Effect of maternal diet on select fecal bacteria of foals

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ABSTRACT: Adult horses depend on the microbial community in the hindgut to digest fiber and produce short-chain fatty acids that are use for energy. Colonization of the foal gastrointestinal tract is essential to develop this symbiosis. However, factors affecting colonization are not well understood. The objectives of this study were to evaluate the age-related changes and effects of maternal diet on select fecal bacterial groups in foals from 1 to 28 d of age. Thoroughbred foals (n = 18) were from dams fed forage and one of two concentrates: an oat-based (OB) or corn and wheat middlings-based (CWB) pelleted concentrate. The mares had access to assigned concentrates, along with a mixed hay and cool-season grass pasture, 28 d before and 28 d after parturition. Fecal samples were collected from foals at 1 d (14 to 36 h), 4, 14, and 28 d after birth. Fecal samples were serially diluted with phosphate-buffered saline before inoculation of enriched, selective media to enumerate *Lactobacillus* spp., amylolytic bacteria, and cellulolytic bacteria. Enumeration data were log-transformed then analyzed with mixed model analysis of variance with repeated measures (SAS 9.3) to test the main effects of maternal diet (OB or CWB), time of sample, and interaction between maternal diet and time. Cellulolytic bacteria first appeared in foal feces between 4 and 14 d of age and increased with age ($P < 0.05$). Amylolytic bacteria and lactobacilli were abundant at 1 d and then increased with age ($P < 0.05$). There was an interaction between maternal diet and time for *Lactobacillus* spp. with OB foals having more lactobacilli than CWB foals at 1 and 4 d ($P < 0.05$); however, there were no differences observed at 14 d ($P > 0.05$). Maternal diet did not influence amylolytic or cellulolytic bacteria ($P > 0.05$). These results indicate that colonization of the hindgut is a sequential process beginning early in the foal's life and that maternal diet may influence some bacteria in the gastrointestinal tract of foals.

Key words: horse, microbial colonization, starch source

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INTRODUCTION

Horses rely on the microbial community of the gastrointestinal tract (GIT) to digest structural carbohydrates and produce short-chain fatty acids that they use as energy. At birth, very few or no bacteria are detected in the foal's GIT; thus, foals are lacking the microbial community needed to ferment carbohydrates (Earing et al., 2012; Faubladier et al., 2013). Cellulolytic bacteria that are important in the GIT of mature horses are not detected at birth and appear to be slow to colonize (Julliand et al., 1996; Faubladier et al., 2013; Rey et al., 2014). Amylolytic bacteria, including lactobacilli, have been detected in foal feces as early as 1 d of age (John et al., 2015). Dietary composition, such as the amount and botanical source of starch, affects the microbiota in adult horses (Potter et al., 1992; Harlow et al., 2016). However, little is known of the factors influencing microbes in the foal's GIT. Earing et al. (2012) found that mare-to-foal microbial similarity was low at birth then increased with maximum similarity at 6 wk. Because foals were more similar to their dam than to other foals, it is possible that maternal factors, including diet, could influence colonization in the foal's GIT. The objectives of the current study were to 1) evaluate changes in fecal cellulolytic bacteria, amylolytic bacteria, and *Lactobacillus* spp. in foals from 1 to 28 d of age, and 2) determine the effect of starch source in the maternal diet on select fecal bacteria of foals. We hypothesized that fecal bacteria would change with age in foals from 1 to 28 d of age and that these changes would be influenced by starch sources in the maternal diet.

MATERIALS AND METHODS

All procedures were approved by the Institutional Animal Care and Use Committee at the University of Kentucky.

Animals

Eighteen thoroughbred foals from mares with a mean age of 13 yr (range 6 to 19 yr) were used in this study at the University of Kentucky's Maine Chance Farm, Lexington, KY. Foals were born between March 4, 2015, and May 21, 2015. Mean body weight (BW) of mares was 632.3 kg (range 551.0 to 721.0 kg) when mares were assigned to a treatment. Body condition score of pre-foaling mares ranged from 4.0 to 6.0 using a 9-point scale (Henneke et al., 1983). Before the beginning of the study, the pregnant mares were maintained on pastures consisting of cool-season grasses with free access to automatic waterers. The mares were fed approximately 4 kg of a commercial pelleted concentrate (McCauley Bros., Inc., Versailles, KY) daily in two equal meals and had ad libitum access to a grass and alfalfa-mixed hay.

Diets and Treatment Assignments

Pregnant mares were paired by last breeding date and then randomly assigned to one of two treatments: an oat-based (OB) or a corn and wheat middlings-based (CWB) pelleted concentrate. The two pelleted concentrates were custom-formulated (McCauley Bros., Inc.) so that the OB concentrate contained no corn or wheat middlings and the CWB concentrate contained no oats. The two concentrates were similar in all nutrients (Table 1). Mixed grass and alfalfa hay were available ad libitum to the mares and foals throughout the study along with pasture. The two mares with the earliest due dates began adaptation at 333 d of gestation. All other mares were adapted to their assigned diet when they reached 310 d of gestation. Before parturition, each mare received 3.2 kg (dry matter, DM) daily of the assigned concentrate that was divided into two meals fed at 0645 and 1530. After parturition the daily intake of concentrate was gradually increased to 4.8 kg (DM) per horse, fed in three meals.

Housing and Feeding Management

Pregnant mares were housed in groups of two to four in paddocks. When parturition was imminent, the mares stayed in box stalls at night to facilitate observation of foaling. After parturition, foals remained in box stalls (4.5 m × 4.5 m) with their dam for 1 to 3 d, moved into individual small round pens, and then rotated through progressively larger paddocks. Mare and foal pairs were comingled with other mare and foal pairs in outdoor paddocks approximately 1 wk after birth. Assigned concentrates were fed individually using various methods. Before parturition mares were fed assigned concentrate in box stalls or using nose bags. After parturition mares and foals were kept in box stalls before moving into individual pens where hanging fence feeders were used for concentrate meals. Foals had access to their dam's assigned concentrate as well as hay and pasture. When mare and foal pairs were comingled, the mares and foals were closely monitored during meals to prevent consumption of any nonassigned concentrate. Foals were weighed within a few hours of birth and then weekly. Mare BW was recorded pre-foaling and within 1 wk postpartum.

Table 1. Ingredients and chemical composition of the oat-based (OB) and a corn and wheat middlings-based (CWB) pelleted concentrates (dry matter basis)

Item	OB	CWB
Ingredient, %		
Ground oats	74.00	-
Ground corn	-	35.00
Wheat middlings	-	34.75
Soybean meal (48%)	12.50	9.70
Soybean hulls	-	8.00
Molasses with fat blend	6.90	6.93
Alfalfa meal	2.50	2.50
Dicalcium phosphate	1.60	0.55
Calcium carbonate	0.95	1.75
Soybean oil	0.75	-
White salt	0.50	0.50
Pres Toxi-Chek[1]	0.10	0.10
McCauley Trace Mineral Premix[2]	0.10	0.10
McCauley Vitamin Premix[3]	0.10	0.10
L-lysine	-	0.02
Chemical composition		
Dry matter, %[4]	88.50	88.70
Digestible energy, Mcal/kg[5]	3.43	3.41
Crude protein, %[4]	15.80	15.40
Acid detergent fiber, %[4]	11.55	12.05
Neutral detergent fiber, %[4]	21.10	23.45
Starch, %[4]	38.00	36.15
Nonstructural carbohydrates, %[6]	44.55	45.00
Crude fat, %[4]	5.90	4.10
Calcium, %[4]	1.37	1.27
Phosphorus, %[4]	0.85	0.74
Magnesium, %[4]	0.24	0.30

[1]Mold inhibitor additive (Lucta USA Inc., Northbrook, IL).

[2]Provides 26.25 ppm Cu, 60 ppm Mn, 0.4 ppm Se, and 75 ppm Zn.

[3]Provides 9.75 IU/kg vitamin A, 1.76 IU/kg vitamin D_3, 233.91 IU/kg vitamin E, 0.10 mg/kg biotin, 40.77 mg/kg pantothenic acid, 1.90 mg/kg vitamin K, 9.71 mg/kg thiamine, 61.06 mg/kg niacin, 6.54 mg/kg riboflavin, 2.89 mg/kg folic acid, 6.77 mg/kg pyridoxine, and 0.03 mg/kg vitamin B_{12}.

[4]Analyses performed by Dairy One (Ithaca, NY) presented as means (n = 2) on a DM basis.

[5]Estimated with the following equation (NRC, 2007): Digestible energy (Mcal/kg) = 4.07 − 0.055 × (% acid detergent fiber).

[6]Nonstructural carbohydrates calculated by adding % starch and % water-soluble carbohydrates.

Fecal Samples

Fecal samples were collected from foals at 1, 4, 14, and 28 d after birth. Foals were continuously monitored in box stalls or in outdoor paddocks with their dam to collect fecal samples during defecation by catch and the time of collection was recorded. To avoid bacterial contamination of samples, sterile gloves and 100 mL sterile specimen cups were used to collect fecal samples from foals. Samples were placed in a prewarmed (37 °C) insulated cooler, homogenized in the sterile specimen cup, and a subsample of approximately 1 g was placed in a sterile, prewarmed, and preweighed Hungate tube. The tube was then purged of air with CO_2 and then transported to the laboratory in an insulated cooler (37 °C) for analysis.

Bacterial Enumerations

Upon arrival at the laboratory, the fecal samples were reweighed and then diluted 1:10 (w/w) with anaerobic phosphate-buffered saline (PBS) using anaerobic technique. The samples were homogenized by vortex and serially diluted (10^{-1} to 10^{-10}) with PBS before inoculation of enriched and selective media. Selective media were used to enumerate cellulolytic bacteria, *Lactobacillus* spp., and amylolytic bacteria.

Cellulolytic bacteria were enumerated as described by Harlow et al. (2015b) by inoculating 9 mL of anaerobic defined liquid medium with 1 mL from each serial dilution (10^{-1} to 10^{-10}). The medium for enumeration of cellulolytic bacteria contained a strip of filter paper (4 g filter paper per liter of media) as the growth substrate. After 10 d of incubation (37 °C), the dissolution of cellulose was visually evaluated and the highest dilution with dissolution of cellulose was recorded.

Amylolytic bacteria were enumerated by inoculating anaerobic medium (9 mL) containing starch as the substrate (4 g soluble starch per liter of media) with 1 mL from each serial dilution (10^{-1} to 10^{-10}). The inoculated media were incubated for 3 d at 37 °C then visually evaluated for bacterial growth. The highest dilution exhibiting bacterial growth was recorded.

Enumeration of *Lactobacillus* spp. was accomplished by inoculating Rogosa SL Agar (Rogosa et al., 1951) (BD, Franklin Lake, NJ) with 0.20 mL from each serial dilution (10^{-1} to 10^{-7}) with a sterile spreader. The inoculated media were incubated aerobically at 37 °C for 3 d, then colonies were counted. Plates containing 20 to 200 colonies were counted and the highest dilution counted was recorded.

Statistical Analysis

Enumeration data were $\log_{10}$-transformed before statistical analysis. Data were analyzed using the MIXED procedure of SAS 9.3 (SAS Institute Inc., Cary, NC). Mixed model analysis of variance with repeated measures design was used to test the main effects of sample day, maternal diet (OB or

CWB), and the interaction between sample day and maternal diet. When there was a significant main effect or interaction ($P < 0.05$), least squares means were separated using least significant difference tests. Regression analysis was used to estimate foal average daily gain (ADG). The effects of maternal diet on foal ADG, foal birth weight, and birth weight as a percentage of mare BW were compared using Student's *t*-test (SAS 9.3). Results were considered significant when $P < 0.05$ and a trend was recognized when $P < 0.10$.

RESULTS AND DISCUSSION

Initially, 10 mares were assigned to each diet (OB and CWB) with the intention of obtaining fecal samples from 10 foals per diet at 1, 4, 14, and 28 after birth. Owing to logistical limitations, including the irregularity of defecation in newborn foals, fecal samples were not collected from all foals on all sampling days. For foals born between 2200 and 0600, it was not feasible to collect the 1 d samples at precisely 24 h after birth. The mean time for the 1 d sample was 27 h after birth with a range of 14 to 36 h after birth.

Foal BW and ADG

Starch source in the maternal diet did not influence prepartum ($P = 0.1598$) or postpartum mare BW ($P = 0.1896$; Table 2). There was no effect of maternal diet on birth weight of foals ($P = 0.2164$). Birth weight of individual foals ranged from 47 to 72 kg across all treatments with a mean of 56.2 ± 1.8 and 59.3 ± 1.9 kg for OB and CWB foals, respectively. Foal birth weight as a percentage of mare post-foaling weight was not influenced by maternal diet ($P = 0.9327$; Table 2). The mean birth weight as a percentage of mare post-foaling weight was 10.3 ± 0.3% and 10.2 ± 0.3% for OB and CWB foals, respectively. Foal birth weight as a percentage of mare BW is estimated to be 9.7% (NRC, 2007), which was consistent with the results of this study. Foal ADG from birth to 28 d was similar for OB and CWB ($P = 0.8634$, 1.77 and 1.66 kg/d, respectively). Inclusion of different starch sources in pelleted concentrates fed to mares did not appear to alter BW of mares pre- or postpartum, foal birth weights, or foal ADG. These results suggest that the two concentrates provided similar amounts of digestible nutrients.

Table 2. Effect of treatment on BWs of foals from mares fed either an oat-based (OB) or a corn and wheat middlings-based (CWB) pelleted concentrate[1]

	OB	CWB	*P* value
Prepartum mare BW, kg	633.6 (17.1)	670.1 (18.0)	0.1598
Postpartum mare BW, kg	553.6 (16.7)	579.3 (15.8)	0.1896
Foal birth BW, kg	56.2 (1.8)	59.3 (1.9)	0.2164
Foal birth wt, as % of postpartum mare BW	10.3 (0.3)	10.2 (0.3)	0.9327
Foal ADG[2], kg/d	1.77 (0.1)	1.66 (0.1)	0.8634

[1]Data presented as least squares means (SEM).

[2]Calculated using regression.

Effect of Foal Age on Fecal Bacteria

There were no detectible cellulolytic bacteria in fecal samples of foals at 1 d of age. The absence of detectible cellulolytic bacteria 1 d after birth is similar to previous work in foals (Julliand et al., 1996; Faubladier et al., 2013). Cellulolytic bacteria first appeared in fecal samples of foals at 4 or 14 d of age. Only one foal had detectable cellulolytic bacteria in the feces at 4 d of age; however by 14 d of age, all but one foal had detectible cellulolytic bacteria. Faubladier et al. (2013) reported that low numbers of cellulolytic bacteria were detected in the feces of foals 2 d after birth but it was not clear whether cellulolytic bacteria were found in all foals at 2 d. Similarly in ruminants, cellulolytic bacteria have been detected within the first week after birth (Anderson et al., 1987; Fonty et al., 1987); however, it is unknown if all of the calves and lambs in those studies had detectable cellulolytic bacteria. Milk is the primary nutrient source in the neonatal period and a minimal amount of forage is consumed. Therefore, the need for cellulolytic bacteria is low initially, but cellulolytics will be needed once the foal starts to consume forages that contain structural carbohydrates. Actual intake of forage by individual foals was not measured in this study but would be expected to be minimal the first week after birth, then increase with age (Crowell-Davis et al., 1985). Cellulolytic bacteria appeared to colonize between 4 and 14 d of age, concurrent with the assumed increase in consumption of forage and other solid feeds.

The number of cellulolytic bacteria continuously increased with age in foals ($P < 0.05$, Figure 1). Cellulolytic bacteria from fecal samples of individual foals at 28 d of age ranged between 10^4 to 10^6 colony-forming units (cfu)/g of feces with a least squares mean of 3.2×10^4 cfu/g of feces across all treatments (Figure 1). Previous work in foals (Julliand et al., 1996; Faubladier et al., 2013) and ruminants (Anderson et al., 1987; Rey et al., 2014; Guzman et al., 2015) have also observed a

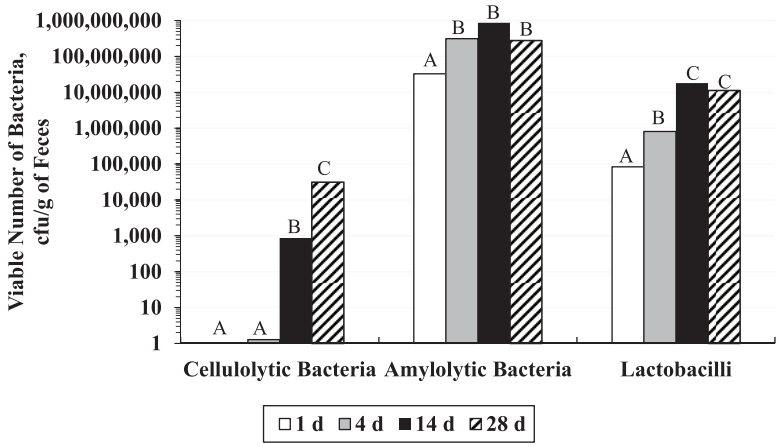

Figure 1. Effect of time on fecal bacterial groups of foals. Data presented as back-transformed least squares means. cfu = colony-forming units or viable cell number. Letters A, B, and C represent differences between time points within bacterial group ($P < 0.05$). There was an effect of time on cellulolytic bacteria ($P < 0.0001$), amylolytic bacteria ($P = 0.0028$), and lactobacilli ($P < 0.0001$). Pooled SEM are $\log_{10}$-transformed: cellulolytics 0.187, amylolytics 0.252, and lactobacilli 0.205.

continual increase in cellulolytic bacteria with age. Faubladier et al. (2013) found that the number of cellulolytic bacteria in foals reached adult levels by 1 mo of age whereas Julliand et al. (1996) reported that foals reached adult values by 2 mo of age. Cellulolytic bacteria in the feces of adult horses have been reported between 10^5 and 10^7 cfu/g of feces (Muhonen et al., 2009; Harlow et al., 2016). In this study, the average number of cellulolytic bacteria in foal feces was less than previously reported in adult horses, but at least one foal in this study had reached adult values by 28 d of age.

Cellulolytic bacteria were not detected in the feces of 1 d old foals; however, there was an abundance of fecal amylolytic bacteria at 1 d of age. Amylolytic bacteria were detected in all fecal samples from foals at 1 d of age and ranged between 10^2 and 10^9 cfu/g of feces in individual foals. The average number of amylolytic bacteria in foal feces increased from 1 to 4 d of age ($P < 0.05$), then remained stable from 4 to 28 d of age ($P > 0.05$; Figure 1). Similarly, Faubladier et al. (2013) reported an increase in amylolytic bacteria in foal feces from 1 to 3 d after birth and little variation from 3 to 60 d of age. We have previously observed amylolytic bacteria in adult horses between 10^6 and 10^8 cfu/g of feces (Harlow et al., 2016). Most of the foals in this study had already reached adult values by 1 d of age. One foal at 1 d of age had relatively few detectible amylolytic bacteria (10^2 cfu/g of feces) and no detectable lactobacilli. The fecal sample from this foal was collected 15 h after birth and the average time of sample collected from all the foals was 26 h after birth. Because of the relatively early sample time, the fecal sample may have been more similar to meconium with low abundance of microorganisms. By 4 d of age, 50% of foals had more fecal amylolytic bacteria than adult values (>10^8 cfu/g of feces), which increased to 88% of foals at 14 d of age. Lactobacilli, which are a type of amylolytic bacteria, were also detected in fecal samples from some individual foals at 1 d and ranged from 0 to 8.3×10^5 cfu/g of feces. Fecal lactobacilli in foals increased from 1 to 14 d of age ($P < 0.05$) then remained stable to 28 d of age ($P > 0.05$; Figure 1).

The first microorganisms to colonize an environment are termed "pioneer species." We found in this study that some bacteria, such as amylolytics and lactobacilli, colonize the GIT of the newborn foal rapidly after birth reaching adult values by 1 d of age then exceeding adult values. Many bacteria in this functional group are facultative anaerobes with the ability to survive in the presence of oxygen, enabling them to survive in the surrounding environment and colonize the GIT of the newborn foal, as seen in other species (Anderson et al., 1987; Agarwal et al., 2002; Rey et al., 2014). Lactobacilli have been identified as early colonizers in the GIT of human infants (Sela and Mills, 2010). Furthermore, anaerobic and facultative anaerobic bacteria are thought to be pioneer species in ruminants; specifically *Streptococcus* spp. and *Escherichia coli* have been identified as predominant bacteria in newborn calves (Fonty et al., 1987; Minato et al., 1992).

In contrast to amylolytic bacteria, cellulolytic bacteria were slower to colonize the foal's GIT in this study. Cellulolytic bacteria were first detected in foal feces between 4 and 14 d of age and did not reach adult values by 28 d of age. Likewise in a previous study, foal amylolytic bacteria were greater than adult values soon after birth whereas cellulolytic bacteria were similar to adult levels by 1 mo of age (Faubladier et al., 2013). In ruminants,

cellulolytic bacteria do not appear to be pioneer species of the GIT, but once colonized, cellulolytic bacteria increase with age (Minato et al., 1992).

Together with previous studies, the current observations indicate a change in microbial community as the foal ages. The microbial community in the GIT of human infants also increases in complexity with age and resembles that of adults by 12 mo of age (Stark and Lee, 1982; Koenig et al., 2011). Colonization of microbes in the GIT of foals appears to be a sequential process, as seen in other species (Kim et al., 2011). The diet of newborn foals consists primarily of milk, which does not contain starch; therefore, the amylolytic bacteria detected 1 d after birth most likely use substrates other than starch, such as lactose and other oligosaccharides (Faubladier et al., 2013). Further research is required to fully understand the role of amylolytic bacteria in the GIT of neonatal foals.

Effect of Maternal Diet on Foal Fecal Bacteria

The microbial community in the GIT changes over time in foals. Previous work has found greater similarity of the microbiota between mare and foal pairs than between individual foals (Earing et al., 2012). In adult horses, dietary starch sources (oats, corn, barley, etc.) have differential effects on the fecal microbial community (Harlow et al., 2015a, 2016). Faubladier et al. (2013) found changes to the microbial community of foals when their dams were fed a fermented feed product. Because exposure to maternal feces is one route of inoculation of the foal's GIT, it was of interest to determine whether starch source in the maternal diet would affect the microbial community in the foal's GIT.

There was no main effect of starch source in maternal diet on fecal cellulolytic bacteria in foals (P = 0.3768; Table 3) and there was no treatment by time interaction for foal cellulolytic bacteria (P = 0.2482; Table 3). Likewise, there was no main effect of starch source in maternal diet on fecal amylolytic bacteria in foals (P = 0.1272). However, there was a trend for a treatment by time interaction for amylolytic bacteria (P = 0.0611; Table 3). At 1 d of age, foals from mares fed the OB concentrate had more fecal amylolytic bacteria than foals from mares fed CWB concentrate ($P < 0.05$), and there was a trend for OB foals to have more amylolytics than CWB foals at 4 d ($P < 0.10$; Table 3). Differences between treatments for foal fecal amylolytic bacteria were no longer present at 14 and 28 d of age ($P > 0.05$). Similar to fecal amylolytic bacteria, starch source in maternal diet had no main effect on fecal lactobacilli in foals (P = 0.1093) but there was a treatment by time interaction (P = 0.0368; Table 3). There were more lactobacilli in OB foals compared to CWB foals at 1 and 4 d ($P < 0.05$), but the differences were no longer present at 14 and 28 d of age ($P > 0.05$; Table 3).

Foals were not observed eating concentrate during the first week of life when treatment differences in the number of fecal amylolytics and lactobacilli were present. Therefore, we cannot attribute the treatment differences in fecal amylolytic bacteria and lactobacilli in foals to the direct consumption

Table 3. Main effects of treatment, time, and treatment by time interaction on fecal bacterial groups of foals from mares fed an oat-based (OB) or a corn and wheat middlings-based (CWB) pelleted concentrate[1]

	Sample				*P* value		
	1 d	4 d	14 d	28 d			
n (OB)	7	7	8	9			
n (CWB)	9	9	9	9	Diet	Time	Diet × time
Amylolytic bacteria, cfu/g of feces (SEM)[2]							
OB	$1.4 \times 10^{8, a}$ (0.388)	$9.7 \times 10^{8, c}$ (0.388)	7.6×10^{8} (0.363)	1.7×10^{8} (0.342)	0.1272	0.0028	0.0611
CWB	$7.7 \times 10^{6, b}$ (0.342)	$1.0 \times 10^{8, d}$ (0.342)	1.0×10^{9} (0.342)	4.6×10^{8} (0.342)			
Lactobacilli, cfu/g of feces (SEM)							
OB	$3.2 \times 10^{5, a}$ (0.314)	$2.2 \times 10^{6, a}$ (0.313)	1.4×10^{7} (0.293)	8.4×10^{6} (0.277)	0.1093	<0.0001	0.0368
CWB	$2.2 \times 10^{4, b}$ (0.277)	$2.9 \times 10^{5, b}$ (0.277)	2.4×10^{7} (0.277)	1.5×10^{7} (0.277)			
Cellulolytic bacteria, cfu/g of feces (SEM)							
OB	0	0	977 (0.269)	77,428 (0.254)	0.3768	<0.0001	0.2482
CWB[3]	0	2 (0.254)	774 (0.254)	12,915 (0.254)			

[1]Data presented as back-transformed least squares means (SEM). SEM are $\log_{10}$-transformed.

[2]cfu = colony forming units or viable cell number. Means with a superscript [a, b] indicate differences between treatments within sample time and bacterial group ($P < 0.05$). Means with a superscript [c, d] indicate differences between treatments within sample time and bacterial group ($P < 0.10$).

[3]One foal at 4 d had 100 cfu/g of feces of cellulolytics, the rest had 0 cfu/g of feces at 4 d.

of concentrates by the foals. Faubladier et al. (2013) also found that foal fecal bacteria may be influenced by maternal diet soon after birth, but the effects were transient.

The proportion of cereal grains in the diet has been shown to influence the composition of mare's milk; protein and fat content decreases with an increase in the proportion of concentrate (Doreau et al., 1992). Furthermore, the composition of concentrate also influences milk composition. Hoffman et al. (1998) found that mares fed a concentrate high in fat and fiber had greater protein content in colostrum whereas mares fed a concentrate high in sugar and starch had greater lactose content in colostrum. However, there is little information on the influence of dietary starch source on mares' milk composition. It is known that different starch sources (oats, corn, barley, etc.) vary in their susceptibility to enzymatic digestion in the foregut of adult horses (Potter et al., 1992; Meyer et al., 1995; de Fombelle et al., 2004). Oat starch is thought to have greater foregut digestibility than corn starch. It is possible in the current study that foregut digestion of the OB concentrate fed to mares yielded more simple sugars available for absorption by the small intestine than the CWB concentrate and that difference in glucose availability to the mammary gland affected milk composition.

The OB foals had more fecal lactobacilli and amylolytics within the first 4 d after birth but differences were no longer present by 14 d of age. Perhaps the greater number of these bacteria in the feces of foals from dams fed the OB diet reflected a greater amount of available substrate in milk (such as carbohydrates) to support microbial growth. Further research is needed to fully understand the influence of milk composition on the development of the GIT of the foal.

CONCLUSIONS

It is evident from our results that colonization of the hindgut is a sequential process beginning early in the foal's life. There appears to be an abundance of amylolytic bacteria in the GIT of the foal soon after birth, whereas cellulolytic bacteria are slower to colonize. Maternal diet had transient effects on foal fecal bacteria; however, further research is required to fully understand the influence of maternal diet on the process of microbial colonization of the GIT of the foal. Understanding the GIT colonization process in the foal may enable researchers to develop strategies to improve nutrient utilization and minimize gastrointestinal disease.

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