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Foreword and Acknowledgements

Equine Veterinary Journal is delighted to publish this special issue containing some of the Clinical Research Abstracts presented at BEVA’s Annual Congress. These abstracts focus on topics of interest and relevance to equine clinicians, and while they often describe work in progress, are of extremely high scientific quality. With this initiative, now in its second year, BEVA hopes to provide both members and equine practitioners in general with up-to-date information that will help inform their clinical practice.

The abstracts have been compiled and edited by David Hicks and Sue Wright and I am grateful to David Mountford, BEVA’s Scientific Programme Guardians and the reviewers of these clinical research abstracts: Keith Chandler, Sandy Love, James Crabtree, Sue Dyson, Luise Harrison, Raphael Labens, Vicki Nicholls and David Rendle.

Celia M. Marr  
EVJ Editor-in-Chief

BEVA Congress Sponsors
DEVELOPMENT AND CLINICAL APPLICATION OF PUDENDAL NERVE BLOCK USING A PERIPHERAL NERVE LOCATOR FOR REPRODUCTIVE SURGERY IN HORSES

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Aims: To develop, describe and evaluate clinical applicability of pudendal nerve block using a peripheral nerve locator.

Methods: In a pilot study, 6 ponies were used to determine the correlation between nerves serving the perineal region (namely pudendal, caudorectal and perineal nerves) and the muscle contractions observed when an electrolocation technique was used. After appropriate electrolocation a small volume of lidocaine/methylene blue solution was injected under general anaesthesia. The dye location relative to the specified nerves was examined after humane euthanasia followed by dissection of the relevant region. In a second experiment, 7 Thoroughbred horses were used to evaluate the appropriate volume of lidocaine solution for the nerve block. Lidocaine/methylene blue solution was injected after positive electrolocation immediately (5–40 min) after euthanasia. A stained segment of 2 cm or more of the nerve was deemed to be sufficient and was evaluated after dissection. Finally, evaluation of a bilateral pudendal nerve block was used to provide anaesthesia and analgesia in 10 clinical cases involving both mares and geldings.

Results: Anal twitch alone was associated with injection of the caudal rectal nerve, whereas combined twitch of the anus and perineum was required for reliable location and injection of the pudendal nerve. Injection was more accurate in standing rather than laterally recumbent horses. A volume of 10–20 ml bilaterally, reliably provided at least 60 min of clinical analgesia for perineal surgery. Procedures satisfactorily completed included: urethral extension and perineal body repair in mares and penile examination, removal of penile tumours and penile amputation in male patients.

Conclusions and practical significance: Pudendal nerve block is a practical and effective alternative to epidural analgesia for selected standing surgical procedures of the reproductive tract in male and female horses.

Ethical animal research: The Cornell University Institutional Animal Care and Use Committee approved the project and informed client consent was obtained. Sources of funding: None. Competing interests: None.

A PROSPECTIVE RANDOMISED STUDY TO COMPARE THE EFFECTS OF PREOPERATIVE HYPERTONIC SALINE OR PENTASTARCH ON HAEMATOLOGICAL VARIABLES AND LONG-TERM SURVIVAL OF SURGICAL COLIC CASES

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Aims: To compare the effects of hypertonic saline and pentastarch on packed cell volume (PCV) and total protein (TP) immediately preoperatively and on long-term survival of horses undergoing emergency exploratory laparotomy.

Methods: One hundred horses presenting to the Philip Leverhulme Equine Hospital between 2004 and 2008 with signs of abdominal pain and PCV ≥45% were recruited. Horses were randomly allocated to receive 4 ml/kg bwt of either hypertonic saline (HS; Vetivex, 7.2%; n = 49) or pentastarch (PS; Haes-Steril, 10%; n = 51) preanaesthesia. Blood samples were collected at presentation and post fluid resuscitation. Horse survival was tracked for 4 years following the final recruited case.

Results: There were no significant differences between treatment groups at presentation for: age (HS, mean 13 ± 6 years; PS, mean 14 ± 6 years); body mass (HS, median 516 [130–698] kg; PS, median 525 [312–660] kg); sex (HS, 1 stallion, 34 geldings, 16 mares; PS, 2 stallions, 32 geldings, 15 mares); baseline PCV (HS, median 52 [46–70]%; PS, median 50 [45–70]%); baseline TP (HS, median 72 [46–103] g/l; PS, median 72 [50–106] g/l); and heart rate (HS, mean 80 ± 20 beats/min; PS, mean 77 ± 18 beats/min) at presentation. Hypertonic saline treatment resulted in a significantly
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greater reduction in both PCV (HS, mean 13 ± 7%; PS mean 7 ± 5%; P<0.001) and TP (HS, median 16 (6–32) g/l; PS, median 2 (8 to 17) g/l; P<0.001), compared with pentastarch. There was no significant difference in long-term survival between treatments (Cox proportional hazards model, P = 0.73).

Conclusions and practical significance: Despite the greater reduction in haemoconcentration and likely reduction in blood viscosity incurred by HS, no benefit to overall long-term survival was demonstrated. In a clinical setting, either of these fluids remains appropriate for preoperative fluid resuscitation in colic horses.

Acknowledgements: All the veterinary anaesthetists who assisted in data-gathering.

Ethical animal research: Permission for this study was granted by the Ethical Review Committee of the University of Liverpool’s Veterinary School. Signed owner consent was obtained for all cases. Sources of funding: None. Competing interests: None.

04

ABNORMAL PLASMA NEUROACTIVE PROGESTAGEN DERIVATIVES IN ILL, NEONATAL FOALS PRESENTED TO THE NEONATAL INTENSIVE CARE UNIT

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Aims: To determine the pregnane profile of foals with neonatal maladjustment syndrome (NMS) and compare it with that of healthy controls and sick, non-NMS foals.

Methods: Thirty-two foals with a clinical diagnosis of NMS, 12 foals with other neonatal disorders and 10 healthy control foals were selected for the study. Heparinised blood samples were collected from each group of foals and pregnane and androgen concentrations were determined using liquid chromatography mass spectrometry at 0, 24 and 48 h of age.

Results: Healthy foals showed a significant decrease in pregnane concentrations over the first 48 h of life (P<0.01). Foals with NMS and sick, non-NMS foals had significantly increased progesterone, pregnenolone, androstenedione, dehydroepiandrosterone and epitestosterone concentrations compared with healthy foals (P<0.05). Progesterone and pregnenolone concentrations of sick, non-NMS foals decreased significantly over 48 h (P<0.05), whereas concentrations in NMS foals remained elevated.

Conclusions and practical relevance: Pregnanes concentrations of ill, neonatal foals remain elevated following birth, reflective of a delayed, or interrupted, transition from intra- to extra-uterine life. These pregnanes are potent allosteric modulators of the GABAA receptor and are important in providing tonic inhibition of fetal central nervous system activity and damping movement to prevent maternal damage. Infusion of the pregnane allopregnanolone into neonatal foals leads to somnolence and loss of affinity for the dam (Madigan et al. 2012). Together, these findings suggest that the pathogenesis of NMS may be associated with the persistence of high concentrations of pregnanes. Serial progesterone and pregnenolone measurement may be useful in aiding diagnosis of NMS.

Ethical animal research: The UC Davis IACUC approved the project. Sources of funding: Private donation. Competing interests: None.

Reference


05

TO DETERMINE THE EFFECT OF AN ORAL JOINT SUPPLEMENT ON ORTHOPAEDIC, PHYSIOTHERAPY AND HANDLER EVALUATION SCORES IN HORSES

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Introduction: Despite the range of oral joint supplements available, there has been very limited research into their efficacy.

Aims: To determine effect of an oral joint supplement on orthopaedic, physiotherapy and handler evaluation in horses.

Methods: Twenty-four mature horses were included in the study. Horses were excluded if they were in poor body condition, had health problems or greater than 2.5 lameness. Supplement S (containing chondroitin sulphate 1.62 g/100 kg bwt, glucosamine 1.9 g/100 kg bwt, vitamin C 0.8 g/100 kg bwt, methyl sulphonil methane 2.56 g/100 kg bwt, DHA 0.66 g/100 kg bwt), EPA 0.34 g/100 kg bwt or placebo P (carrier/flavours only) were given to horses in their feed for 21 days each in a triple-blind crossover design; all horses received supplement and placebo in random order. Horses were evaluated at Days 0 (baseline), 21 (after first treatment) and 42 (after second treatment). Assessments included: clinical orthopaedica evaluation for straight line and lunging exercise (walk and trot), and during ridden exercise (walk, trot and canter); handler field evaluation, during groundwork and while ridden, grading-specific criteria; grading of range of motion (ROM) and muscle tone based on standardised physiotherapy criteria. All evaluators were blinded to treatment. Significance indicates P<0.05.

Results: S was associated with significantly lower lameness grade in a straight line and circle than either P or baseline. Both S and P were associated with significantly improved ROM and muscle tone over baseline. Handler scores for ridden and groundwork were significantly higher with S compared with P or baseline. After S, horses were graded significantly higher for field ‘ease-of-movement’ compared with P or baseline.

Conclusions and practical significance: Oral administration of this supplement was associated with less lameness, improved ridden/groundwork scores and improved ‘ease-of-movement’ in the field. Improvement in physiotherapy assessment with both treatments over time suggests effects of ongoing training on ROM and muscle tone.

Ethical animal research: This study involved informed consent of the persons responsible for horses used in the study. Sources of funding: World Horse Welfare. Competing interests: David Marlin and Rebecca Frost are employed by Science Supplements. Vicki Adams and Rachel Murray are involved with Science Supplements on a consultancy basis.

Manufacturer’s address

*FlexAbility™, Freedom Flex™, Science Supplements.
EFFECT OF TWO DIETS ON ANTIOXIDANT STATUS IN RACING STEEPLECHASERS DURING INTENSIVE TRAINING

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Aims: High-intensity training, racing and inappropriate antioxidant supply generates high levels of deleterious oxidative stress. This study aimed at comparing the effect of 2 commercial feeds on oxidative stress levels in steeplechasers, over a 3-month period.

Methods: A random double-blind study was undertaken to compare the effect of 2 diets in 40 racehorses. The first group received a regular pelleted commercial diet (R) and the second group received a low-starch high-fibre diet (L). The horses were examined after a 6-week habituation period (T0) and after 6 (T6) and 12 weeks of reinforced training (T12). Horses were raced regularly. At each step, horses were weighed; body and clinical scores were attributed. Several blood markers were studied including vitamin A (Vit A), vitamin E (Vit E), beta-carotene, superoxide dismutase (SOD), glutathione peroxidase (GPx), Coenzyme Q10 (CoQ10), copper (Cu), zinc (Zn), selenium (Se) and pre- and post effort creatine kinase (CK) activity. Data were analysed using an ANOVA for repeated measurements and a t test.

Results: In the L-group, GPx levels were significantly higher than in the R-group at all times and a progressive and constant increase in GPx was observed from T0 to T12. Vit E levels and CoQ10 increased at T6 in both groups but levels were significantly higher in group-L. Selenium values were significantly higher at T0 in the L-group vs. R-group and remained stable in time in both groups. All other markers were not significantly different between groups and did not change with time. The CK levels did not differ between groups, however 2 horses from group-R displayed different between groups and did not change with time. The CK levels did not differ between groups, however 2 horses from group-R displayed different between groups and did not change with time.

Conclusions and practical significance: The choice of an appropriate diet can effectively increase antioxidant protection and prevent training-induced oxidative stress even in intensively trained racehorses during racing season.

Acknowledgements: The authors thank Mr E. Clayeux for his collaboration.

Ethical animal research: The trainer of all horses gave informed consent for this study. Sources of funding: Lambey SA, France. Competing interests: J.-L. Lambe is owner of Lambey SA and S. Benoit is a consultant for this feed company.

THE EFFECT OF INJECTION NEEDLE GAUGE SIZE ON THE VIABILITY OF EQUINE MESENCHYMAL STEM CELLS

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Aims: Performance horses are at high risk of injury to the superficial digital flexor tendon (SDFT). Studies have shown that autologous mesenchymal stem cells (MSCs) injected into SDFT lesions subsequently result in a significant reduction of re-injury in National Hunt racehorses; however, recent studies show that only 25% of implanted MSCs survive 24 h post injection. The reason for this loss is unclear but may relate to cell quiescence or injection-related mortality. We hypothesised that cell viability and mortality is increased with needle gauge.

Methods: Equine MSCs cultured in vitro were resuspended to a final suspension density of 5 x 10^5 cells/ml to mimic that used for implantation in clinic. The cell suspension was injected through a 19 gauge, 21 gauge (current practice in clinic) or 23 gauge needle. Mesenchymal stem cells viability and mortality was analysed over a 24 h period post injection using alamarBlue® and Annexin V (apoptosis) assays, respectively.

Results: There was a 25% reduction in viability (P<0.01) and mortality (P<0.01) compared with noninjected MSCs over the 24 h period post injection. All needle gauges also induced a decrease in cell metabolic activity immediately post injection but with recovery by 2 h post injection. Furthermore, 21 gauge and 23 gauge needles increased early apoptotic cells immediately post injection, whereas the 19 gauge needle showed a delayed increase in apoptosis until 2 h post injection.

Conclusions: The delayed apoptosis may correlate with a subpopulation of quiescent cells subsequently becoming apoptotic. The proportion of early and late apoptotic MSCs, while significant, does not account for the total cell loss reported after intra-lesional injection.

CAN WE USE INFORMATION ON THE MECHANICAL PROPERTIES OF WAXED SAND/FIBRE, SAND/FIBRE AND SAND/RUBBER ARENA SURFACES TO HELP UNDERSTAND INJURY PREVENTION?

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Introduction: Recent research demonstrated that there is a link between arena surfaces and lameness in the dressage horse, that horses alter their gait on different surfaces and that cross-training on different surfaces was protective (Murray et al. 2010a,b; Walker et al. 2012). However, there are minimal data on arena mechanical properties and how these could relate to injury.

Aims: To compare the mechanical properties of 3 common arena surface types.

Methods: A dual-axis synthetic-hoof drop hammer fitted with accelerometers and a 3-axis load cell was used to test 49 arenas: waxed sand with fibre (WSF) (n = 16), un-waxed sand with fibre (SF) (n = 19) and un-waxed sand with rubber (SR) (n = 14). Ten different locations were tested on each arena. Maximum load and load rate (representing firmness), maximum vertical and horizontal accelerations (representing friction), shear angle, and hysteresis (representing elasticity), were compared between the 3 surfaces using an independent samples Student’s t test.

Results: There were significant differences in firmness, friction and elasticity between all 3 surfaces, with WSF having the greatest values for all properties. When looking at the un-waxed surfaces, adding fibre resulted in increased friction, decreased firmness and decreased elasticity compared with adding rubber (Table 1).
Conclusions and practical significance: The mechanical properties of the surface types are significantly different. These findings suggest that a horse’s limbs may experience more rapid deceleration and higher impact on WSF, while on SF or SR a horse has more capacity to slide through the surface with less impact but experiences less energy return. This has implications in potential development of injury on different surfaces, and reinforces the requirement for proprioceptive training on varying surfaces prior to undertaking peak loading on a surface the horse has not trained on.

Ethical animal research: Not applicable. Sources of funding: Funding from World Horse Welfare, the Swedish-Norwegian Foundation for Equine Research and UK Sport lottery funding for the BEF World Class Programme. Competing interests: None.

Table 1: Summary of the mean and standard deviation for maximum load and load rate, maximum vertical and horizontal accelerations, shear angle and hysteresis for waxed sand/fibre, sand/fibre and sand/rubber arena surfaces

<table>
<thead>
<tr>
<th>Property</th>
<th>Waxed sand with fibre</th>
<th>Sand with fibre</th>
<th>Sand with rubber</th>
</tr>
</thead>
<tbody>
<tr>
<td>Max load (kN)</td>
<td>13.35 ± 2.89</td>
<td>6.83 ± 4.09</td>
<td>9.88 ± 3.04</td>
</tr>
<tr>
<td>Max load rate (kN/sec)</td>
<td>406.75 ± 34.41</td>
<td>297.55 ± 110.18</td>
<td>382 ± 45.86</td>
</tr>
<tr>
<td>Max vertical acceleration (m/sec²)</td>
<td>71.37 ± 27.48</td>
<td>37.54 ± 12.33</td>
<td>34.03 ± 15.92</td>
</tr>
<tr>
<td>Max horizontal acceleration (m/sec²)</td>
<td>11.29 ± 10.44</td>
<td>5.67 ± 4.95</td>
<td>4.22 ± 3.95</td>
</tr>
<tr>
<td>Shear angle (radians)</td>
<td>0.285 ± 0.08</td>
<td>0.320 ± 0.11</td>
<td>0.320 ± 0.11</td>
</tr>
<tr>
<td>Hysteresis</td>
<td>0.135 ± 0.042</td>
<td>0.056 ± 0.036</td>
<td>0.085 ± 0.066</td>
</tr>
</tbody>
</table>

References


REFERENCE

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Aims: Synovial inflammation is characterised by increases in catabolic cytokines, notably tumour necrosis factor-α (TNF-α) and interleukin-1β (IL-1β), to understand their role in equine arthritis, model systems to evaluate small interfering RNAs (siRNAs) targeted against these cytokines are being developed. Specifically, we aim to investigate the change in TNF-α and IL-1β expression following transfection of equine synoviocytes with TNF-α siRNA and IL-1β siRNA. It is hypothesised that gene expression would decrease following treatment with siRNAs, and that off-target effects would not be induced.

Methods: Synoviocytes from healthy metacarpophalangeal joints of 6 horses were digested using 0.2% trypsin and 0.2% collagenase type-II. Synoviocytes were cultured in DMEM with 10% FCS, with and without 1 μg/ml LPS, and with and without one of 3 siRNAs at 20 nmol: generic scrambled control siRNA, TNF-α siRNA or IL-1β siRNA. Gene expression was analysed with RT-qPCR for TNF-α and IL-1β; GAPDH was used for normalisation.

Results: The patterns of TNF-α gene expression in 2 experiments were similar; expression levels decreased (51%) relative to LPS stimulated control siRNA. Efficiency of transfection with the IL-1β siRNA was less reliable: expression levels of IL-1β actually increased when using IL-1β siRNA.

Conclusions: The pattern of TNF-α gene expression supported our hypothesis but the degree of gene knockdown was disappointing. This may be due to a number of reasons; LPS concentration, siRNA transfection method, or siRNA sequence. The IL-1β siRNA did not work in these experiments and indeed may be inducing ‘off-target’ effects. The mixed culture of synoviocytes used was predominantly synovial fibroblasts.

Further work will be carried out using peripheral blood mononuclear cells, and multiple siRNA sequences.
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**THE EFFECT OF DISPLACED VERSUS NONDISPLACED PELVIC FRACTURES ON LONG-TERM RACING PERFORMANCE IN 31 THOROUGHBRED RACEHORSES**

Aims: To evaluate the long-term racing prognosis for Thoroughbred racehorses with displaced vs. nondisplaced fractures of the pelvis, identified scintigraphically.

Methods: Medical records of 31 Thoroughbred racehorses presenting to the University of Melbourne Equine Centre, that had fractures of the pelvis identified scintigraphically were reviewed. Pelvic fracture site was determined and defined as displaced or nondisplaced based on ultrasound and/or radiographic findings. Race records were analysed for each horse, with a minimum of 24 months follow-up, and correlated to fracture type to determine long-term prognosis for racing. Results are expressed as median and range.

Results: Fractures at a single site were more common (n = 22) than fractures involving 2 sites (n = 9) and the ilial wing was the most commonly affected (n = 12). Thoroughbred racehorses with displaced pelvic fractures at any site (n = 12) raced fewer times within 24 months of diagnosis than horses with nondisplaced fractures (n = 19) (0.5, 0–13, vs. 7, 0–24, P = 0.037) but there was no clear statistical difference in race earnings between the 2 groups (0, 0–123,250, vs. 14,440, 0–325,500, P = 0.080). Four horses with displaced fractures (33%) were subjected to euthanasia on humane grounds due to persistent severe pain. When these were excluded from the analysis there were no differences in performance variables between horses with displaced and nondisplaced fractures.

Conclusions and practical significance: Thoroughbred racehorses with displaced and nondisplaced pelvic fractures that survive the initial post injury period have a good prognosis for racing.

Ethical animal research: All owners consented to use of horse details within this retrospective analysis. Sources of funding: University of Melbourne Equine Centre. Conflicts of interest: None.

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**Abstracts**

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**RISKS OF SYNOVIAL SEPSIS FOLLOWING INTRASYNOVIAL MEDICATION IN AMBULATORY PRACTICE, 2006–2011: 9456 INTRASYNOVIAL INJECTIONS**

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Aims: To determine the incidence of synovial sepsis following intra-articular and intrathecal injections. Synovial sepsis is a serious potential complication of intrasynovial medication.

Methods: Available case records for all horses receiving intrasynovial medications (ISMs) performed by 9 ambulatory clinicians were examined over 5 years (2006–2011). Intrasyovial medications were defined as therapeutic interventions, based on clinical history and orthopaedic examination; nontherapeutic injections for diagnostic analgesia were excluded. Route protocol was not to clip injection sites; the skin was prepared using standard aseptic technique. Scrupulous aseptic injection technique was employed at all times. All horses were under the care of a single practice, dealing with mainly Thoroughbred racehorses. Records were cross-referenced against synovial cytology submissions and hospital admissions for synovial sepsis. Development of 2 out of 4 clinicopathological signs of synovial sepsis (lameness, joint distension, synovial white blood cell count >10,000/l, synovial total protein >25 g/l) within 8 weeks of medication of the same synovial space was considered to represent a post medication complication (PMC). Failure of the PMC to resolve with conservative therapy was considered to represent post medication synovial sepsis (PMSS).

Results: During the study period 9456 ISMs were recorded, in 4332 sessions, in 1732 horses. Corticosteroids were included in 92.3% of ISMs, 94.8% included amikacin and 0.15% (14/9456) included polysulphated glycosaminoglycans (PSGAGs). Twelve horses developed PMC (0.0013% ISM), 4 horses developed PMSS (0.0004% ISM). All 4 horses returned to use following joint lavage. Administration of intrasyovial PSGAGs, was significantly associated with PMSS (P<0.0001 OR = 787 95% CI 145–20,337). Intrasyovial medications that included amikacin were less likely to develop PMSS (P = 0.005 OR = 0.018 95% CI 0.0019–0.174); however if the PSGAGs group was excluded the difference became nonsignificant (P = 0.0981).

Conclusions and practical significance: The risk of iatrogenic sepsis following intrasynovial medication is extremely low. Intrasyovial medication with PSGAGs should be avoided without concomitant antimicrobials.

Acknowledgements: A. Wilson and K. Batteate for case collation.

Ethical animal research: Not required by this Congress: retrospective analysis of case records. Sources of funding: None. Competing interests: None.
Methods: A new technique is described and records were retrieved for 25 cases which had undergone the procedure at Newmarket Equine Hospital between 2009 and 2011. Case records were evaluated for anamnesis, clinical findings, and details of surgical and post operative management. Outcome of surgery was assessed by telephone questionnaire with owners, relating to both functional and cosmetic outcome of surgery.

Results: A total of 78.9% of cases treated using the described technique had resolution of clinical signs and returned to full work, and a further 18.2% were improved. Outcome was unrelated to number of SPs resected or mean radiographic grade of impingement. Cosmetic outcome was described as excellent in 81.8% and good in the remainder. Surgery time ranged from 20 to 70 (median 30) min and there were no intraoperative or post operative complications.

Conclusions: Previous descriptions of overriding SPs have failed to consider the shape of the SPs, and common site of impingement. As a result, previously reported techniques have been exceedingly destructive. Additionally, the changing relationships of SPs between standing and recumbent positions complicates surgical procedures when the previously reported technique is employed.

Practical significance: Based on the above and the presented follow-up data, subtotal (cranial wedge) ostectomy represents a logical and effective treatment for overriding SPs. If performed under general anaesthesia it results in short surgery time, maintains the contour of the spine, has few complications, and produces a functionally and cosmetically better outcome than previously reported nondefined amputation of the dorsal portion of implicated SPs.

Ethical animal research: Not required by this Congress: retrospective clinical study. Sources of funding: None. Competing interests: None.

TEARS OF THE ACCESSORY LIGAMENT OF THE SUPERFICIAL DIGITAL FLEXOR AND THEIR RELATIONSHIP TO THE CARPAL SHEATH

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Aims: The study aimed to describe an ultrasonographic technique for evaluation of the accessory ligament of the superficial digital flexor (ALSDF) and to report the presentation, clinical, ultrasonographic and endoscopic features associated with intrathecal tears of the ligament.

Methods: The case records of 83 horses that underwent ultrasonography features associated with intrathecal tears of the ligament. The areas of torn ALSDF were in consistent locations and with colour Doppler to assess intra- and periligamentous vasculature. The curved array transducer was used for transverse studies orientations and with colour Doppler to assess intra- and periligamentous vasculature. The curved array transducer was used for transverse studies both with the limb weightbearing and with slight carpal flexion. This combination of imaging studies detected tearing of the ALSDF which communicated with the carpal sheath in all 10 cases. Seven of the 10 cases underwent tenoscopic examination which confirmed the ultrasonographic findings. The areas of torn ALSDF were in consistent locations and with similar lesion morphology.

Conclusions: Disruption of the ALSDF can communicate with the carpal sheath resulting in lameness and intrathecal haemorrhage. The latter appears to result from vessels which are primary branches of the median artery and tears in the ALSDF can extend to this. The injuries are reliably predicted by ultrasonography. This also allows assessment of the proximity of the tear to the median artery which in turn is an important guide for subsequent tenoscopic surgery.

Practical significance: Ultrasonographic evaluation of the ALSDF can identify tears which communicate with the carpal sheath. Tenoscopy should then be considered as a treatment option.

Ethical animal research: Not required by this Congress: retrospective clinical study. Sources of funding: None. Competing interests: None.

IN VIVO EVALUATION OF ACOUSTOELASTOGRAPHY IN THE NORMAL EQUINE SUPERFICIAL DIGITAL FLEXOR TENDON

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Introduction: Superficial digital flexor tendon (SDFT) injury is common in athletic horses. Recovery requires balancing rest and controlled exercise, resulting in high risk of re-injury. It is difficult to determine the level of work the damaged tissue can sustain based on serial ultrasound examination alone. However, as small changes in sonographic appearance are associated with large changes in biomechanical strength, it is difficult to determine the degree of activity the tissue can sustain.

Aims: Acoustoelastography (AEG) is a new ultrasound-based model to evaluate tendon function. Acoustoelastography deduces stiffness gradient, the rate of change of normalised stiffness as a function of strain, by analysing the changes in echo intensity observed in cine loops captured from gradually deforming tendon. The goal of this project was to establish a reproducible method for applying AEG in the normal equine SDFT.

Methods: Fifteen horses with no history of lameness and normal lameness examinations were recruited with informed owner consent and in accordance with the University Research Animal Resources Center. Stiffness gradient index (SGI) and dispersion values (DV) for the palmar SDFT were evaluated at 3 sites (5, 10 and 15 cm distal to the accessory carpal bone [DACB]) by 2 observers. Lifting of the contralateral forelimb during image acquisition resulted in the required SDFT deformation. Interobserver repeatability, intraobserver repeatability, and right-to-left limb symmetry were evaluated.

Results: The SGI and DVs for the SDFT at different locations, as well as effects of age or gender, showed no statistical difference (P > 0.05). Interclass correlation evaluating repeatability within the same observer, between observers, and symmetry between right and left limbs demonstrated excellent agreement.

Conclusions and practical significance: This study shows that AEG is a feasible and repeatable technique for measuring stiffness gradients in normal equine SDFTs. This study will provide the basis for developing a simple, noninvasive evaluation of tendon function that could vastly improve the ability to detect, monitor and treat tendon injuries.

Ethical animal research: All animal protocols were approved by the University of Wisconsin Research Animal Resources Center. Client-owned horses were used for the study and informed consent was obtained.

Sources of funding: Research supported by Companion Animal Grant through University of Wisconsin. Competing interests: Co-author collaborator Ray Vanderby has patent on acoustoelastography post processing ultrasound-based tissue evaluation technique.
**Abstracts**

**HORSE, RIDER AND VENUE-RELATED RISK FACTORS FOR ELIMINATIONS FROM FÉDÉRATION EQUESTRE INTERNATIONALE ENDURANCE RIDES DUE TO LAMENESS AND METABOLIC REASONS**

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**Aims:** To assess risk factors for elimination due to lameness and metabolic reasons from Fédération Equestre Internationale (FEI) endurance rides of 80–160 km distance.

**Methods:** Venue, horse and rider-related variables (n = 33, including data on speed, signalment, previous experience) were collected from the FEI website. Weather and terrain data were collected at the venue. Univariable and multivariable logistic regression and generalised estimated equation (GEE) statistics were performed to assess risk factors for eliminations due to lameness and metabolic reasons, respectively.

**Results:** Multivariable logistic regression on 1435 horse starts revealed that the venue (n = 11) (P = 0.013), the horse’s previous experience >90 days) elapsed since the last FEI ride (OR = 0.78, P = 0.044) and time were significantly associated with elimination for lameness; all 3 predictors remained significant in the GEE model. In the multivariable model for elimination for metabolic reasons, the venue (P = 0.011), increasing number of entries (OR = 1.008, P = 0.001) and deep sand or soil on the track (OR = 1.98, P = 0.001) significantly increased the risk of elimination for metabolic reasons.

**Conclusions and practical significance:** Decreasing the frequency of racing schedule may contribute to decreased risk of elimination for lameness. Competing in deep sand or soil may contribute to exhaustion leading to elimination for metabolic reasons. Venue was associated with both outcomes; a number of reasons other than terrain and going are likely to contribute to this; e.g. unmeasured horse-level factors (training, previous injuries etc.) and the riders' aim (e.g. training, qualification, competition). Elimination due to lameness or metabolic reason is likely to be the end result of a complex process, of which not every aspect was or can be measured. However, further studies with a larger number of horse starts and assessing variables that could not be measured in this study may identify risk factors that can be modified.

**Ethical animal research:** Not required by this Congress: retrospective analysis of data in public domain. **Sources of funding:** A. Nagy’s PhD scholarship was funded by the University of Bristol. **Competing interests:** None.

**IS THE MOVEMENT OF THE THORACOLUMBAR AND LUMBOSacRAL JOINTS IN THE RIDDEN DRESSAGE HORSE AFFECTED BY MUSCLE DEVELOPMENT?**

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**Introduction:** Stabilisation of the back is essential for rider support and to moderate intervertebral movement. Epidemiological data has shown that 25% of UK dressage horses had back pain over a 2-year period (Murray et al. 2010). It has been established that back pain in sports horses is performance limiting (Stubbs et al. 2011). Intervertebral range of motion (ROM) and flexion–extension movements of the back have been previously described. However, the relationship between muscle development and ridden ROM has not been investigated.

**Aims:** To investigate the relationship between grade for development of muscles visible on examination and ridden ROM of the thoracolumbar (TL) and lumbosacral (LS) regions of the back.

**Methods:** Thirty-five dressage horses (novice to Grand Prix competition level) were evaluated by an experienced clinician and assigned muscle development grades. High-speed motion-capture (250Hz) was used to measure TL and LS angle for 4 strides of collected trot from the left side. Spearman’s rank correlation tests were used to test for associations between muscle development grade and ridden angles and ROM.

**Results:** Increased development of visible muscle groups was correlated with increased TL and LS flexion at different parts of the stride (Table 1). Decreased LS ROM was associated with increase muscle development of the thoracic and lumbar regions.

**Conclusions and practical significance:** Results suggest that increased development of muscle groups visible on examination is important for...
The Effect of Trotting Speed, Direction and Line of Travel on Asymmetry in Standardbred Racehorses During High-Speed Locomotion on the Racetrack

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Aims: To evaluate the effects of racing and training conditions on motion symmetry we set out to quantify hindlimb symmetry using an objective inertial sensor-based system in a cohort of in-training Standardbred racehorses during high-speed trotting and to assess the influence of speed, line taken (straight vs. bend) and direction of travel (clockwise vs. anticlockwise).

Methods: Eight Standardbred racehorses in full training were instrumented with a GPS-enhanced inertial sensor located at the tuber sacrale and trotted around an oval racetrack at a range of speeds and in both directions. Symmetry index (SI), MinDiff, MaxDiff, vector sum (VS) and range of motion (ROM) were quantified for the hindlimbs for each stride using vertical displacement data derived from the inertial output.

Results: A total of 9108 strides were collected on the track. Overall, all horses displayed a right hindlimb asymmetry or ‘lame ness’. Horses were more asymmetrical when trotting around bends compared with on straights. The main factor influencing asymmetry was trotting around a clockwise bend. Speed had a small but significant influence on asymmetry that varied between measures.

Conclusions: The inertial sensor system was suitable for collecting on-track, high-speed locomotion data. The magnitude of asymmetry was dependent on direction, possibly indicating subclinical lameness or laterality in these horses.

Practical significance: The inertial sensor system is practical for on-track objective high-speed lameness evaluation. Further studies with larger numbers of objectively sound and lame Standardbred horses are needed to determine the effect of lameness on high-speed trotting. The influence of factors such as training regimes and racing equipment should also be evaluated.

Ethical animal research: All procedures were performed with approval of the Royal Veterinary College Ethics Committee. The study was carried out in accordance with local regulations and in collaboration with the Swiss National Stud and Vetsuisse Bern. Informed consent was obtained from the owners of all horses used. Approval no. 2012/P337. Sources of funding: None. Competing interests: None.

Table 1: Table summarising association of muscle development grade with back angles at specific points of the stride within the hindlimb (HL) and forelimb (FL)

<table>
<thead>
<tr>
<th>Angle</th>
<th>Point of stride</th>
<th>Muscle group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thoracolumbar</td>
<td>HL stance</td>
<td>Lumbosacral, pelvic</td>
</tr>
<tr>
<td>(TL)</td>
<td>HL maximum protraction</td>
<td>Neck, lumbar, lumbosacral, pelvic</td>
</tr>
<tr>
<td>Lumbosacral</td>
<td>FL stance</td>
<td>Neck, lumbar, lumbosacral, pelvic</td>
</tr>
<tr>
<td>(LS)</td>
<td>HL stance</td>
<td>Abdominal, thoracic, lumbar, lumbosacral, pelvic</td>
</tr>
<tr>
<td></td>
<td>HL maximum protraction</td>
<td>Abdominal</td>
</tr>
<tr>
<td></td>
<td>FL stance</td>
<td>Abdominal, lumbosacral, hindlimb</td>
</tr>
</tbody>
</table>

References

Objective Assessment of Gait Asymmetry in Polo Ponies

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Aims: Polo ponies have demanding athletic careers and are likely to display a degree of movement asymmetry due to subclinical lameness. Due to training and management practices in polo, the prevalence of lameness may be underestimated. Subjective assessment of lameness has been proven to be unreliable and a growing body of evidence supports the use of trunk mounted inertial sensors as a practical field method of objective gait analysis. We set out to test the hypothesis that all polo ponies display some degree of movement asymmetry. It is likely that some of these asymmetries will be severe enough to be classified as lameness. In horses with asymmetry we hypothesise that left forelimb lameness will predominate.

Methods: Forty polo ponies were equipped with trunk mounted inertial sensors and trotted along a hard, straight surface. Data were analysed according to published protocols and standard objective symmetry indices were derived and compared with published thresholds.

Results: All 40 horses displayed asymmetrical movement. Twenty-one (52.3%) were left forelimb asymmetric, and 19 (47.5%) were right forelimb asymmetric. Eighteen of these asymmetric horses had asymmetry values consistent with lameness. Twelve horses were identified as left forelimb lame and 6 horses identified as right forelimb lame. There was an equal division between right and left hindlimb asymmetry. Nine horses had values exceeding the lameness thresholds; 6 were right hind lame and 4 were left hind lame.

Conclusions: Polo ponies display asymmetry and some asymmetries are consistent with lameness. There appears to be a bias in left forelimb lameness in this population.

Practical significance: These findings may highlight underlying orthopaedic pathology that may be discipline specific or be a reflection of entrained or inherent laterality/handedness.

Ethical animal research: All procedures were performed with approval of the Royal Veterinary College Ethics Committee. Informed consent was obtained from all owners of the horses used in this study. Approval ref no. 2012/P337. Sources of funding: None. Competing interests: None.
MUSCULOSKELETAL INJURY IN ARABIAN RACEHORSES: A STUDY OF INJURY DISTRIBUTION AND PREVALENCE IN ONE TRAINING YARD IN THE UNITED KINGDOM (2005–2012)

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Aims: There has been limited information regarding musculoskeletal injury in Arabian racehorses. This study aims to document the distribution of musculoskeletal injuries and their prevalence in a population of Arabians in flat training in one Newmarket yard.

Methods: A retrospective analysis of individual veterinary records for all horses in training in one Arabian racing yard during the period from 1 January 2005 to 31 December 2012 was performed. Information including injury type, sex, age and limb affected were recorded. Injury categories included in the study were: 1) fractures involving the carpus, tarsus, proximal sesamoid bones and metacarpophalangeal/metatarsophalangeal joints; 2) stress fractures (pelvis, tibia, scapula, humerus, radius, metacarpus, metatarsus); 3) suspensory ligament branch desmitis; 4) superficial digital flexor (SDFT) tendinitis. Injuries which occurred as a result of trauma and those which may be associated with variable clinical signs were excluded.

Results: In 144 horses, 42 individual injuries occurring in 34 horses were recorded. Overall prevalence of horses sustaining musculoskeletal injury was 23.6%. Eight horses sustained more than one injury and 3 horses suffered re-injury (same site, same limb). Fractures of the proximal phalanx (P1) were the most common injury type (25/42, 59.5%), followed by SDFT tendinitis (8/42, 19.0%) and suspensory ligament branch desmitis (3/42, 7.1%). Musculoskeletal injury was most common in 4-year-olds. Gender was not significantly associated with overall musculoskeletal injury.

Conclusions and practical significance: A better knowledge of the distribution and prevalence of musculoskeletal injury in racing Arabians may allow earlier intervention and focus veterinary diagnostic efforts for injury detection.

Acknowledgements: The author is grateful for the co-operation of Mrs. G. Duffield and Shadwell Estate Management, Newmarket, UK.

Ethical animal research: Approved by the Ethical Committee of KLM Cargo. Horse owners gave consent for blood sampling. Sources of funding: Team Amstelstreek, Ronde Hoep, the Netherlands; Air France-KLM-Martinair Cargo; G. Klatte, Germany. Competing interests: None.

THE EFFECTS OF AMBIENT TEMPERATURE AND RELATIVE HUMIDITY ON BLOOD PARAMETERS IN HORSES DURING LONG-DISTANCE FLIGHTS

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Aim: At Schiphol Amsterdam airport more horses needed treatment for fever arriving after long-distance flights (>5 h) from hot and humid places than from temperate climate places. Our aim was to explain this finding by determining blood parameters with predictive value for early onset of illness in horses during commercial long-distance flights.

Methods: Blood was drawn prior to loading (Sample 1), halfway during the flight (Sample 2) and immediately after unloading (Sample 3) from 11 horses shipped from Houston to Amsterdam (test-group) and from 6 horses shipped from Amsterdam to New York (control-group) for complete routine screening. Prior to loading both groups had a 5 h resting period at the airport. Before and during the flights ambient air temperature (AT) and relative humidity (RH) were measured in each jet-stall.

Results: The AT for the test-group from loading until take off was 35.3 ± 1.89°C (max. 41.1°C) and RH 41%. For the control-group AT was 18.2 ± 2.1°C (max. 21.1°C) and RH 65%. From take off to arrival AT and RH for the test-group were 23.3 ± 3.0°C and 28%, and for the control-group 20.6 ± 1.4°C and 40%, respectively. Samples 1, 2 and 3 showed in the test-group higher (P<0.05) urea concentration (4.87 ± 1.06, 5.22 ± 1.06 and 5.06 ± 1.13 mmol/l) and lymphocyte count (3.85 ± 1.06, 3.48 ± 1.55 and 3.20 ± 2.12 10⁹/l) than in the control-group (3.68 ± 0.53, 4.12 ± 0.59, 4.25 ± 0.58 and 1.95 ± 0.72, 1.97 ± 0.68, 1.83 ± 0.56). In the test-group lactate concentrations (mmol/l) were significantly higher in Samples 1 and 2 (test-group 1.53 ± 0.35 and 1.84 ± 0.51; control-group 1.15 ± 0.83 and 1.35 ± 0.40), and creatinine kinase (iu/l) was higher in Sample 1 (test-group 326.0 ± 123.04; control-group 201.2 ± 69.5).

Conclusions and practical significance: Earlier studies proved that air transport is less stressful than road transport. This study demonstrates that high AT with moderate RH prior to take off resulted in significant elevation of some blood parameters. These findings support an increased susceptibility to illness, e.g. shipping-fever in horses after long-distance flights from hot and humid places.

Ethical animal research: Approved by the Ethical Commitment of KLM Cargo. Horse owners gave consent for blood sampling. Sources of funding: Team Amstelstreek, Ronde Hoep, the Netherlands; Air France-KLM-Martinair Cargo; G. Klatte, Germany. Competing interests: None.

RADIOLOGICAL CHARACTERISATION OF OSSIFIED UNGULAR CARTILAGES OF THE FOOT

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Aim: Ossified ungular cartilages may be associated with foot pain. There are no detailed descriptions of their radiological appearance. Dorsopalmar images are considered best for evaluation of ossification; the usefulness of oblique images has not been assessed. The aims were to describe the radiological appearance of ossified ungular cartilages and to determine the usefulness of oblique images.

Methods: Radiographs of all feet (n = 169, 2005–2012) with ≥ Grade 2 ossification (Dyson et al. 2010) of one/both ungular cartilages were assessed subjectively using a purpose-designed grading system, after a repeatability study in which 10 sets of radiographs were examined 10 times. Detection of abnormalities was compared among conventional and oblique images.

Results: Radiological abnormalities including focal/diffuse increase in trabecular bone opacity, loss of trabecular architecture, and loss of definition between the trabecular bone and the cortices were more frequent in lateral cartilages (72, 59 and 60, respectively) than medial (41, 36 and 33, respectively). Cartilage shape varied in dorsopalmar images, being straight (27%), curving axially (19%) or abaxially (10%). There was modelling of the cortices in 52 lateral (33 mild, 19 moderate) and 23 medial (17 mild, 6 moderate) cartilages. There were separate centres of ossification (SCsO) in 30 lateral and 14 medial cartilages. Fractures were present in 18 medial and 13 lateral cartilages, 20 at the base. Accurately distinguishing between fractures and osseous reaction around junctions between ossification centres was impossible in 8 cartilages. In total SCsO or fractures were seen in 59 dorsopalmar and 81 oblique images. Fracture sites and junctions between ossification centres were consistently best seen in oblique images. Palmar curvature of the cartilages could only be accurately detected in oblique images and was present in 31 cartilages.
Conclusions and practical significance: Lateral cartilages had a greater variation in radiological appearance than medial cartilages. Oblique images can provide information not available from other images.

Ethical animal research: Not required by this Congress: retrospective analysis of archived radiographs. Sources of funding: None. Competing interests: None.

Reference

CHARACTERISATION OF KERATOMAS OF THE EQUINE DIGIT USING CONTRAST ENHANCED COMPUTED TOMOGRAPHY (CECT) TO FACILITATE SURGICAL REMOVAL AND PROVIDE PROGNOSTIC INFORMATION REGARDING POST OPERATIVE MORBIDITY

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Aims: To characterise keratomas using contrast enhanced computed tomography (CECT) to facilitate complete surgical excision and provide information regarding post operative morbidity.

Methods: Records of horses with histologically confirmed keratomas who underwent CECT were reviewed. Historical and clinical details, CECT characteristics, surgical approach and outcome were reviewed.

Results: Ten horses, mean age 9.3 years, exhibiting recurrent or chronic grade 3 to 5/5 lameness for 5 weeks to 2 years, had histologically confirmed keratomas characterised and removed following CECT. Radiographs failed to reveal evidence of keratomas in 5/10 cases. Eleven keratoma lesions affecting the dorsal midline (n = 5), lateral hoof wall (n = 2), medial hoof wall (n = 3) and solar surface of the third phalanx (P3) (n = 1) were identified. Keratomas had a broad (n = 6) or narrow (n = 4) based attachment to the laminar surface, were classified as columnar (n = 5) or spherical (n = 5), measured 3–15 mm wide and 5–52 mm in length. Contrast material delineated keratoma margins by contrast enhancement within the lesion (n = 4) or attenuation of contrast material by the lesion (n = 6) suggesting surrounding hypervascularity or compression of surrounding vasculature, respectively. Defects in the distal phalanx (n = 5), abnormal vascularity (n = 10) and abscessation (n = 6) around the lesion were also identified. Proximal to distal and medial to lateral limits of the keratoma were marked on the hoof wall allowing complete excision via a partial hoof wall (PHWR [n = 7]) or complete hoof wall resection (CHWR [n = 3]). Soundness (mean 7 weeks) and no recurrence was achieved in all cases. Hoof wall defect healing was protracted in cases in which CECT attenuation of vascularity was evident around the defect.

Conclusions: Contrast enhanced computed tomography allows comprehensive characterisation of keratomas, determining location, vascularity and extent of laminar and P3 involvement. Accurate surgical landmarks are obtained so complete surgical excision is facilitated, the degree of hoof wall resection is minimised and recurrence is prevented. Those lesions with attenuation of vascularity around them on CECT are more likely to have protracted healing of the hoof wall defect.

Ethical animal research: Not required by this Congress: retrospective clinical study. Sources of funding: None. Competing interests: None.

USE OF TRANSOEOSOPHAGEAL ULTRASOUND TO VISUALISE THE AORTOPULMONARY REGION IN TWO NORMAL FRIESIAN HORSES AND THREE FRIEISAN WITH AORTIC RUPTURE OR AORTOPULMONARY FISTULATION

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Aims: Aortic rupture and aortopulmonary fistulation are reported with increased incidence in the Friesian breed. In contrast to Warmblood horses, the aorta in Friesian horses typically ruptures at the level of the ligamentum arteriosum (Ploeg et al. 2013). This remote location often creates difficulties in identifying the rupture with transthoracic ultrasound.

The aim of this study was to evaluate the usefulness of transoesophageal visualisation of the aortopulmonary region with a linear probe in both standing sedated horses and horses suffering from aortic rupture.

HIGH-SPEED FLUOROSCOPY: A NOVEL METHOD FOR DYNAMIC IMAGING OF THE EQUINE FOOT
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Aims: This study describes the distal limb kinematics including intra-horse and inter-horse variability, and variability between gait in sound horses using high-speed fluoroscopy which allows cineradiographic examination at speed.

Methods: Distal limb kinematics were collected at walk and trot from 6 sound horses using a high-speed fluoroscopy system set over a force plate. The dorsal proximal interphalangeal joint (PIPJ) angle and the dorsal distal interphalangeal joint (DIPI) angle were repeatedly measured at toe-on, 25°, 50° and 75% stance.

Results: The PIPJ and DIPI showed overall extension through stance. The mean (± s.d.) range of motion (ROM) during stance of the PIPJ was 10 ± 3° (walk) and 9 ± 3° (trot) and for the DIPI was 29 ± 5° (walk) and 27 ± 6° (trot) showing significant differences between strides, gait and horses (P<0.001).

Conclusions: High-speed fluoroscopy allows for kinematic assessment of the distal limb. The ROM of the PIPJ was observed to be similar to the literature whilst the ROM for DIPI was less than previously reported.

Practical significance: Kinematic analysis allows investigation of forces acting on bones, joints, ligaments and tendons. This is of special interest in the foot as the most common site of forelimb lameness in the horse; however, kinematic analysis of the foot has to date been a challenge due to the presence of the hoof capsule. The described method allows reliable assessment of foot kinematics at different gaits and speeds, which can be used for future studies to assess the effectiveness of treatment and monitor disease progression.

Ethical animal research: Ethical approval was granted by the Royal Veterinary College Ethics Committee. Sources of funding: The Royal Veterinary College. Competing interests: None.
**Methods:** Five Friesian horses, 2 healthy and 3 affected, were subjected to transthoracic ultrasound (2.5 MHz sectorial probe GE) followed by transoesophageal ultrasound (7.5 or 10 MHz linear probe). Correct oesophageal introduction of the probe was guided by means of endoscopy. After visualisation of the aortopulmonary region, horses were anaesthetised, and the transoesophageal ultrasound was repeated. In 4 horses, simultaneous carotid artery catheterisation was performed, to visualise the catheter at the zone of interest. After obtaining a diagnosis, all affected horses were subjected to euthanasia and autopsies were performed.

**Results:** The aortopulmonary region could be visualised in all horses. In the affected horses presence or absence of a periaortic blood cuff, aortic rupture and or aortopulmonary fistulation could be clearly identified. In one horse during carotid catheterisation, the catheter could be guided into the fistulation site, which was clearly visualised by means of transoesophageal ultrasound.

**Conclusions and practical significance:** Transoesophageal ultrasound by means of a 7.5 or 10 MHz linear probe allows for good visualisation of the aortopulmonary region and for diagnosing aortic rupture in Friesian horses. The technique will be further developed as an elegant noninvasive way to screen Friesian horses for this pathology, even in the standing nonsedated horse.

**Ethical animal research:** Owner informed consent was obtained for all horses. **Sources of funding:** This study was funded by the Dutch Royal Friesian Studbook and Scil Animal Care. **Competing interests:** None.

**Reference**


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**CHANGES IN INTESTINAL MUCOSAL MICROVASCULAR PERFUSION ASSESSED USING ORTHOGONAL POLARISATION SPECTRAL IMAGING IN THE HORSE**

**Aims:** Although several markers for measuring global tissue perfusion are available, there are currently no practical measurements of capillary microvascular perfusion in horses with hypovolaemia. Orthogonal polarisation spectral (OPS) imaging allows assessment of capillary microvascular perfusion by visualisation of mucosal blood flow. This study aimed to demonstrate that administration of the α2 adrenoceptor agonist detomidine, results in measurable changes in mucosal blood flow that can be determined using OPS. We hypothesise that these changes will mirror known aberrations in total peripheral resistance and cardiac output.

**Methods:** Microvascular blood flow was recorded using OPS placed manually, per rectum in 6 normal horses (weighing 603 ± 134 kg) undergoing sedation for a range of clinical procedures. The OPS recordings were made prior to and following sedation (5, 10, 20 min) with detomidine (10 μg/kg bwt) and butorphanol (10 μg/kg bwt) administered by i.v. injection. Microvascular perfusion was determined using standardised methods from OPS recordings including proportion of perfused vessels (PPV), functional capillary density (FCD), microvascular flow index (MFI) and vessel density (VD).

**Results:** Detomidine had a significant effect on microvascular blood flow as demonstrated by changes in MFI, PPV and FCD 5 min following sedation (P<0.001) and changes in VD 10 min after sedation (P<0.02).

**Microvasculature had returned to the presedation baseline by 20 min for all criterions.**

**Conclusions:** These data demonstrate that changes in organ perfusion known to be caused by α2 adrenoceptor agonists result in observable changes in mucosal blood flow. This appears to mirror changes in cardiac output and total peripheral resistance previously demonstrated after administration of detomidine in the horse.

**Practical significance:** This technique may be a useful marker to use in early goal-directed therapy in horses with systemic inflammatory response syndrome (SIRS) and severe intestinal pathology.

**Acknowledgements:** The staff at DAC Melton Mowbray for their assistance with the conduct of the study.

**Ethical animal research:** This study was approved by the University of Nottingham ethical review committee. Horses were admitted into the DAC veterinary facility for routine veterinary procedures and sedated for routine dental care or orthopaedic investigation. Full consent was given by the DAC for use of these horses in the study. **Sources of funding:** Student project undertaken at the University of Nottingham. **Competing interests:** None.

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**PREVALENCY AND RISK FACTORS FOR OWNER-REPORTED OBESITY IN HORSES AND PONIES IN GREAT BRITAIN**

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**Aims:** To estimate prevalence of owner-reported obesity in British veterinary-registered horses and ponies, and identify risk factors associated with obesity.

**Methods:** Thirty veterinary practices randomly selected horse owners to complete an ethically approved, self-administered postal questionnaire. Owners estimated body condition score using a modified Carroll and Huntington method (1–6) and animals were classified as obese if they were scored as 5 (fat) or 6 (very fat). Factors associated with obesity were assessed using logistic regression analysis.

**Results:** Owner-reported prevalence of obesity was 31.2% (n = 247/792, 95% CI 27.9–34.2). A greater proportion of obese animals (n = 47/225, 20.9%) had a history of laminitis compared with normal/underweight animals (n = 69/511, 13.3%, P = 0.01). Univariable logistic regression analysis identified several management and horse-level risk factors. Data from 785 horses were included in the final multivariable logistic regression model, and factors associated with an increased risk of obesity were breed (P<0.001), ease of maintaining weight (P<0.001) and primary use (P = 0.002). Compared with Thoroughbreds, draught-type (odds ratio [OR] 7.3; 95% CI 3.5–17.1), cob-type (OR 5.8; 95% CI 2.6–12.8), native (OR 3.1; 95% CI 1.7–5.7), and Welsh breeds (OR 3.5; 95% CI 1.9–6.2) were more likely to be obese. Animals described as ‘good doers’ were more likely to be obese compared those described as readily maintaining normal weight (OR 3.7; 95% CI 2.6–5.3). Compared with animals whose primary use was competition, animals predominantly used for pleasure riding were more likely to be obese (OR 2.5; 95% CI 1.4–4.3), and risk increased in non-ridden horses compared with competition horses (OR 2.9; 95% CI 1.5–5.5, P = 0.002).
Conclusion and practical significance: Identification of breed and other horse characteristics as risk factors for obesity will enable optimal targeting of owner education regarding preventive management to reduce the risk of obesity among the British horse population.

Ethical animal research: Owner informed consent was obtained for the questionnaire. Sources of funding: This project was funded by World Horse Welfare. Competing interests: None.

VACCINATION WITH VIRUS-LIKE PARTICLES PROTECTS HORSES FROM EXPERIMENTAL BPV-1 INFECTION

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Aims: Bovine papillomavirus types 1 and 2 (BPV-1/2) are closely related serotypes, and that intradermal inoculation of horses with cow wart-derived BPV-1 virions reliably results in the development of transient pseudo-sarcoids. Based on these data, we next performed a virus challenge study.

Methods: Fourteen horses were immunised with 100 µg of BPV-1 L1 VLP in adjuvant on Days 1 and 28; 7 horses served as nonvaccinated controls. On Day 42, all 21 horses were intradermally inoculated on their neck with 5 x 10^6 BPV-1 virions per wheal (10 wheals per horse). The horses were monitored for adverse reactions and pseudo-sarcoïd formation. Antibody titres were determined for Days 0 and 42.

Results: All 7 control horses developed pseudo-sarcoïds at every inoculation site (10/10). Tumours reached sizes of up to 16 mm in diameter and persisted for a minimum of 20 weeks. Immunisation with BPV-1 L1 VLPs resulted in complete protection in 13 of 14 vaccinated horses, with serum antibody titres ranging between 6.400 and 25.600. One vaccinated horse developed 7 tumours, which reached a maximum size of 2 mm and completely regressed within 5 weeks, with antibody titres of 800.

Conclusions and practical significance: Immunisation of 14 horses with BPV-1 L1 VLPs conferred complete protection from experimental infection with BPV-1 virion in 13 cases, and partial protection in one horse. The old age of this individual may account for the poor response to vaccination. Provided that the upcoming BPV-2 challenge will yield similar results, BPV-1 L1 VLP may be used for routine immunisation of equids.

Ethical animal research: This work was approved by the institutional ethics committee and the national authority according to Austrian Law for Animal Experiments, Tierversuchsgesetz -TVG. Approval Number: BMWF-68.205/0236-II/3b/2011. Sources of funding: This project is funded by the Veterinary University of Vienna Austrian Science Fund (FWF).

Competing interests: None.
VACCINATION AGAINST EQUINE GRASS SICKNESS: PILOTING A CLINICAL FIELD TRIAL OF A CLOSTRIDIUM BOTULINUM TYPE-C TOXOID IN SCOTLAND IN 2012–13

Methods: Only healthy horses/ponies residing on premises previously affected by a high incidence and frequency of EGS were included. Enrolled horses/ponies were randomly allocated, at premises-level, stratified by age, to vaccine or placebo treatment groups at a 1:1 ratio. Baseline and follow-up premises and horse data were obtained via telephone questionnaires. Veterinary surgeons responsible for the primary care of enrolled animals administered the primary course of vaccine/placebo on Days 0, 21, 42. Following appropriate training, owners completed post treatment daily observations for 7 days following each treatment.

Results: There were 5 participating practices, recruiting 10 EGS-affected premises in eastern Scotland. A total of 109 horses/ponies were enrolled: 13 were excluded prior to randomisation, and one was excluded following randomisation due to absence of a valid passport. Median age at enrolment was 5.5 years. Age (P = 0.34), gender (P = 0.19) and breed (P = 0.94) distributions did not differ significantly between vaccine and placebo groups. Ninety-five horses/ponies completed the primary treatment course. No significant adverse events were reported and overall prevalence of minor injection site reactions was 1.4% (n = 4/285; 95% CI 0.4–2.8%).

Conclusions and practical significance: Participant compliance has been excellent, and findings of this pilot study will be used to refine sample size calculations and inform modifications to trial methodology for a proposed future full-scale vaccine trial.

Ethical animal research: This study was conducted under an Animal Test Certificate and granted institutional ethical approval. Informed written owner consent was obtained for each animal, and procedures were in line with recognised veterinary practice as defined in The Animals (Scientific Procedures) Act 1986. Sources of funding: This study was generously funded by Neogen Corporation. Competing interests: None.
EFFECT OF PHENYL BUTAZONE, FLUNIXIN MEGLUMINE AND FIROCOXIB ON EX VIVO CYCLO-OXYGENASE ACTIVITY IN HORSES UNDERGOING ELECTIVE SURGERY

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Aims: Nonsteroidal anti-inflammatory drugs (NSAIDs) inhibit the production of prostaglandins and other inflammatory mediators by inhibiting the activity of the cyclo-oxygenase enzymes (COX). Two major isoforms of COX enzymes exist: COX-2, which is expressed during the inflammatory response, and COX-1, which is responsible for the physiological production of prostaglandin that regulates tissue homeostasis. The study aims to evaluate the effect of firocoxib ex vivo in the horse as, to the authors’ knowledge, published studies assess its effect only in vitro.

Methods: Horses (n = 18) undergoing elective surgery were recruited and allocated to treatment groups depending on clinician preference (1) phenylbutazone (4.4 mg/kg bwt i.v. b.i.d.), (2) flunixin meglumine (FM, 1.1 mg/kg bwt i.v. b.i.d.) and (3) firocoxib (FIR, 0.1 mg/kg bwt i.v. s.i.d.). Residual blood samples were collected prior to NSAIDs (T0), 2 h after NSAIDs (T2), and 24 h following surgery (T24). The COX activity was measured using validated immune-enzymatic assays. A Kruskal–Wallis test was used to determine the effect of time and treatment on COX-1 and COX-2 activity. Bonferroni corrections were used to identify the level of significance accounting for multiple comparisons (P < 0.017).

Results: At T2 and T24, the relative COX-1 activity was significantly greater in horses receiving firocoxib compared with horses receiving either phenylbutazone (P < 0.008) or flunixin meglumine (P < 0.005). At T2 and T24, COX-1 activity was reduced (compared with baseline) in horses receiving phenylbutazone or flunixin meglumine. The effect on COX-2 activity was not significantly different between drugs (P = 0.471).

Conclusions and practical significance: Cyclo-oxygenase selectivity of firocoxib is demonstrated ex vivo. Firocoxib is as effective as phenylbutazone or flunixin meglumine in modulating the production of prostaglandins by COX-2 isoenzyme, whilst the physiological action of COX-1 isoenzyme is preserved with firocoxib, but not with phenylbutazone and flunixin ex vivo.

Ethical animal research: Study approved by Ethics and Welfare Committee - School of Veterinary Medicine, College of Medical, Veterinary and Life Sciences, University of Glasgow. Sources of funding: Funding provided by John Crawford Endowment Fund, and Mrs I.J. Gates Charity Fund, University of Glasgow. Competing interests: None.

THE EFFECT OF TEMPERATURE CHANGES ON IN VITRO SLOW WAVE ACTIVITY IN THE EQUINE ILEUM

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Aims: To evaluate the effect of temperature changes on in vitro slow wave activity of the equine ileum using intracellular recording techniques.

Methods: A section of ileum was collected immediately following euthanasia from 9 normal horses euthanised for clinical reasons unrelated to the gastrointestinal tract. Intestinal tissue samples were cut into 1 mm thick sections, pinned out on a Sylgard plate and superfused with warmed, oxygenated Krebs solution. Intracellular recordings of membrane potential were made from smooth muscle cells using glass microelectrodes. All experiments were performed in the presence of a calcium channel blocker to ensure stable impalements. The temperature of the tissue bath was altered during the course of the experiment at a range of 27–41°C. All data were recorded and stored using a computer interfaced acquisition system. A software package was used to analyse the resting potentials, the amplitude, frequency and duration of slow waves.

Results: In all 9 horses slow wave frequency appeared to be approximately linearly related to the temperature over the range studied increasing by 0.5 cycles/min for each 1 degree increase in temperature (P < 0.01). The initial slow wave frequency resumed when the temperature was returned to 37°C. The recovery time appeared to be directly related to the duration for which the temperature had been changed.

Conclusions: Slow wave frequency in the equine ileum is highly temperature sensitive.

Practical significance: As post operative ileus is a major cause of morbidity and mortality in the horse, the negative effect of lower temperatures on slow wave activity should be considered. During colic surgery close attention should be paid to minimising extra-abdominal gut exposure time and keeping the temperature of the intestinal and abdominal lavage fluids at body temperature.

Ethical animal research: Post mortem samples obtained with the consent of the owners. Sources of funding: The Norwegian Agricultural Agreement Research Fund, Norsk Rikstoto and the Research Council of Norway as part of the Norwegian/Swedish research collaboration. Competing interests: None.

CHARACTERISATION OF INTESTINAL STEM CELL NICHE CONSTITUENTS IN NORMAL AND STRANGULATED EQUINE SMALL INTESTINE

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Aim: Death from colic results from acute circulatory failure secondary to intestinal ischaemia and sepsis. Multipotent stem cells normally reside within intestinal crypts and are the source of mucosal renewal that maintains barrier function. A better understanding of changes to intestinal cell populations during mucosal repair will facilitate future efforts directed at regenerative medicine. The aim of this research was to characterise the constituents of the stem cell niche in normal and strangulated equine intestine using biomarkers validated in other animal models.

Methods: Tissues were collected from 3 healthy horses subjected to euthanasia for reasons unrelated to this project, and intra-operatively from 2 horses admitted to the NCSU Veterinary Health Complex that required small intestinal resection. Tissues were examined using immunofluorescence (IF) and western blots (WB). For IF, fixed tissues were embedded and sectioned. Protein was isolated from snap frozen mucosal scrapings, and semi-quantitative analysis of protein levels between groups was conducted using WB.

Results: Stem/progenitor cells were labelled using sex determining region Y-box 9 (SOX9), a marker of stem/progenitor cells, whereas the entire population of proliferative cells was identified by labelling proliferative cell nuclear antigen. Post mitotic cell types were labelled using mucin2 [goblet cells], chromogranin A [enteroendocrine cells], beta-catenin [epithelial cells] and sucrase isomaltase [absorptive cells].
ASSOCIATIONS BETWEEN LOCAL WEATHER PATTERNS AND THE FREQUENCY OF SAND ENTERITIS IN AN EAST ANGLIAN EQUINE HOSPITAL

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Aim: To identify climatic trends associated with the frequency of sand enteritis.

Methods: Horses were included if they: (1) had radio-dense intestinal contents or sand impaction confirmed at exploratory laparotomy; (2) originated from East Anglia; (3) presented between 1 January 2005 and 31 December 2012. Historical weather data for East Anglia were obtained from the Met Office website. Associations between prevalence and climatic variables were examined separately for March–April and October–November.

Results: Ninety-two horses were included. Peaks in admissions of sand enteritis cases were seen in March–April (n = 21, 23%) and October–November (n = 25, 27%). The frequency of March–April cases was positively correlated with mean temperature in those months (r² = 0.37); negatively correlated with frost (r² = 0.58) and rainfall (r² = 0.54) in the 2 previous months; and negatively correlated with sunshine in the preceding 3 months (r² = 0.44). The frequency of October–November cases was positively correlated with higher average temperatures (r² = 0.14) and fewer frost days (r² = 0.16) in the preceding 3 months. Twenty-nine (32%) horses presented in a temporal cluster from September 2010 to April 2011 (P<0.001), coinciding with peak rainfall (121.6 mm; August 2010); peak frost (23.2 days); lowest mean temperature (0.0°C; both December 2010); and least sunshine (39.3 h/month; January 2011) recorded over the 8-year period.

Conclusions: In our population, spring cases of sand enteritis are more likely following dark, dry winters, and autumn cases more likely following dark, dry winters, and autumn cases more likely following hot summers. A wet summer followed by a cold dark winter may have contributed to the cluster of cases in 2010/2011. These weather combinations may adversely affect grass growth.

Practical significance: Weather patterns may have a role in the epidemiology of sand enteritis. Horses at pasture are likely to graze closer to the ground and ingest sand particles when grass growth is poor.

Ethical animal research: Not required by this Congress: retrospective analysis of clinical cases. Sources of funding: Polly Compston is supported by the Margaret Giffen Trust. Competing interests: None.

EQUINE HERPESVIRUS TYPE 1 (EHV-1) IN THE ENVIRONMENT: HOW LONG WILL IT STAY INFECTIVE?

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Introduction: Equine herpesvirus type 1 (EHV-1) is a highly contagious agent for horses, able to cause outbreaks of respiratory disease, myelopathy and/or abortion. Horizontal transmission is directly throughonasphyngaeal droplet transmission or, indirectly, through fomite transmission. Once in the environment viral maintenance of infectivity will depend on a variety of factors associated with viral envelope integrity. Adsorptive and surface tension forces, temperature fluctuations and UV-light exposure have been shown to affect envelope integrity in other herpesviridae.

Aims: We hypothesised that viral survival will be different if placed on various environmental surfaces or materials, and if placed in different environmental conditions.

Methods: An EHV-1 suspension was placed on surfaces or materials: plastic, fabric, leather and stall bedding materials: shavings and straw. Materials were placed in different environments: constant 4°C, ‘barn’ and ‘outdoor environment’. Samples of each material and environment were collected at time points 0, 3, 12, 24, 36 and 48 h followed by viral titration and quantitative PCR analysis. Statistical analysis used generalised linear models with random-effects mixed models controlling for repeated measures. Statistical significance was assumed when P<0.05.

Results: Results showed significant differences upon contact (t = 0) of the viral suspension with materials, most noticeable with shavings and leather. Most materials and in environmental conditions other than 4°C showed a rapid decrease in viral survival, especially during the first 3 h. While results show significant reduction on some surfaces and materials over others, it is important to realise that viral maintenance of infectivity was still significant under simulated ‘barn conditions’ following the 3 h time point.

Conclusions and practical significance: These results emphasise the importance of the prudent use of biosecurity protocols when mitigating an EHV-1 outbreak.

Ethical animal research: Not applicable. Sources of funding: 1) Student was funded from an internal grant (DVM student grant) at CSU. 2) CSU Equine Neurology Research Fund. Competing interests: None.
Day 56. Tetanus antibody titres were determined with the toxin binding inhibition test (ToBI). Immunity to equine influenza virus (EIV) induced by vaccination was measured by single radial haemolysis (SRH; antibody response) and IFN gamma assay (a marker of cell-mediated immunity; CMI). Immune responses to Herpesvirus 1,4 were measured by complement fixation (CF) and IFN gamma assay.

Results: Concurrent administration of EIV and EHV 1,4 vaccines was well tolerated. Concurrent administration was proven to be efficacious against tetanus with similar serological immune response of treatment group T04 as compared with T02. Mean SRH results for ponies that received both vaccines were above 85 mm² for A/eq/Borlange/91 and A/eq/Kentucky/98 antigens, a threshold associated with clinical protection against closely related EIV strains. The highest levels of IFN gamma response were measured in group T02. Although concurrent vaccination seemed to have an effect on both equine herpesvirus (EHV) serology and EHV CMI, EHV results obtained during this study were not conclusive and may have been influenced by a natural EHV-4 infection occurring prior to study start.

Conclusion and practical relevance: Concurrent administration of Equip® FT and Equip® EHV 1,4 was well tolerated and induced mean serum antibody levels consistent with clinical protection for tetanus and EIV. However, it was not possible to gather relevant information about the EHV immune response under the conditions of this study.

Ethical animal research: The study was conducted under the Animal Health Trust Home Office Project License and with approval of the Zaventem Ethics Review Assessment team (Zoetis). Sources of funding: The study was sponsored by Pfizer/Zoetis Animal Health. Competing interests: Equip® FT is owned by Zoetis, who sponsored this study. Romain Paillot reports no conflict of interest. All other authors are employed by the study sponsor.

COMPUTED TOMOGRAPHY VALIDATION OF THE TECHNIQUE OF DIAGNOSTIC LOCAL ANALGESIA OF THE CAUDAL PART OF THE INFRAORBITAL NERVE AND CAUDAL NASAL NERVE USED FOR THE INVESTIGATION OF IDIOPATHIC HEADSHAKING IN HORSES

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Introduction: Diagnostic local analgesia of the caudal portion of the infraorbital nerve (CPIN) and caudal nasal nerve (CNN) is a valuable aid to the diagnosis of idiopathic headshaking in horses. The site of deposition of local anaesthetic has not been verified.

Aims: To verify the site of deposition of local anaesthetic in this procedure, and to identify any correlation between accuracy of the technique and operator experience.

Methods: The procedure was performed bilaterally using contrast material on 30 cadaver horse heads by 3 groups of veterinarians and veterinary students with varying levels of experience in the technique. Location of deposition was identified by use of computed tomography (CT).

Results: Contrast was deposited around the target site in 53.3% (32/60) of injections. The most experienced operator performed the procedure accurately significantly (P<0.05) more often (80% [16/20]) than did the less and nonexperienced performers (40% [16/40]).

Conclusions: A negative response to diagnostic local analgesia of the CPIN and CNN in the investigation of headshaking does not disprove facial pain as the cause of headshaking in that horse. A negative response could arise due to failure to deposit local anaesthetic around the target area. Sufficient experience of performing the procedure decreases the probability of false negative results.

Practical significance: Clinicians performing diagnostic local analgesia of the CPIN and CNN must be aware of the possibility of false negative results. Experience improves the reliability of results.

Ethical animal research: Not required by this Congress. Horse cadaver heads obtained from horses subjected to euthanasia for reasons other than this study were used. Sources of funding: The Langford Trust for Animal Health and Welfare funded this study and Langford Veterinary Services funded the first author’s residency training. Competing interests: None.
REDUCED THRESHOLD POTENTIAL OF THE TRIGEMINAL NERVE IN EQUINE HEADSHAKING

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Aim: Although the aetiopathogenesis of headshaking remains elusive, trigeminal neuralgia is regarded as the likely explanation of observed clinical signs. The aim of this study was to compare sensory nerve conduction threshold and velocity of the trigeminal nerve using the infraorbital nerve in control and headshaking horses.

Methods: Control group (n = 6 horses) and headshaking group (n = 6) were subject to general anaesthesia. A Nicolet Viking IV evoked potential system was used for sensory nerve conduction study. Stimulating, recording and reference electrodes were used. A pair of stimulating electrodes was placed at the gingival mucosa, one each at the rostral and caudal aspects of the maxillary canine tooth. Four pairs of recording electrodes were placed at 4 different points along the tract of the infraorbital nerve (point 1 at infraorbital foramen), maxillary nerve (point 2 at exit of trigeminal canal), spinal somatosensory (point 3 at level of C1 spinal cord segment), and cortical somatosensory evoked potentials (point 4 at level of frontal cerebral cortex). A reference electrode was placed at a distance half way between the stimulating and recording electrodes at point 1. Stimuli were applied at 2.5, 5, 10, 15, 20 mA. The duration of each stimulus was 0.1 ms. Thresholds were recorded and conduction velocities calculated for each group.

Results: The threshold of sensory nerve action potential occurred at low stimuli (2.5 and 5 mA) in horses with headshaking; and at higher stimuli (10 mA in 3 horses, 15 mA in 1 horse, and 25 mA in 2 horses) in control horses. Although there were differences in conduction velocity between both groups, this was not significant.

Conclusions and practical significance: Headshaking horses have a low threshold for inducing sensory action potentials upon minimal stimulation compared with control horses supporting involvement of the trigeminal nerve in the pathogenesis of affected horses.

Ethical animal research: Study approved by the UC Davis IACUC.
Sources of funding: Private donation. Competing interests: None.

PRELIMINARY DATA OF A RETROSPECTIVE STUDY ON NEUROLOGICAL SIDE EFFECTS AFTER ADMINISTRATION OF POLYMIXIN B TO ENDOTOXAEMIC HORSES

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Aims: This retrospective study reports the occurrence of neurological side effects attributed to the use of polymyxin B in horses treated for endotoxaemia.

Methods: Between January 2012 and January 2013 18 horses were treated for endotoxaemia with 5000 iu polymyxin/kg bw intravenously q. 8 h. For this purpose a sterile polymyxin solution was compounded by a pharmacy: 500 ml 0.9% NaCl contained 2.5 Mio. iu polymyxin B. Depending on the disease which led to endotoxaemia the horses received other treatments as well. Horses were examined at regular intervals and ataxia was graded using the modified Mayhew grading scale.

Results: Sixty-six per cent of patients were mares, 28% were geldings and the rest stallions. Age ranged from one to 23 years, with a mean (± s.d.) of 12 (± 7) years. Ten of 18 horses were treated for colitis, 2 of 18 each for small intestinal strangulating lesion, aspiration pneumonia and large colon torsion. In 6 horses a weak, ataxic gait was noted between 24 and 36 h after start of polymyxin treatment. The only factors significantly associated with ataxia were the number of doses of polymyxin the horses received (P = 0.011) and the concurrent use of neostigmine (P = 0.025). No other treatments were associated with occurrence of ataxia. The level of ataxia observed was correlated with the time necessary for ataxia to resolve. Horses which had shown ataxia after polymyxin treatment had a significantly longer overall hospitalisation time (P = 0.004).

Conclusions and practical significance: Self-limiting weak, ataxic gait was observed in horses treated with polymyxin B for endotoxaemia.
Horses receiving multiple doses of polymyxin seem to be at risk. A cumulative effect of polymyxin might be suspected. Furthermore neostigmine could be responsible for potentiating polymyxin side effects. Neurological side effects of polymyxin at dose rates used for anti-endotoxic treatment need to be further elucidated.

**Ethical animal research:** Not required by this Congress: retrospective study. **Sources of funding:** None. **Competing interests:** None.

### PHARMACOKINETICS OF PERGOLIDE MESYLATE IN HORSES

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**Aims:** Pergolide mesylate is the treatment of choice for equine pituitary pars intermedia dysfunction (PPID) and veterinary preparations are now licensed for this use in a number of countries. Pharmacokinetic properties of the drug have not been established completely in horses and current dosing recommendations are based upon clinical experience. This study aimed to establish the pharmacokinetic properties of the drug following intravenous administration.

**Methods:** Eight healthy Thoroughbred or Standardbred geldings were administered 0.02 mg/kg bwt pergolide mesylate via an intravenous jugular catheter. Blood samples were collected over a period of 48 h from a catheter in the contralateral jugular vein for determination of plasma pergolide concentrations. Pergolide concentrations in plasma were determined using a high-performance liquid chromatography–tandem mass spectrometry assay. Maximum concentration of pergolide was determined directly from the data. Other pharmacokinetic parameters were determined for each horse by use of noncompartmental analysis with a commercial software program. Area under the curve was calculated by the linear trapezoidal rule. The terminal elimination rate constant and terminal half-life were calculated by means of log-linear regression. Initial volume of distribution, mean residence time, and clearance were calculated using standard noncompartmental formulae.

**Results:** See Table 1.

<table>
<thead>
<tr>
<th>Table 1: Results</th>
<th>Mean (± s.d.)</th>
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</thead>
<tbody>
<tr>
<td>Maximum concentration (C_{max})</td>
<td>14.90 ± 5.02</td>
</tr>
<tr>
<td>Terminal elimination rate constant (λ) (h⁻¹)</td>
<td>0.14 ± 0.06</td>
</tr>
<tr>
<td>Terminal half-life (h)</td>
<td>5.80 ± 2.26</td>
</tr>
<tr>
<td>Area under the curve 0–∞ (ng·h/ml)</td>
<td>18.63 ± 7.30</td>
</tr>
<tr>
<td>Mean residence time (h)</td>
<td>6.30 ± 2.43</td>
</tr>
<tr>
<td>Clearance (ml/h/kg bwt)</td>
<td>964.30 ± 460.95</td>
</tr>
<tr>
<td>Initial volume of distribution (/kg bwt)</td>
<td>1.13 ± 0.39</td>
</tr>
</tbody>
</table>

**Conclusions:** Volume of distribution and half-life for pergolide in horses are shorter than reported previously. There is no rationale for administering a loading dose of pergolide mesylate; however, based on its pharmacokinetic properties twice daily dosing with pergolide mesylate may be more appropriate than once daily dosing.

**Ethical animal research:** Approved by the Animal Care and Ethics Committee, Charles Sturt University. **Sources of funding:** Internal funding. **Competing interests:** Although not used in the current study, David Rendle has acted as a paid speaker and consultant for Boehringer Ingelheim, manufacturers of Prascend: the licensed form of pergolide in Europe and the USA.

### EFFECTS OF PERGOLIDE MESYLATE ON PLASMA ADRENOCORTICOTROPIC HORMONE CONCENTRATION IN HORSES WITH PITUITARY PARS INTERMEDIA DYSFUNCTION

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**Aims:** Pituitary pars intermedia dysfunction (PPID) is a common degenerative neuropathy of horses associated with ageing. Pergolide mesylate (pergolide) is the only licensed treatment for PPID in the UK; however, published evidence of its efficacy is limited to small case series and anecdotal reports. This study evaluated adrenocorticotropic hormone (ACTH) responses in horses treated with pergolide and investigated factors that may influence response to treatment.

**Methods:** A retrospective review of submissions to The Liphook Equine Hospital Laboratory was performed from January 2007 to December 2012 and cases in which ACTH concentration was measured before and after instigation of pergolide treatment were identified. Data were analysed using Rv.2.15 software (R Development Core Team). Improvement was defined as a reduction of ACTH concentration of ≥75% or a return of ACTH concentration to within seasonally adjusted reference intervals.

**Results:** A total of 2122 cases satisfied the inclusion criteria. Improvement was identified in 54.8% of horses at the first follow-up assessment; however, ACTH concentration returned to within reference intervals in only 28%. Equids with a higher ACTH concentration pretreatment were more likely to improve (odds ratio [OR] 0.56, 95% confidence intervals [CI] 1.00–1.02; P = 0.027), but less likely to return to within the reference interval (OR 0.97; 95% CI 0.96–0.98; P<0.001). Older equids were significantly less likely to improve (OR 0.56, 95% CI 0.46–0.67; P<0.001). Improvement was more likely after a cumulative dose of 50 mg (OR 1.59, 95% CI 1.10–2.29; P = 0.013) and duration of treatment was positively associated with treatment response (P = 0.039). A daily dose >0.5 g/kg bwt was less likely to be associated with a reduction in ACTH concentration (OR 0.86, 95% CI 0.77–0.96; P = 0.009). Native breeds, mini and draught horses were more likely to return to the reference range than other breeds (OR 1.6, 95% CI 1.24–2.07; P<0.001). Neither gender nor season were associated with response to treatment.

**Conclusions and practical significance:** Pergolide is an effective means of reducing ACTH concentration in equids with PPID.

**Ethical animal research:** Not required by this Congress: retrospective clinical study. **Sources of funding:** None. **Competing interests:** Rendle and Durham have acted as paid speakers and consultants for Boehringer Ingelheim, manufacturers of Prascend, the licensed equine pergolide product.