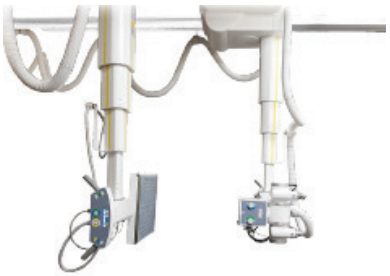

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