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Fecal pH and Microbial Populations in Thoroughbred Horses During Transition from Pasture to Concentrate Feeding

Mariette van den Berg MSc, Simone O. Hoskin PhD, Chris W. Rogers PhD, Alex Grinberg PhD

Institute of Veterinary, Animal and Biomedical Sciences, Massey University, Palmerston North, New Zealand

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ABSTRACT

Abrupt dietary transitions and feeding of rapidly fermentable diets are common practices in the horse industry and have been associated with digestive and metabolic disorders that can impair the performance of horses. The present study investigated the effect of dietary transition from pasture grazing to confinement with concentrate feeding, and back, on fecal pH and bacterial populations of *Streptococcus* spp and *Lactobacillus* spp. Six Thoroughbred fillies, previously grazing perennial ryegrass and white clover-based pasture, were housed in individual stalls and fed an increasing ratio of concentrate to conserved forages for 13 days (days 1-13), followed by an abrupt transition back to only pasture-grazing for 3 days (days 14-16). The concentrate was initially offered at 0.83 kg dry matter (DM)/d and increased to 5 kg DM/d, whereas ensiled alfalfa was initially offered at 0.61 kg DM/d, increasing to 1.22 kg DM/d. Meadow hay was initially offered at 6.73 kg DM/d, decreasing to 1.6 kg DM/d. Fecal specimens were collected daily for determination of pH, and every 2 days for quantitative analysis of *Streptococcus* spp and *Lactobacillus* spp. Mean fecal pH increased significantly from pasture baseline values (pH 6.18) during the initial confinement and supplementation on day 1 (6.37), day 2 (6.52), day 3 (6.58), and day 4 (6.43) (standard error of mean [SEM]: 0.056; $P < .001$). By day 5, mean fecal pH values had decreased to, and remained at, baseline values until the horses returned to pasture, when another increase occurred at day 15 (6.45). Fecal colony forming units (cfu) of *Streptococcus* spp and *Lactobacillus* spp increased linearly ($r = 0.94$; $P < .001$) from 6.0 and 6.1 $\log_{10}$ cfu/g on day -4, to 7.8 $\log_{10}$ cfu/g on day 14 (SEM: 0.2 $P < .001$), respectively. Fecal cfu decreased on return to a pasture-only diet ($P < .001$). In this study, the increment of bacterial populations was associated with a relatively stable fecal pH and highlights the difficulty in identifying the effects of dietary transition on the equine hindgut health, without microbial culture.

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1. Introduction

Intensive management of horses for sport and racing is often associated with dietary transition and the feeding of rapidly fermentable carbohydrates. In New Zealand, the preparation of young horses often involves rotations of short periods (2-3 weeks) of stall confinement and concentrate feeding, followed by a return to the pasture environment [1,2]. During the period of transition from pasture to confinement and concentrate feeding, the concentrate component of the diet is commonly increased by as much as 500 g/d [3]. This is significantly higher than the 200 g/d for a 550-kg horse recommended internationally to minimize digestive and metabolic disorders [4].

The equine hindgut microflora can be highly and rapidly reactive to abrupt changes in diet, with changes in colon and cecum microflora observed as early as 5 hours after the introduction of concentrate to horses previously fed

Corresponding author at: Chris W. Rogers, PhD, Massey Equine, Institute of Veterinary, Animal and Biomedical Sciences, Massey University, Private Bag 11222, 4442 Palmerston North, New Zealand.
E-mail address: c.w.rogers@massey.ac.nz (C.W. Rogers).

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a hay-based diet [5]. Abrupt dietary changes, especially those involving pastures rapidly increasing in levels of water-soluble carbohydrates (WSC), or feeding excess levels of starch-containing grain, or cereal concentrates, are the known risk factors for the development of digestive and metabolic disorders such as hindgut acidosis and laminitis [6-8]. Equine hindgut acidosis occurs when resistant starch escapes digestion in the small intestine, or dietary starch or WSC concentrations exceed the prececal digestive and absorptive capacity. The starch or WSC are then rapidly fermented in the hindgut by anaerobic bacteria that excessively produce lactate, causing a rapid decrease in hindgut pH (hindgut acidosis) [9,10].

Various studies monitoring changes in cecal microbial populations of the equine hindgut in response to excess starch, or fructan (oligofructose), suggest that gram-positive bacteria such as *Lactobacillus* spp [6,11], *Streptococcus bovis* [6,11,12], and *Streptococcus equinus* [12] play an integral role during the onset of hindgut acidosis and laminitis. Recent advances in the identification of equine gut microbiology show that specific members of the *Streptococcus* genus, collectively described as equine hindgut streptococcal species, may be involved in the series of events that lead to lactic acidosis and subsequently to laminitis in the horse, proliferating in response to carbohydrate administration, before rapidly declining in numbers [13,14]. Even mild lactic cases of acidosis and laminitis can affect the well-being and performance potential of horses owing to subtle radiographic changes [15]. Therefore, monitoring of equine gut health can assist with the early detection of unfavorable changes and identifying those horses at risk of developing acidosis.

Acute and subacute hindgut acidosis cases are not distinctly separate disorders, thereby making diagnosis difficult. The critical hindgut pH threshold for the development of subacute or acute acidosis in horses is related to the optimal pH for lactate using bacteria to establish. An intestinal and hindgut pH of $\leq$6.0 is known to inhibit cellulolytic bacteria and protozoa and favors the rapid growth of acid-tolerant bacteria such as *Streptococcus bovis/equinus* and *Lactobacillus* spp [12,16]. A cecal pH of 6.0 in horses is considered to represent subclinical acidosis [17], and a hindgut pH of $<$6.0 is associated with clinical conditions, such as osmotic diarrhea, overgrowth of gram-positive bacteria, and lyses of gram-negative bacteria, which may culminate in laminitis in horses [6].

Direct measurement of gastrointestinal changes in pH and microbial populations requires fistulation [18], which is invasive and not practical in a nonlaboratory setting. In several species, measurement of fecal pH and microbial culture has been validated as an indirect measure of gastrointestinal changes [19-21]. In the horse, fecal pH and fecal microbial culture have been successfully used to monitor changes in the equine hindgut in response to carbohydrate challenges and evaluation of different feeding strategies [22,23].

Despite the general view linking the composition of the intestinal microflora with the development of acidosis, the microbial changes in the equine hindgut during transition from the feeding of concentrates with conserved forage to the grazing of fresh pasture, or vice versa, are poorly understood. The aim of the current study was to investigate the effect of abrupt dietary transition from pasture grazing to confinement with concentrate feeding, and back, on fecal pH and bacterial populations of *Streptococcus* spp and *Lactobacillus* spp in the hindgut of a young Thoroughbred horse.

2. Materials and Methods

This study was conducted in early spring (September) 2005 and was approved by the Massey University Animal Ethics Committee (Palmerston North, New Zealand).

2.1. Horses and Management

Six Thoroughbred fillies, with an average age of 12 (SD: 1) months and weight of 365 (SD: 27) kg, which were bred and reared on a commercial Thoroughbred stud farm (Manawatu, New Zealand), were included in the study. Before the experiment initiation, the fillies had been reared on perennial ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*)-based pasture typical of most New Zealand Thoroughbred stud farms [1,2]. The fillies were then maintained in a cohort of six horses per paddock, with an average pasture sward height of 6-7 cm (approximately 2,240 kg/dry matter [DM]/Ha), and in addition to available pasture were offered 1.9 kg DM/d concentrate (Sweetfeed, NRM Ltd, Auckland New Zealand), 1.9 kg DM/d crushed barley *(Hordeum vulgare)*, and 0.45 kg DM/d ensiled alfalfa chaff (*Medicago sativa*; FiberPro, Fiber Fresh Feeds Ltd, Reporoa, New Zealand). Fourteen days before the start of this experiment, the fillies only had access to the pasture and no supplements or concentrates were fed (washout period).

On day 0, the horses were removed from pasture, housed individually in 6 m $\times$ 6 m loose boxes with wood shavings as bedding, and gradually offered an increasing ratio of concentrate to conserved forage during a period of 13 days (days 1-13). During the 13-day treatment period, the horses were fed conventional meadow hay (ryegrass/clover pasture), ensiled alfalfa chaff (FiberPro), and a 1:1 mix of a commercially manufactured concentrate-pelleted diet (Racehorse pellets; NRM Ltd, Auckland New Zealand) and crushed barley.

Initially, the horses were offered 0.83 kg DM/d concentrate mix (day 1), which increased daily by 0.41 kg DM/d up to a maximum of 5 kg DM/d by day 11 of the trial. The FiberPro was initially offered at 0.61 kg DM/d, increasing by 0.2 kg DM/d every third day, up to a maximum of 1.22 kg DM/d by day 10. Hay was initially offered at 6.73 kg DM/d and was reduced linearly to a minimum of 1.6 kg DM/d by day 7. The horses were fed the concentrate and ensiled alfalfa as two feeds daily (08:00 and 17:00 hours), and the meadow hay was offered three times daily (8:00, 13:00, and 17:00 hours). Water was available ad libitum, and the horses were not exercised.

The amounts fed were formulated to meet the National Research Council (NRC) fiber and energy requirements for growing horses (Anonymous, 1989). After the treatment period, horses were transferred back to the pasture of similar composition, size, and available pasture DM as used for the pre-treatment period, for 3 days (post-treatment; days 14-16). The horses were not exercised.

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