11.00–11.15
A nested case–control study to identify risk factors for recurrent colic

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Aims: This study aimed to quantify time-varying and non-time-varying risk factors for recurrent colic among the veterinary-visiting horse population in North West UK. Methods: Cases and unmatched, randomly selected controls were selected from a previous longitudinal cohort study of 127 horses recruited via first opinion equine veterinary surgeons subsequent to an episode of medical colic (Scantlebury et al. 2011). Telephone questionnaires collected data on management exposures and recurrence of colic at 4-monthly intervals. All recurrent colic episodes were selected as cases and the data relating to 30 days prior to the colic were used to determine exposure status. Three controls per case were randomly selected from the cumulative horse time at risk. Multivariable logistic regression analyses were used to determine risk factors for recurrent colic with a random effect term for ‘horse’. Results: In total, 59 cases and 177 controls were included. The final model showed that horses that displayed crib-biting/lvinsucking behaviour (CBWS) (OR 10.1, 95% CI 2.5–41.0 P < 0.001), were weaned (OR 3.9, 95% CI 1.5–10.1, P = 0.004), had reduced access to pasture (OR 0.99, 95% CI 0.99–1.0, P = 0.005) and were fed probiotics (OR 2.4, 95% CI 0.99–6.0, P = 0.06 included as improved model fit) had an increased risk of colic. Conclusions and practical significance: In line with recognised veterinary practice as defined in The Animals (Scientific Procedures) Act 1986, 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.

References

11.15–11.30
Vaccination against equine grass sickness: Piloting a clinical field trial of a Clostridium botulinum type-C toxoid in Scotland in 2012–13

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Aims: To conduct a pilot study to inform sample size and trial methodology for a proposed triple-blinded randomised placebo-controlled field trial for a Clostridium botulinum type C toxoid vaccine in the prevention of equine grass sickness (EGS). 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.
Validation of two novel hand-held lactate monitors in horses

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Aims: Increased blood L-lactate concentration is commonly associated with disease severity and mortality in equine adults and neonates. The ISTAT1 portable blood gas analyser has been validated for measuring lactate concentration in equine blood. A modified version of the ISTAT (EPOC), and a small lactate-only device (EDGE), have not been validated in horses previously. The aim of this study was to validate these devices using samples from adult colic cases and critical care cases. Methods: Heparinised blood samples were collected from horses exhibiting clinical signs of abdominal disease (n = 35) or other illnesses (n = 20) necessitating critical care. Samples were tested within 5 min of collection and were separated into aliquots for simultaneous analysis with the EPOC, EDGE and ISTAT (gold standard). Intra-assay precision of EPOC and EDGE was assessed by duplicate analysis of the first 25 samples. Results: Linear regression analyses between lactate concentrations obtained from EPOC vs. ISTAT (r² = 0.989; P<0.0001) and EDGE vs. ISTAT (r² = 0.904; P<0.0001) correlated well. Compared with the ISTAT, the EPOC and EDGE underestimated lactate concentrations, with a mean bias of -0.352 mmol/l (95% limits of agreement: -1.274 to 0.569 mmol/l) and -0.413 mmol/l (95% limits of agreement: -1.966 to 1.140 mmol/l) respectively. Good intra-assay agreement was shown between duplicate EPOC samples (r² = 0.991; P<0.0001) and duplicate EDGE samples (r² = 0.975; P<0.0001). Conclusions: Both devices demonstrated good agreement with the previously validated ISTAT. A degree of bias was detected, which is unlikely to be clinically relevant at high lactate concentrations. Around the upper reference interval for normal horses the bias may influence clinical decision making. Practical significance: The results of this study support the use of the EPOC and EDGE in a clinical setting, particularly for diagnostic and prognostic purposes. However, when lactate concentrations are near the upper reference interval, and the devices are used for serial monitoring of critical care cases, results may need to be interpreted with caution.

Manufacturers’ addresses

†Heska Corporation, Fort Collins, Colorado, USA.
‡EPOC – EPCAL, INC. Ottawa, Canada.
†Edwards Hand-Held Lactate Monitor: APEX Biotechnology Corporation, Hsinchu, Taiwan.

Is detomidine suitable for colic in horses?

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Aims: Use of detomidine, a potent α2 agonist equine sedative and analgesic is still controversial in horses with colic, especially regarding gut motility or re-administration. Methods: In order to learn more about induced ileus and re-administration effects, 10 healthy adult horses were administered nasogastric charcoal (1 g/kg bwt) prior to one of 4 sedation protocols, alternatively given to all of them: S1 = no sedation, S2 = 0.02 mg/kg bwt i.v. detomidine, S3 = 0.04 mg/kg bwt sublingual detomidine, S4 = 0.2 mg/kg bwt, 0–45 min later. Heart rate, sedation and head position (0 to 2), as well as gut motility (abdominal sounds, 0 to 3), auscultation) were recorded every 5 then 15 min for 210 mins. Time for dark faeces (hours) and side effects were also monitored. Results: S2, S3 and S4 provided similar profound sedation. S4 induced a significant longer max-sedation time (200 min vs. 110 min) and 210 mins with more sweating. Head raised back to nearly normal position at T210. Gut sounds decreased with all protocols, without any statistical difference, but never disappeared. Conclusions: Use of detomidine and re-administration can be considered safe, regarding gut motility, for practical use in horses with colic.

Effect of phenylbutazone, 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.), (2) flunixin meglumine (1.1 mg/kg bwt i.v.) and (3) firocoxib (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 Kruskall–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 co-treated 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 (P<0.05). At T0 and T24, COX-2 activity was reduced compared with baseline.
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 euthanased 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 27–41ºC. 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 37ºC. 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), betacatenin (epithelial cells) and sucrase isomaltase (absorptive cells). Additionally, caspase 3 positively marked apoptotic cells. Western blots indicated increased apoptosis and decreased proliferation, but preservation of stem/progenitor cell populations in ischaemic cases. Sample collection continues on normal and ischaemic-injured intestinal epithelium to support statistical differences. Conclusions and practical significance: These findings provide the technical platform for identifying distinct cell populations within the stem cell niche. These preliminary results indicate that surgically resected segments may be a viable source of stem/progenitor cells to induce regeneration in injured intestine. Ethical animal research: The Institutional Animal Care and Use Committee approved all animal studies and client consent was obtained. Sources of funding: 1) NJH/NCSU Comparative Medicine and Translational Research Training Program (CMTRTP) T32RR024394. 2) North Carolina Horse Council. Competing interests: None.