Diagnosis and Treatment of Undifferentiated and Infectious Acute Diarrhea in the Adult Horse

Sarah D. Shaw, DVM[a,b,1], Henry Stämpfli, DVM, Dr Med Vet[b,*]

KEYWORDS

- Equine - Colitis - Typhlocolitis - Infectious diarrhea - Intravenous fluid therapy

KEY POINTS

- Strict biosecurity measures should be enforced for all cases of acute diarrhea in adult horses.
- Diagnostic tests for salmonellosis, clostridiosis, coronavirus, and Potomac horse fever are evolving.
- Aims of treatment of acute diarrhea include fluid resuscitation, correction of electrolyte abnormalities, and limiting the systemic inflammatory response.
- Limited evidence exists to support many of the medications used to treat acute diarrhea and the judicious use of therapeutics is warranted.

INTRODUCTION

Acute diarrhea associated with colitis or typhlocolitis is a major cause of morbidity in horses and is life-threatening. Clinical signs of colic, hypovolemia, and endotoxemia result from altered motility, hypersecretion of fluid, and disruption of the mucosal barrier secondary to intestinal inflammation.

Undifferentiated and infectious acute diarrhea is a diagnostic and therapeutic challenge. Differential diagnoses for the acutely diarrheic horse share similar clinical and clinicopathologic features. Determination of causation is rarely possible. The fundamental diagnostic approach includes assessment of hydration, electrolyte and

---

acid-base abnormalities, mucosal integrity, organ function, and the inflammatory response. Immediate therapeutic intervention, often in the absence of a definitive diagnosis, reduces comorbidities and mortality.

INITIAL DIAGNOSTICS
Bloodwork

Hematologic abnormalities seen early in the course of gastrointestinal disease reflect stress and endotoxemia. Leukopenia may be characterized by lymphopenia, neutropenia with or without a left shift, and toxic changes in neutrophils. A neutrophilic leukocytosis may be seen later. Degenerative left shifts and the presence of metamyelocytes and myelocytes are poor prognostic indicators.[1] Hemoconcentration and thrombocytopenia are common. Horses with a packed cell volume greater than 45% were 3.5 times less likely to survive.[2] Hyperfibrinogenemia and elevated serum amyloid A may be seen in acute, severe, colitis[3]

Diarrhea in horses is associated with hemodynamic and electrolyte changes caused by intraluminal sequestration of fluid. Serum biochemistry often reveals renal or prerenal azotemia and electrolyte abnormalities including hyponatremia, hypochloremia, hypokalemia, and hypocalcemia. In one retrospective, negative base excess was the best prognostic indicator.[4] In a study of 101 horses, plasma lactate at admission was not associated with survival status. However, reduction in serial lactate concentration by greater than or equal to 30% 4 to 8 hours and by greater than or equal to 50% 24 hours after admission was significantly associated with survival.[5] A creatinine concentration greater than 2.0 mg/dL (176.8 μmol/L) was also associated with a lower likelihood of survival.[2] Hyperproteinemia may be present with severe dehydration, although mild to severe hypoproteinemia is also seen as a result of gastrointestinal protein loss.

Ultrasonography

Abdominal ultrasonography is an important diagnostic aid in cases of acute diarrhea and should be performed to assess large and small intestinal wall thickness and peritoneal fluid volume and character. See Nicola C. Cribb, Luis G. Arroyo's article, "Techniques and Accuracy of Abdominal Ultrasound in Gastro-Intestinal Diseases of Horses and Foals," in this issue for more details.

Pathogen-Specific Serologic and Fecal Diagnostics

Commercial laboratories frequently offer diarrhea panels, which are helpful for screening for numerous pathogens. However, like any screening test, specificity may be compromised and results should be interpreted with caution. Furthermore, some diagnostic laboratories include tests that are unnecessary or not clinically relevant in the adult horse. **Table 1** provides a summary of commercially available fecal and serologic diagnostics and their limitations.

DIFFERENTIAL DIAGNOSES
Salmonellosis

Salmonellosis is a well-recognized cause of acute diarrhea in the horse. It is reported as a result of infection with *Salmonella enterica* subsp. *enterica*, a G⁻, facultative anaerobic bacterium. Numerous serovars are associated with clinical disease but *Salmonella typhimurium* is commonly isolated from diarrheic horses and associated with high pathogenicity.[6] *Salmonella* infection in adult horses ranges from the inapparent carrier state, to pyrexia, anorexia, leukopenia, and depression without diarrhea, to acute, severe, enterocolitis with diarrhea.

Table 1
Diagnostic tests for acute diarrhea in the adult horse

Pathogen	Diagnostic Tests	Comments
Salmonella spp	Fecal PCR	Potentially highly sensitive and specific. Fast turnaround time. Serial samples needed.
	Fecal culture	Slower but provides isolates for susceptibility testing and typing. Serial samples needed. Sensitivity is impacted greatly by laboratory methods, such as enrichment culture, and laboratory experience.
Clostridium difficile	Culture (isolation with selective media)	Extra testing required to determine if isolates are toxigenic.
	Antigen ELISA	Quick and inexpensive. Very sensitive but lower specificity. Will detect toxigenic and nontoxigenic strains. Most often used as in initial screening test because of high negative predictive value, with positives tested by ELISA or PCR.
	Toxin A/B II ELISA, (or cytopathic effect cell culture)	Performance of different commercial assays on horse feces is likely highly variable. Has been clinical standard for diagnosis.
	PCR for TcdA and TcdB	Can be quick and highly sensitive. False-positives can occur because of detection of carriers, which is common in some populations.
Clostridium perfringens	Culture	Low diagnostic yield because of common shedding by healthy horses.
	Culture + PCR	PCR to detect selected toxin genes can increase diagnostic relevance, particularly with genes more commonly associated with disease (eg, NetF, beta2).
	Toxin-ELISA	Currently restricted to enterotoxin. Positive results are suggestive but enterotoxin is detected in some healthy horses.
	PCR	Genotyping is available in specialized laboratories.
Coronavirus	Fecal PCR	Epidemiology of ECoV is not well established but positive results in horses with disease consistent with ECoV infection provides a presumptive diagnosis.
Neorickettsia risticii	Fecal and blood PCR	Detection in blood may be more sensitive early in the course of disease; horses become PCR positive on feces later.
	Serology (IFA)	Single serum samples are not diagnostic.

Abbreviations: ECoV, equine coronavirus; ELISA, enzyme-linked immunosorbent assay; IFA, indirect fluorescent antibody; PCR, polymerase chain reaction; TcdA, *C difficile* toxin A; TcdB, *C difficile* are toxin B.

Clinically apparent salmonellosis in adult horses most commonly manifests as enterocolitis with large-volume, watery diarrhea with subsequent bacteremia.[7] Profound neutropenia is characteristic in *Salmonella* infections and results from neutrophil margination, invasion into the intestinal villi, and loss in the intestinal lumen.[8] Intestinal mucosal damage permits bacterial entry into the portal circulation, potentially damaging hepatocytes and increasing serum levels of sorbitol dehydrogenase and aspartate transaminase.[8] Dehydration is severe and is associated with prerenal

azotemia and decreased blood pH. Clinical signs of pyrexia, inappetance, diarrhea, and colic are largely a result of the host inflammatory response.

Risk factors for infection include recent abdominal surgery, antimicrobial administration, transportation, gastrointestinal disease, immunosuppression, diet changes, respiratory disease, high ambient temperature, and general anesthesia.[9–11] Historically, a diagnosis of *Salmonella* colitis was made through identification of the organism on fecal culture, but it is only intermittently shed. Real-time quantitative polymerase chain reaction (qPCR) has largely replaced culture based on shorter time to results, increased relative sensitivity, and potentially fewer serial samples required to detect *Salmonella*.[7,12] Between three and five serial fecal samples (for PCR or culture), depending on laboratory methods, are recommended to determine negative *Salmonella* status.

Treatment in early disease is mainly supportive with intravenous, isotonic fluids directed at volume replacement and to buy time for tissue recovery. Acid-base and electrolyte abnormalities should be corrected and inflammation controlled. Severe leukopenia frequently occurs in cases of *Salmonella* infection and warrants consideration of antimicrobial therapy with aminoglycosides, fluoroquinolones, or cephalosporins. Given the propensity for this organism to cause microthrombosis and infarction, anticoagulant therapy may be considered. Nosocomial *Salmonella* infections and hospital biosecurity have been the focus of recent literature. Outbreaks in teaching hospitals have resulted in prolonged closures, significant economic losses, and decreased clinic time for veterinary students. Strict biosecurity measures should be enforced in cases of acute diarrhea to prevent the potential nosocomial spread of *Salmonella* spp and other highly infectious organisms.

Clostridiosis

Colitis as a result of *Clostridium difficile* infection (CDI) was first described in horses in 1987. *Clostridium perfringens* and *C difficile* are the clostridial species most commonly associated with diarrhea, with sporadic cases attributed to *Clostridium septicum*, *Clostridium sordelli*, and *Clostridium cadaveris*.

Clostridium difficile

C difficile is a common isolate in cases of colitis in adult horses. CDI has received recent attention in the human literature, although the most prevalent ribotypes responsible for disease differ between humans and animals. *C difficile* is isolated from the manure of healthy horses and prevalence rates range from 0% to 33%.[13,14] Thus, the carrier state in horses and livestock has been suggested as a reservoir for human CDI.

The most important risk factors for CDI in adult horses are antimicrobial administration and hospitalization. Isolation of toxigenic *C difficile* has been associated with the administration of all antimicrobials, most notably β-lactams, gentamicin, trimethoprim sulfonamides, clindamycin, erythromycin, and rifampicin.[14] Proton pump inhibitors and H_2 antagonists have been implicated in the development of CDI in horses, but strong evidence is lacking.

The most important virulence factors of *C difficile* are toxin A (TcdA, an enterotoxin) and toxin B (TcdB, a cytotoxin). Clinical disease, however, only occurs when the normal gastrointestinal flora is disrupted and toxigenic strains of *C difficile* proliferate. In humans CDI is acute or chronic, whereas chronic CDI in horses has not been described.

C difficile colitis varies from peracute colitis with a rapid decline to a milder, more prolonged course. Horses may be pyrexic, depressed, and anorexic before

developing diarrhea and gastrointestinal signs. Neutropenia, hypoproteinemia, dehydration, hemoconcentration, and electrolyte and acid-base abnormalities may all be appreciated. *C difficile* is challenging to culture so false-negative test results are possible and CDI is therefore likely underdiagnosed. Enzyme immunoassays have been developed for the identification of TcdA and TcdB, although their performance is highly variable. More recently, molecular detection methods following bacterial culture have been used to identify genes for TcdA and TcdB in the feces of horses.[15,16]

Aside from supportive care, specific treatments for CDI remain controversial. Evidence exists that *Saccharomyces boulardii*, a nonpathogenic yeast, can destroy TcdA.[17] Di-tri-octahedral smectite binds TcdA and TcdB in a dose-dependent manner in vitro and may be useful to limit the toxemic effects of these toxins. Metronidazole is commonly used to treat *C difficile* colitis in horses to reduce the *C difficile* population, whereas the normal flora is restored. Although metronidazole resistance has been documented in cases of human infection, it is not a prevalent feature in equine CDI.[14] However, even targeted antimicrobial therapy can have deleterious effects on the microbiota and may disrupt or delay recolonization with beneficial bacteria. Recently, fecal transfaunation, also known as fecal microbial transplantation or fecal bacteriotherapy, has re-emerged as a treatment of CDI in humans and horses. Proximal duodenal infusion with donor feces was significantly more effective than standard vancomycin therapy in humans with recurrent CDI.[18] Anecdotal evidence supports the use of fecal microbial slurries via nasogastric intubation for treatment of acute cases of equine CDI.[19] Further investigation is needed to provide evidence-based support.

Clostridium perfringens

C perfringens is associated with occasional cases of acute diarrhea in adult horses. It has been isolated from the gastrointestinal flora of normal horses, although reported prevalence rates are low, ranging from 0% to 8% in healthy adult horses.[13] It is classified into five types (A to E) based on exotoxin production. *C perfringens* type A is the isolate most commonly cultured from healthy and diarrheic adult horses. β2 toxin, suspected to be produced by a subtype of *C perfringens* type A, has been associated with clinical *C perfringens* infections in horses with colitis.[20,21] β-toxin produced by types B and C has direct cytotoxic effects resulting in enterocyte necrosis, mucosal ulceration, and hemorrhagic diarrhea. Enterotoxin, produced by both type A and type C strains, creates pores and alters membrane permeability, leading to cell necrosis. The poreforming toxin NetF has also been recently identified as a major virulence determinant and has been associated with necrotizing enteritis in foals and dogs.

Clinical signs of *C perfringens* vary from peracute fatal colitis to a milder presentation with anorexia, depression, and pyrexia. Gastrointestinal signs include profuse, watery diarrhea that may be hemorrhagic. Affected horses demonstrate nonspecific signs of colic, endotoxemia, and dehydration. Given its presence in the feces of normal horses, the diagnosis of *C perfringens* colitis is challenging. Quantitative fecal culture no longer supports a diagnosis of *C perfringens* colitis. Enzyme immunoassays for the *C perfringens* enterotoxin detect its presence in healthy and clinically affected horses. More recently, PCR has been used to detect toxin genes, although not proving a causal relation.[15] Therapy is mainly supportive. Specific therapy aimed at binding exotoxins includes treatment with di-tri-octahedral smectite.[22,23] There is limited evidence of the benefits of metronidazole therapy in cases of *C perfringens* colitis.

Coronavirus

Equine coronavirus (ECoV) has recently emerged as a significant enteric pathogen in adult horses.[24,25] Although coronaviruses have been long identified in the feces of

adult horses and foals, their role in the pathogenesis of enteric fever and diarrhea was only recently elucidated after outbreaks of clinical disease. ECoV is an enveloped, positive-stranded RNA virus belonging to the Betacoronavirus-1 genus. Transmission is believed to be via the fecal-oral route. Infection begins in the small intestine. Lesions include necrosis and sloughing of enterocytes in the intestinal villi, dilated crypts filled with necrotic debris, and mucosal inflammation.[25,26]

Hematologic abnormalities include leukopenia, characterized by neutropenia and/or lymphopenia.[25] The most common clinical manifestations of ECoV infection include anorexia, lethargy, and fever.[22,25] In a report of 59 clinical cases, diarrhea and colic were only seen 20% and 7% of affected horses, respectively.[25] Neurologic signs associated with ECoV infection have been described and seem to be associated with hyperammonemic encephalopathy with Alzheimer type II astrocytosis in the cerebral cortex.[26]

Diagnosis of ECoV colitis is currently based on PCR of feces, gastrointestinal contents, or tissue samples. In one study there was no significant statistical difference in absolute ECoV quantification between positive sick and positive healthy horses. However, the overall agreement between clinical status (horses demonstrating clinical signs of ECoV infection) and ECoV-positive status on PCR was 91%.[25] Therapy is directed at supportive care and prevention of hyperammonemic encephalopathy.

Potomac Horse Fever

Potomac horse fever (PHF), or equine neorickettsiosis, was first recognized as a cause of acute typhlocolitis in horses in the United States in 1984 along the Potomac River area and has since been described in Canada, Uruguay, Brazil, and Europe. This acute, potentially fatal disease is an infection with the gram-negative, obligate, intracellular, bacterial endosymbiont *Neorickettsia risticii*.[27] The bacterium survives in digenic trematodes, such as *Acanthatrium* and *Lecithodendrium* spp.[28] Trematodes use freshwater and lymnaeid snails as first intermediate hosts and aquatic insects (mayflies, caddisflies, damselflies, dragonflies, and stoneflies) as second intermediate hosts.[29,30] The definitive hosts of these trematodes are brown bats (*Eptesicus fuscus* and *Myotis lucifugus*). Horses become accidental hosts of digenic trematodes when they inadvertently ingest infected intermediate hosts. *N risticii* is released from the trematode and once inside the equine gastrointestinal tract it invades and replicates within the colonic epithelial cells. It also translocates into blood monocytes (equine monocytic ehrlichiosis), mast cells, and tissue macrophages. A seasonal rise in disease incidence is seen during warmer months.

The most common clinical signs of PHF are diarrhea, fever, anorexia, depression, and colic.[30] Abortions may also occur as a result of *N risticii* infection. In one retrospective of 44 horses with PHF, laminitis was identified in 36%, of which 88% were affected in all four feet.[30] Horses present with clinicopathologic abnormalities typically seen in equine colitis. The same study identified that serum creatinine and urea nitrogen concentrations, hematocrit, red blood count, blood hemoglobin concentration, band neutrophils, serum aspartate transaminase, serum CK, and anion gap were significantly higher in nonsurvivors. Further, serum chloride, serum sodium, and duration of hospitalization were significantly lower in nonsurvivors.[30]

Diagnostics for PHF include serum indirect fluorescent antibody testing, enzyme-linked immunosorbent assay, culture of *N risticii* from buffy coat or feces, and PCR on whole blood or feces. Both exposure and vaccination lead to many false positives with antibody testing and in one study 16% of healthy horses had a rise in paired indirect fluorescent antibody titers,[31] so antigen testing is recommended. Culture of *N*

risticii remains the gold standard of diagnosis, although PCR was reported to identify 81% of culture-positive, naturally infected horses.[32]

Tetracycline antibiotics effectively kill *N risticii* and oxytetracycline administration to horses with clinical signs of disease has been positively associated with survival.[30] Additional therapy is supportive and includes fluid replacement and laminitis prevention. Particular attention should be paid to fluid resuscitation in hypovolemic horses either before, or in conjunction with, administration of nephrotoxic drugs (oxytetracycline and nonsteroidal anti-inflammatory drugs [NSAIDs]). Several killed and adjuvants vaccines have been marketed. However, vaccine failure rates are high,[33,34] likely caused by the antigenic differences present among greater than 14 *N rickettsia* strains isolated from clinical cases.[27]

TREATMENT AND SUPPORTIVE CARE
Fluid Therapy

Fluid therapy in gastrointestinal disease has been covered in depth.[35] Fluid plans should be based on clinical and clinicopathologic assessment. Fluid deficits are based on the formula:

Volume deficit (L) = Bwt (kg) * % dehydration

A generally accepted approach is replacement of the volume deficit at a rate of 10 to 20 mL/kg/h.[35] The patient should be reassessed every 4 to 6 hours and changes to the fluid plan made according to changes in hydration status. Maintenance requirements and ongoing fluid losses should also be calculated to administer fluids at an appropriate rate:

Maintenance fluid volume = 50 to 100 mL/kg per 24 hours

The goal of fluid therapy is volume resuscitation and correction of lactic acidosis; commercially available, balanced, isotonic fluids should be administered. Fluid supplementation decisions should be made based on serum electrolytes and acid-base status. Lactic acidosis caused by dehydration is common. Hypokalemia is common in anorectic horses with gastrointestinal disease and should be addressed when serum potassium is less than 3 mEq/L. Sodium bicarbonate supplementation is considered if the base deficit is greater than -10 mEq/L or the pH is less than 7.2. The required amount of sodium bicarbonate should be administered as an isotonic solution over a period of 24 hours.[35] It is important to remember that the administration of sodium bicarbonate does not correct a metabolic acidosis (lactic acidosis) secondary to dehydration. Therefore, sodium bicarbonate should be used as adjunct therapy to balanced isotonic fluids, and only if metabolic acidosis is severe. The amount of sodium bicarbonate required is estimated using the following formula:

$Na-HCO_3$ (mmoL/L) = BE x BW x 0.3

An ionized calcium level of less than 1.4 mg/dL (0.3493 mmol/L)[35] and decreased intestinal motility are indications for calcium supplementation.

Colloid Oncotic Support

The general indications for the use of colloids include hypovolemia, hyproteinemia, and decreased osmotic pressure. Because hypoproteinemia is common in horses with diarrhea, synthetic colloids and commercial equine plasma are often administered.

The literature evaluating the use of synthetic colloids in horses is limited to experimental studies and small population clinical evaluations. Hydroxyethyl starch (HES) increased colloid osmotic pressure (COP) in normal ponies but had dose-dependent effects on hemostatic variables, leading a trend in prolongation of bleeding times.[36] In hypoproteinemic horses, HES had a modest effect on COP but administering typical doses did not restore COP to normal.[37,38] In an experimental endotoxemia, the use of HES with hypertonic saline solution exerted no benefit on cardiac output or systemic vascular resistance compared with isotonic fluid resuscitation.[39] Furthermore, no significant differences in coagulation measures were noted between horses treated with HES versus isotonic fluids.[40] However, HES has been shown to adversely affect platelet function in vitro and in healthy horses.[41,42]

The use of synthetic colloids over crystalloids in human patients with sepsis has been questioned. Colloid administration has been associated with acute kidney injury, osmotic nephrosis, hemorrhage, anaphylactoid reactions, tissue accumulation, hepatic organ failure, and pruritus. Despite numerous randomized clinical trials and meta-analyses, there is conflicting evidence correlating colloid administration with increased mortality rates and increased need for renal-replacement therapy. However, the 2012 Surviving Sepsis Campaign recommended against the use of HES in sepsis.[43] As a result, the European Medicine Agency concluded that HES solutions should not be used in patients with sepsis or critically ill patients and the Food and Drug Administration placed a boxed warning on HES for increased mortality and renal-replacement therapy in 2013.

A recent study comparing the effects of HES versus commercial equine plasma on clinicopathologic values and COP in healthy horses revealed both products produced equivalent, although modest, increases in plasma COP. Meanwhile, plasma had a less profound and less prolonged dilutional effect on hematocrit, blood hemoglobin, serum total protein, and albumin concentration compared with HES.[44]

The benefits of HES over plasma include lower cost, easier storage, higher oncotic pressure, and no risk of infectious disease transmission.[44] However, equine plasma provides immunoglobulins, coagulation factors, and antithrombin. Plasma may also be less likely to result in acute kidney injury or coagulation abnormalities in critically ill patients. Although evidence from the human literature cannot be applied to equine medicine, there is a need for further evaluation of HES use in critically ill and septic horses. In the meantime, judicious use of colloids in these patients is recommended.

Antimicrobial Therapy

The advantages and disadvantages of antimicrobial administration should be considered before use in cases of acute diarrhea. In one retrospective, horses presenting with colitis that were previously treated with antimicrobials were 4.5 times less likely to survive than those not treated.[2] Antimicrobial use affects the fecal microbiota,[45,46] which is likely already disturbed.[47] Procaine penicillin, ceftiofur sodium, and trimethoprim sulfadiazine administration all impacted the fecal microbiota in healthy horses, and changes often persisted for 25 days.[46]

The identification or clinical suspicion of the presence of certain pathogens warrants targeted antimicrobial use. Oxytetracycline is considered the treatment of choice for horses with PHF.[30] Metronidazole is recommended for the treatment of C difficile–associated diarrhea (CDAD) in humans and is often used to treat horses with confirmed or presumed CDAD. However, Magdesian and colleagues[15] demonstrated an association between metronidazole administration and identification of metronidazole-resistant C difficile strains. Although metronidazole resistance in horses with CDIs is not considered clinically important at this time, these strains are

suspected to be more virulent.[14] To this effect, horses with metronidazole-resistant strains of *C difficile* had an increased risk of mortality compared with horses infected with metronidazole-susceptible strains.[15]

Antimicrobial therapy may be warranted in acute diarrhea cases demonstrating severe neutropenia and/or evidence of septic foci, such as in the lungs, liver, or jugular veins. Antimicrobial drug selection should be based on culture and susceptibility patterns of the causative agents whenever possible. However, in cases of acute undifferentiated diarrhea in horses, antibiotics may be contraindicated because of further disruption of the microbiota. Furthermore, increased shedding of *Salmonella* was associated with the administration of antimicrobials[48] and multidrug resistance in *Salmonella* spp is an emerging threat. Therefore, judicious use of systemic antimicrobials is strongly recommended.

Limiting the Effects of Endotoxemia

Efforts should be made to limit endotoxin entry into circulation, reduce the release of inflammatory mediators, neutralize circulating endotoxin, and provide supportive care.[49] NSAIDs inhibit cyclooxygenase enzymes associated with early hemodynamic responses to endotoxin. Both flunixin meglumine and ketoprofen significantly decreased the production of thromboxane B_2 production by blood monocytes in vitro.[50] Low doses of flunixin meglumine (0.25 mg/kg) suppressed thromboxane and prostaglandin production in horses challenged with endotoxin.[51] Although a dose of 1.1 mg/kg is also antiendotoxic, the lower dose may be associated with fewer side effects, especially in cases with renal insufficiency. Administration of NSAIDs to horses with endotoxemia is a mainstay of treatment. Albeit in practice, treatment is initiated following challenge with endogenous endotoxins, whereas experimentally, benefits were noted when NSAIDs were administered before endotoxin exposure. Clinicians often administer flunixin meglumine every 8 hours to control the clinical signs of endotoxemia while limiting adverse side effects, such as renal papillary necrosis and gastrointestinal ulceration.[49]

Polymyxin B is a cationic polypeptide antibiotic that forms a stable complex with the lipid A component of lipopolysaccharide. Once bound, lipopolysaccharide does not interact with equine inflammatory cells, thus preventing initiation of the proinflammatory cascade.[49,52] Initial high doses reportedly produced signs of neurotoxicity and nephrotoxicity. When administered to healthy horses at doses between 1000 and 5000 IU/kg, polymyxin B inhibited 75% of endotoxin-induced tumor necrosis factor activity[53] and no alterations in either creatinine[53] nor urine γ-glutamyltransferase-to-creatinine ratios[52] were noted. However, polymyxin B should be used with caution in hypovolemic and azotemic animals and should always be administered diluted in isotonic fluids. If side effects of neuromuscular blockage or other neurologic side effects occur, administration should be discontinued.

Hyperimmune plasma or serum, derived from horses immunized against G-bacteria and endotoxin, has the proposed benefits of neutralizing endotoxin and modulating leukocyte activation. However, there is conflicting evidence of the benefits of administration of hyperimmune plasma in experimental models of endotoxemia in horses.[49] A recent study[54] revealed that pretreatment with hyperimmune equine plasma failed to modify clinical signs of experimentally induced endotoxemia and had no impact on leukocyte activation, but reduced the bioactivity of tumor necrosis factor-α. Further studies are needed to evaluate the clinical benefits of hyperimmune plasma or serum in endotoxemic horses.

Pentoxifylline is a methylxanthine derivative that increases the deformability of red blood cells, suppresses production of proinflammatory cytokines, and can inhibit

the activation of B and T cells.[55] Experimentally, pentoxifylline administration had limited beneficial effects in endotoxemic horses[56] and slight benefits in combination therapy with flunixin meglumine.[57] Pentoxifylline is most commonly used in horses with laminitis and placentitis to increase microvascular blood flow, but there are currently no published controlled clinical trials supporting its use.

Intraluminal Intestinal Binding Agents

In theory, binding exotoxins and endotoxin within the gut lumen may decrease systemic absorption in horses with colitis. Di-tri-octaheadral smectite is a negatively charged, hydrated, aluminomagnesium that binds positively charged organic cations. Dose-dependent binding of Di-tri-octaheadral smectite with *C difficile* toxins A and B, *C perfringens* enterotoxin, and endotoxin has been demonstrated in vitro.[23,24] The recommended dose is a 1.4-kg loading dose followed by 454 g every 6 to 12 hours, necessitating frequent nasogastric intubation. Activated charcoal is a nonspecific binding agent used in many monogastric species. A recent in vitro study demonstrated no significant effects of activated charcoal on the microbial population, major metabolites produced, rate of gas production, or pH values.[58] These findings suggest that the impact of activated charcoal on the equine hindgut, and binding of toxic substances, may be minimal. Further in vivo studies are needed to elucidate the benefits of these binding agents.

Probiotics

Probiotics are discussed elsewhere in this issue. In one study of 14 horses with diarrhea, administration of *S boulardii* decreased the duration of diarrhea from 7 to 5 days, compared with control horses.[59] In contrast, there was no significant difference in return to normal manure, return to normal heart rate, appetite improvement, or any other clinical variables measured between *Saccharomyces*-treated and control horses in a randomized prospective study.[60]

The administration of *Lactobacillillus* spp and *Bifidobacterium animalis lactis* had no impact on clostridial shedding and minimal impact on the composition of the fecal microbiota in foals when administered for 3 weeks.[61] Recent literature suggests that orally administered probiotics (*Pediococcus acidilactici* and *S boulardii*) may have immunomodulatory effects in horses.[62] At this time, there is conflicting evidence regarding the clinical benefit of probiotic administration. Furthermore, label claims on veterinary probiotic preparations often contain gross inaccuracies and quality control of products is lacking.[63,64]

Additional Supportive Care Measures

Gastroprotectants such as omeprazole and H_2 antagonists, are often used prophylactically to prevent gastric ulceration in anorectic horses with acute diarrhea. Proton pump inhibitor administration has long been suspected to be a risk factor for CDAD in humans, although high-quality evidence of a cause-effect relationship is lacking.[65] Foals treated with antiulcer medication were two times more likely to develop diarrhea than those left untreated, but this was not associated with CDI.[66] The relationship between antiulcer medications, diarrhea, and CDAD in adult horses is unknown, but increasing gastric pH may eliminate a defense against *C difficile* and other pathogens.

Laminitis is commonly encountered in cases of sepsis. Currently, distal limb cryotherapy (applied from hoof to carpus for 72 hours continuously) is the only method of prevention of laminitis in horses with systemic inflammation that has been validated.[67] Other feasible laminitis prevention techniques include treating the primary disease process, anti-inflammatory therapy (NSAIDs, pentoxifylline), inhibition of

neutrophil and platelet margination (low-molecular-weight heparin, pentoxifylline, aspirin, and possibly corticosteroids), antioxidant therapy (dimethyl sulfoxide), and physical support of the sole with easing of breakover.[68]

Bacterial sepsis has the potential to lead to disseminated intravascular coagulation secondary to widespread inflammation. Salmonellosis has been associated with thrombotic events in ewes and disseminated intravascular coagulation in horses.[8] In a controlled clinical trial in humans with sepsis, heparin improved hypercoagulable states, reduced time in the intensive care unit, and decreased days on mechanical ventilation.[69] In a retrospective study of 360 horses that underwent colic surgery, the prevalence and grade of laminitis was lower in horses treated with low-molecular-weight heparin.[70] Although unfractionated heparin is often used for prophylaxis of coagulation disorders, low-molecular-weight heparin was associated with fewer side effects, such as decreased packed cell volume (PCV), catheter-site complications, and decreased platelet counts.[71] Based on limited data, heparin therapy may be indicated in as a prophylactic measure in horses with severe sepsis.

Nutritional support improves outcomes in critically ill humans and small animal patients. Horses with acute colitis or typhlocolitis are often partially or completely anorexic as a result of ileus, dysmotility, hypoperfusion, and the systemic inflammatory response. A low-bulk diet, such as a complete pelleted feed, good-quality grass hay, or alfalfa, is recommended for horses with acute colitis.[72] Offering fresh grass may stimulate the appetites of anorectic horses, although large amounts of grass may contribute to the development of laminitis or colic signs. Parenteral nutrition is beneficial in severely catabolic cases of acute colitis that do not tolerate enteral feeding. Parenteral nutrition is indicated in diarrheic patients with anorexia persisting more than 48 to 72 hours and in pregnant and lactating mares.

SUMMARY

The cause of acute, infectious, diarrhea in adult horses is often difficult to define. However, aims of therapy are universal and prompt intervention decreases morbidity and mortality. Furthermore, strong evidence to support many common therapeutic options is lacking, and this should be considered when constructing treatment plans.

REFERENCES

1. Sanchez LC. Pathophysiology of Diarrhea. In: Reed SM, Bayly WM, Sellon DC, editors. Equine internal medicine. vol. 3rd edition. St. Louis(MO): Saunders; 2010. p. 793–866.
2. Cohen ND, Woods AW. Characteristics and risk factors for failure of horses with acute diarrhea to survive: 122 cases (1990-1996). J Am Vet Med Assoc 1999; 214(3):382–90.
3. Belgrave RL, Dickey MM, Arheart KL, et al. Assessment of serum amyloid A testing of horses and its clinical application in a specialized equine practice. J Am Vet Med Assoc 2013;243(1):113–9.
4. Staempfli HR, Townsend HGG, Prescott JF. Prognostic features and clinical presentation of acute idiopathic enterocolitis in horses. Can Vet J 1991;32(2):232–7.
5. Hashimoto-Hill S, Magdesian KG, Kass PH. Serial measurement of lactate concentration in horses with acute colitis. J Vet Intern Med 2011;25(6):1414–9.
6. Weese JS, Baird JD, Poppe C, et al. Emergence of Salmonella typhimurium definitive type 104 (DT104) as an important cause of salmonellosis in horses in Ontario. Can Vet J 2001;42(10):788–92.

7. Kurowski B, Traub-dargatz JL, Morley PS, et al. Detection of *Salmonella* spp in fecal specimens by use of real-time polymerase chain reaction assay. Am J Vet Res 2002;63(9):1256–68.

8. Timoney J. Salmonella infections in horses. In: Barrow PA, Methner U, editors. Salmonella in domestic animals. 2nd edition. Oxfordshire(UK): CABI; 2013. p. 305–17.

9. Ekiri AB, MacKay RJ, Gaskin JM, et al. Epidemiologic analysis of nosocomial salmonella infections in hospitalized horses. J Am Vet Med Assoc 2009;234(1): 108–19.

10. Ernst NS, Hernandez JA, MacKay RJ, et al. Risk factors associated with fecal salmonella shedding among hospitalized horses with signs of gastrointestinal tract disease. J Am Vet Med Assoc 2004;225(2):275–81.

11. Alinovi CA, Ward MP, Couëtil LL, et al. Risk factors for fecal shedding of *Salmonella* from horses in a veterinary teaching hospital. Prev Vet Med 2003;60(4): 307–17.

12. Cohen ND, Martin LJ, Simpson RB, et al. Comparison of polymerase chain reaction and microbiological culture for detection of salmonellae in equine feces and environmental samples. Am J Vet Res 1996;57(6):780–6.

13. Schoster A, Arroyo LG, Staempfli HR, et al. Presence and molecular characterization of *Clostridium difficile* and *Clostridium perfringens* in intestinal compartments of healthy horses. BMC Vet Res 2012;8(94):1–6.

14. Schoster A, Staempfli H. Epidemiology and antimicrobial resistance in *Clostridium difficile* with special reference to the horse. Curr Clin Microbiol Rep 2016;3(1):32–41.

15. Magdesian KG, Dujowich M, Madigan JE, et al. Molecular characterization of *Clostridium difficile* isolates from horses in an intensive care unit and association of disease severity with strain type. J Am Vet Med Assoc 2006;228(5):751–5.

16. Magdesian KG, Leutenegger CM. Real-time PCR and typing of *Clostridium difficile* isolates colonizing mare-foal pairs. Vet J 2011;190(1):119–23.

17. Elmer GW, McFarland LV. Biotherapeutic agents in the treatment of infectious diarrhea. Gastroenterol Clin North Am 2001;30(3):837–54.

18. van Nood E, Vrieze A, Nieuwdorp M, et al. Duodenal infusion of donor feces for recurrent *Clostridium difficile*. N Engl J Med 2013;368(5):407–15.

19. Mullen KR, Yasuda K, Divers TJ, et al. Equine faecal microbiota transplant: current knowledge, proposed guidelines and future directions. Equine Vet Educ 2016;1–10.

20. Waters M, Raju D, Garmory HS, et al. Regulated expression of the beta2-toxin gene (cpb2) in *Clostridium perfringens* type A isolates from horses with gastrointestinal diseases. J Clin Microbiol 2005;43(8):4002–9.

21. Gohari IM, Arroyo L, MacInnes JI, et al. Characterization of *Clostridium perfringens* in the feces of adult horses and foals with acute enterocolitis. Can J Vet Res 2014;78(1):1–7.

22. Lawler JB, Hassel DM, Magnuson RJ, et al. Absorptive effects of di-tri-octahedral smectite on *Clostridium perfringens* alpha, beta, and beta-2 exotoxins and equine colostral antibodies. Am J Vet Res 2008;69(2):233–9.

23. Weese JS, Cote NM, deGannes RVG. Evaluation of in vitro properties of di-tri-octahedral smectite on clostridial toxins and growth. Equine Vet J 2003;35(7): 638–41.

24. Oue Y, Ishihara R, Edamatsu H, et al. Isolation of an equine coronavirus from adult horses with pyrogenic and enteric disease and its antigenic and genomic

characterization in comparison with the NC99 strain. Vet Microbiol 2011;150: 41–8.

25. Pusterla N, Mapes S, Wademan C, et al. Emerging outbreaks associated with equine coronavirus in adult horses. Vet Microbiol 2013;162(1):228–31.

26. Giannitti F, Diab S, Mete A, et al. Necrotizing enteritis and hyperammonemic encephalopathy associated with equine coronavirus infection in equids. Vet Pathol 2015;52(6):1148–56.

27. Xiong Q, Bekebrede H, Sharma P, et al. An ecotype of *Neorickettsia risticii* causing Potomac horse fever in Canada. Appl Environ Microbiol 2016;82(19): 6030–6.

28. Pusterla N, Johnson EM, Chae JS, et al. Digenetic trematodes, *Acanthatrium* sp. and *Lecithodendrium* sp., as vectors of *Neorickettsia risticii*, the agent of Potomac horse fever. J Helminthol 2003;77(4):335–9.

29. Mott J, Muramatsu Y, Seaton E, et al. Molecular analysis of *Neorickettsia risticii* in adult aquatic insects in Pennsylvania, in horses infected by ingestion of insects, and isolated in cell culture. J Clin Microbiol 2002;40(2):690–3.

30. Bertin FR, Reising A, Slovis NM, et al. Clinical and clinicopathological factors associated with survival in 44 horses with equine neorickettsiosis (Potomac horse fever). J Vet Intern Med 2013;27(6):1528–34.

31. Madigan JE, Rikihisa Y, Palmer JE, et al. Evidence for a high rate of false-positive results with the indirect fluorescent antibody test for *Ehrlichia risticii* antibody in horses. J Am Vet Med Assoc 1995;207(11):1448–53.

32. Mott J, Rikihisa Y, Zhang Y, et al. Comparison of PCR and culture to the indirect fluorescent-antibody test for diagnosis of Potomac horse fever. J Clin Microbiol 1997;35(9):2215–9.

33. Atwill ER, Mohammed HO. Evaluation of vaccination of horses as a strategy to control equine monocytic ehrlichiosis. J Am Vet Med Assoc 1996;208(8):1290–4.

34. Dutta SK, Vemulapalli R, Biswas B. Association of deficiency in antibody response to vaccine and heterogeneity of *Ehrlichia risticii* strains with Potomac horse fever vaccine failure in horses. J Clin Microbiol 1998;36(2):506–12.

35. Seahorn JL, Seahorn TL. Fluid therapy in horses with gastrointestinal disease. Vet Clin North Am Equine Pract 2003;19(3):665–79.

36. Jones PA, Tomasic M, Gentry PA. Oncotic, hemodilutional, and hemostatic effects of isotonic saline and hydroxyethyl starch solutions in clinically normal ponies. Am J Vet Res 1997;58(5):541–8.

37. Jones PA, Bain FT, Byars TD, et al. Effect of hydroxyethyl starch infusion on colloid oncotic pressure in hypoproteinemic horses. J Am Vet Med Assoc 2001;218(7):1130–5.

38. Bellezzo F, Kuhnmuench T, Hackett ES. The effect of colloid formulation on colloid osmotic pressure in horses with naturally occurring gastrointestinal disease. BMC Vet Res 2014;10(Suppl S8):1–5.

39. Pantaleon LG, Furr MO, McKenzie HC, et al. Cardiovascular and pulmonary effects of hetastarch plus hypertonic saline solutions during experimental endotoxemia in anesthetized horses. J Vet Intern Med 2006;20(6):1422–8.

40. Pantaleon LG, Furr MO, McKenzie HC, et al. Effects of small- and large-volume resuscitation on coagulation and electrolytes during experimental endotoxemia in anesthetized horses. J Vet Intern Med 2007;21(6):1374–9.

41. Blong AE, Epstein KL, Brainard BM. In vitro effects of three formulations of hydroxyethyl starch solutions on coagulation and platelet function in horses. Am J Vet Res 2013;74(5):712–20.

42. Epstein KL, Bergren A, Giguère S, et al. Cardiovascular, colloid osmotic pressure, and hemostatic effects of 2 formulations of hydroxyethyl starch in healthy horses. J Vet Intern Med 2014;28(1):223–33.
43. Dellinger RP, Levy MM, Rhodes A, et al. Surviving sepsis campaign: International guidelines for management of severe sepsis and septic shock, 2012. Intensive Care Med 2013;39(2):165–228.
44. McKenzie EC, Esser MM, McNitt SE, et al. Effect of infusion of equine plasma or 6% hydroxyethyl starch (600/0.75) solution on plasma colloid osmotic pressure in healthy horses. Am J Vet Res 2016;77(7):708–14.
45. Grønvold AMR, L'Abée-Lund TM, Strand E, et al. Fecal microbiota of horses in the clinical setting: potential effects of penicillin and general anesthesia. Vet Microbiol 2010;145(3–4):366–72.
46. Costa MC, Stämpfli HR, Arroyo LG, et al. Changes in the equine fecal microbiota associated with the use of systemic antimicrobial drugs. BMC Vet Res 2015; 11(19):1–12.
47. Costa MC, Arroyo LG, Allen-Vercoe E, et al. Comparison of the fecal microbiota of healthy horses and horses with colitis by high throughput sequencing of the V3-V5 region of the 16s rRNA gene. PLoS One 2012;7(7):1–12.
48. House JK, Mainar-Jaime RC, Smith BP, et al. Risk factors for nosocomial salmonella infection among hospitalized horses. J Am Vet Med Assoc 1999;214(10): 1511–6.
49. Moore JN, Barton MH. Treatment of endotoxemia. Vet Clin North Am Equine Pract 2003;19(3):681–95.
50. Jackman BR, Moore JN, Barton MH, et al. Comparison of the effects of ketoprofen and flunixin meglumine on the in vitro response of equine peripheral blood monocytes to bacterial endotoxin. Can J Vet Res 1994;58(2):138–43.
51. Semrad SD, Hardee GE, Hardee MM, et al. Low dose flunixin meglumine: effects on eicosanoid production and clinical signs induced by experimental endotoxaemia in horses. Equine Vet J 1987;19(3):201–6.
52. Morresey PR, MacKay RJ. Endotoxin-neutralizing activity of polymyxin B in blood after IV administration in horses. Am J Vet Res 2006;67(4):642–7.
53. Parviainen AK, Barton MH, Norton NN. Evaluation of polymyxin B in an ex vivo of endotoxemia in horses. Am J Vet Res 2001;62(1):72–6.
54. Forbes G, Church S, Savage CJ, et al. Effects of hyperimmune equine plasma on clinical and cellular responses in a low-dose endotoxaemia model in horses. Res Vet Sci 2012;92(1):40–4.
55. Liska DA, Akucewich LH, Marsella R, et al. Pharmacokinetics of pentoxifylline and its 5-hydroxyhexyl metabolite after oral and intravenous administration of pentoxifylline to healthy adult horses. Am J Vet Res 2006;67(9):1621–7.
56. Barton MH, Moore JN, Norton N. Effects of pentoxifylline infusion on response of horses to in vivo challenge exposure with endotoxin. Am J Vet Res 1997;58(11): 1300–7.
57. Baskett A, Barton MH, Norton N, et al. Effect of pentoxifylline, flunixin meglumine, and their combination on a model of endotoxemia in horses. Am J Vet Res 1997; 58(11):1291–9.
58. Edmunds JL, Worgan HJ, Dougal K, et al. In vitro analysis of the effect of supplementation with activated charcoal on the equine hindgut. J Equine Sci 2016; 27(2):49–55.
59. Desrochers AM, Dolente BA, Roy M-F, et al. Efficacy of *Saccharomyces boulardii* for treatment of horses with acute enterocolitis. J Am Vet Med Assoc 2005;227: 954–9.

60. Boyle AG, Magdesian KG, Durando MM, et al. *Saccharomyces boulardii* viability and efficacy in horses with antimicrobial-induced diarrhoea. Vet Rec 2013;172:128.
61. Schoster A, Staempfli HR, Abrahams M, et al. Effect of a probiotic on prevention of diarrhea and *Clostridium difficile* and *Clostridium perfringens* shedding in foals. J Vet Intern Med 2015;29(3):925–31.
62. Furr M. Orally administered *Pediococcus acidilactici* and *Saccharomyces boulardii*-based probiotics alter select equine immune function parameters. J Equine Vet Sci 2014;34(10):1156–63.
63. Weese JS. Microbiologic evaluation of commercial probiotics. J Am Vet Med Assoc 2002;220(6):794–7.
64. Scott Weese J, Martin H. Assessment of commercial probiotic bacterial contents and label accuracy. Can Vet J 2011;52(1):43–6.
65. Tleyjeh IM, Bin Abdulhak AA, Riaz M, et al. Association between proton pump inhibitor therapy and *Clostridium difficile* infection: a contemporary systematic review and meta-analysis. PLoS One 2012;7:e50836.
66. Furr M, Cohen ND, Axon JE, et al. Treatment with histamine-type 2 receptor antagonists and omeprazole increase the risk of diarrhoea in neonatal foals treated in intensive care units. Equine Vet J 2012;44(Suppl. 41):80–6.
67. Van Eps AW, Pollitt CC. Equine laminitis model: lamellar histopathology seven days after induction with oligofructose. Equine Vet J 2009;41(8):735–40.
68. Divers TJ. Clinical application of current research findings toward the prevention and treatment of acute laminitis in horses with systemic inflammatory diseases: an internist's perspective. J Equine Vet Sci 2010;30(9):517–24.
69. Liu X-L, Want X-Z, Liu X-X, et al. Low-dose heparin as treatment for early disseminated intravascular coagulation during sepsis: a prospective clinical study. Exp Ther Med 2014;7:604–8.
70. De La Rebière Pouyade G, Grulke S, Detilleux J, et al. Evaluation of low-molecular-weight heparin for the prevention of equine laminitis after colic surgery: retrospective study. J Vet Emerg Crit Care (San Antonio) 2009;19(1):113–9.
71. Feige K, Schwarzwald CC, Bombeli T. Comparison of unfractioned and low molecular weight heparin for prophylaxis of coagulopathies in 52 horses with colic: a randomised double-blind clinical trial. Equine Vet J 2003;35(5):506–13.
72. Gary Magdesian K. Nutrition for critical gastrointestinal illness: feeding horses with diarrhea or colic. Vet Clin North Am Equine Pract 2003;19(3):617–44.