EFFECT OF DIET ON CECAL PH AND FEEDING BEHAVIOR OF HORSES[1,2]

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SUMMARY

Three cecal-fistulated horses were used in a 3 × 3 latin square experiment to determine the influence of diet and of cecal infusions of Na_2CO_3 on cecal fermentation and feeding behavior. The three treatments were hay, concentrate and concentrate plus hourly infusions of Na_2CO_3. Cecal fluid samples and cecal pH readings were taken at zero through 11 hr following feeding at the end of each experimental period, and animal activity was measured by the use of a movie camera set to take 5 sec of film every 5 minutes. Cecal pH was significantly lower at 4, 5 and 6 hr following feeding for the horses receiving the concentrate diet than for those fed hay. The concentrate-fed horses had a significantly lower percentage of cecal acetate and higher cecal propionate than those fed hay, while cecal butyrate was variable for horses receiving both diets. Cecal lactic acid was lower for the horses fed the hay diet than for those fed concentrate but the data were variable.

Infusions of Na_2CO_3 significantly increased cecal pH at 3, 4 and 5 hr post-feeding, compared with that of horses fed only the concentrate diet. Horses receiving the Na_2CO_3 infusion had higher cecal acetate and lower propionate at 1, 3 and 4 hr following feeding than those fed only the all-concentrate diet.

The horses fed the concentrate diet spent significantly more time chewing wood and in coprophagy than did those fed hay. Infusions appeared to reduce the time spent in these activities by the concentrate-fed horses, however the differences were not significant. The amount of time spent chewing wood was found to be significantly correlated with cecal propionate.

(**Key Words:** Horses, Appetite Depravity, Wood Chewing, Volatile Fatty Acids, Cecal pH.)

INTRODUCTION

Feeding completely pelleted diets to horses has become an important economic consideration. Earle (1950) noted several advantages for pelleted diets over separate allowances of hay and grain, including economy of labor, ease of transportation and storage, and convenience in feeding.

Haenlein *et al.* (1966) reported that feeding pelleted hay, or a completely pelleted diet, to ponies caused them to become extremely nervous and to chew wood. In previous work at the University of Kentucky, it was noted that young horses receiving a completely pelleted diet chewed wood and also chewed the manes and tails of other horses.

Briggs *et al.* (1957) reported a relationship among pH, VFA and buffering capacity in sheep fed different diets. Thus, it was of interest to investigate factors which might alter cecal fermentation and feeding behavior in the horse.

EXPERIMENTAL PROCEDURE

Three cecal-fistulated mature Quarter Horse geldings (avg wt 450 kg) were used in a 3 × 3 latin square experiment. The dietary treatments were hay, concentrate and concentrate plus cecal infusions of Na_2CO_3. The horses were fed either 8 kg of mixed grass-legume hay or 6 kg of a sweet feed mixture (table 1) at 12-hr intervals, with trace mineral salt and water offered free-choice. All horses were maintained in metal stalls with concrete floors on which rubber mats were installed to substitute for bedding.

Each experimental period lasted 13 days. The first 3 days of each experimental period were allowed for the horses to adjust to dietary

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TABLE 1. COMPOSITION OF CONCENTRATE DIET

Ingredient	Percentage
Corn (4-02-931)	35
Oats (4-02-313)	35
Wheat bran (4-05-190)	10
Soybean meal (44%) (5-04-604)	10
Molasses (4-04-696)	8
Dicalcium phosphate (6-01-080)	1
Ground limestone (6-02-632)	1
Trace mineral salt	+

changes and on the fourth day a 1" x 8" x 5' white pine board was attached to the front of each stall. Each board was weighed before it was attached to the stall and the final weight taken immediately after it was removed. Motion pictures of the horses were taken on the sixth and seventh days of each experimental period to record the individual activities of the horses before the cecal infusions began. On the eighth day, hourly cecal infusions of 62.5 ml of a 20% w/v solution of Na_2CO_3 was administered to one of the horses receiving the concentrate diet, and the other two horses received sham infusions of water, and the hourly infusions were continued until the end of the experimental period. Motion pictures again were taken on the 11th and 12th days, and, cecal pH readings were made and cecal fluid samples were collected at zero through 11 hr following feeding on the last day of each period. All horses were exercised daily for 1 hour.

The photographic studies of the activities of the horses on each treatment were accomplished by the use of an electrically-timed 8 mm movie camera set to take 5 sec of film at 5-min intervals. The camera was fitted with a wide angle lens to allow photographing the three horses simultaneously. The film was analyzed by noting the activity, or activities, in which a horse was engaged during a given 5-sec interval photographed and the times for each activity totaled for a given hour so that 1 min of shooting time represented approximately 1 hr actual time.

Hourly cecal fluid pH readings were taken with a combination electrode which was covered with a protective shield to allow it to be inserted into the ceca.

The cecal fluid samples collected were deproteinized with metaphosphoric acid, centrifuged and the supernatant frozen for later analysis. Cecal fluid samples were analyzed for VFA content by gas-liquid chromatography and for lactic acid content by an automated enzymatic reaction procedure (Hochella and Weinhouse, 1965) using Technicon auto analysis equipment.

Statistical significance of the cecal VFA, lactic acid and animal activity data was determined by analysis of variance according to methods described by Snedecor and Cochran (1967).

TABLE 2. COMPARISON OF MEAN CECAL pH FOLLOWING FEEDING

No. of hours	Hay	*vs*	Conc	No. of hours	Na_2CO_3 treated	*vs*	Conc
0	7.14		7.22	0	7.60		7.22
1	7.16		7.24	1	7.76		7.24
2	7.04		7.14	2	7.40		7.14
3	7.01		6.77	3	7.26[e]		6.77[f]
4	6.92[a]		6.43[b]	4	7.15[a]		6.43[b]
5	6.89[c]		6.27[d]	5	6.76[e]		6.27[f]
6	6.87[e]		6.12[f]	6	6.82		6.12
7	6.75		6.19	7	6.86		6.19
8	6.83		6.41	8	6.98		6.41
9	6.96		6.44	9	7.07		6.44
10	6.92		6.64	10	7.11		6.64
11	7.12		6.82	11	7.13		6.82

[a]Significantly different ($P<.01$) from b within comparisons.

[c]Significantly different ($P<.05$) from d within comparisons.

[e]Significantly different ($P<.10$) from f within comparisons.

RESULTS AND DISCUSSION

Mean cecal pH values (table 2) were significantly lower at 4, 5 and 6 hr after feeding for the horses fed the concentrate diet than for those fed the hay diet. Alexander *et al.* (1952) reported a range in pH values of 4.5 to 7.0 in the colon contents of ponies when sugar and cellobiose were fermented. Decreasing rumen pH following the addition of concentrates to ruminant diets has been shown by Luther and Trenkle (1967), Clanton and Woods (1966) and Moore (1964). It was suggested by Moore (1964) that the increased soluble carbohydrates produced a possible increase in VFA production and a consequent lowering of rumen pH. The horses receiving the Na_2CO_3 infusions (table 2) appeared to have a consistently higher cecal pH than the horses fed the concentrate diet following feeding, but a significant difference was noted at only 3, 4 and 5 hr after feeding. Kromann and Meyer (1972) and Raun *et al.* (1962) reported an increase in rumen pH due to the addition of $NaHCO_3$ to high concentrate diets, however similar additions to the drinking water of sheep did not appear to have an effect (Loggins *et al.*, 1968). These horse data suggest that a portion of the soluble carbohydrates from the concentrate diet did reach the cecum with a consequent lowering of cecal pH, and that pH also could be increased by infusions as reported for ruminants.

Mean cecal acetate values (Molar %) for the horses receiving the hay diet were consistently higher than for those fed the concentrate diet following feeding, and these differences were significant at all times except for the second and 9th hr after feeding (table 3). Mean cecal propionate followed a reciprocal pattern, with the differences significant at all times following feeding except for the second, 9th and 11th hours. Similar trends in cecal acetate and propionate were noted by Hintz *et al.* (1971) and Stillions *et al.* (1970), and the results are consistent with reported rumen data. Hungate (1966) explained that differences in molar percentages of VFA for cattle fed either hay or concentrate diets were due to alterations in the microbial population. He stated that increased activity of *Rumenococcus albus* produces more acetate in the rumen of hay-fed cattle, whereas increased activities of *Streptococcus bovis* and *Bacteroides amylophilus* produce increased propionate and succinate, respectively, in the rumen of grain-fed cattle. Similar bacteria were isolated by Kern *et al.* (1973) from the ceca of ponies fed hay and oats.

The horses receiving the Na_2CO_3 infusions were found to have a significantly higher percentage of cecal acetate and a significantly lower percentage of propionate at 1, 3 and 5 hr following feeding than those fed the concentrate diet alone (table 4). Kromann and Meyer (1972) and Miller *et al.* (1965) found increased

TABLE 3. COMPARISON OF POST-FEEDING CECAL ACETATE AND PROPIONATE (MOLAR %)-HAY AND CONCENTRATE TREATMENTS

No. of hours	Acetate: Hay	*vs*	Acetate: Conc	No. of hours	Propionate: Hay	*vs*	Propionate: Conc
0	79.72[c]		68.96[d]	0	14.88[c]		23.17[d]
1	78.89[c]		69.30[d]	1	16.25[c]		23.42[d]
2	78.83		73.16	2	16.47		21.61
3	79.18[c]		64.59[d]	3	16.14[a]		26.83[b]
4	80.15[e]		68.81[f]	4	15.72[e]		25.45[f]
5	80.27[a]		64.61[b]	5	15.18[a]		30.28[b]
6	80.06[c]		63.55[d]	6	15.49[c]		31.56[d]
7	79.22[e]		65.81[f]	7	16.03[c]		29.62[d]
8	79.32[e]		66.62[f]	8	15.47[c]		28.86[d]
9	79.09		73.51	9	15.54		22.05
10	79.15[e]		69.42[f]	10	15.92[e]		25.18[f]
11	77.18[c]		70.40[d]	11	17.51		24.87

[a]Significantly different (P<.01) from b within comparisons.

[c]Significantly different (P<.05) from d within comparisons.

[e]Significantly different (P<.10) from f within comparisons.

TABLE 4. COMPARISON OF POST-FEEDING CECAL ACETATE AND PROPIONATE (MOLAR %)-CONCENTRATE AND CONCENTRATE PLUS Na_2CO_3 TREATMENTS

	Acetate				Propionate		
No. of hours	Na_2CO_3 treated	*vs*	Conc	No. of hours	Na_2CO_3 treated	*vs*	Conc
0	68.82		68.96	0	22.88		23.17
1	75.09[e]		69.30[f]	1	16.93[c]		23.42[d]
2	72.15		73.16	2	18.78		21.61
3	72.07[e]		64.57[f]	3	20.13[c]		26.83[d]
4	71.45		68.81	4	20.23		25.45
5	71.53[c]		64.61[d]	5	20.24[a]		30.28[b]
6	66.19		63.55	6	27.40		31.56
7	65.67		65.81	7	29.07		29.62
8	71.80		65.67	8	23.04		28.86
9	62.11		73.31	9	29.29		22.05
10	61.96		69.42	10	31.99		25.18
11	64.21		60.40	11	24.11		24.87

[a]Significantly different (P<.01) from b within comparisons.

[c]Significantly different (P<.05) from d within comparisons.

[e]Significantly different (P<.10) from f within comparisons.

rumen acetate and decreased rumen propionate with dietary additions of $NaHCO_3$. Hungate (1966) pointed out, that, in ruminants, starch digesters require a lower pH than do cellulose fermenters which suggested that a similar condition might exist in the cecum of the horse. These data confirm this since significant differences were found for cecal pH, acetate and propionate for the horses receiving the different treatments.

Mean cecal butyrate values (Molar %) for the 12-hr collection periods were 5.00 ± .15, 5.82 ± .39, and 7.41 ± .38, respectively, for the hay, concentrate and concentrate plus Na_2CO_3 treatments, and these differences were not significant. Increases in rumen butyrate in ruminants fed buffered high-concentrate diets were noted by Kromann and Meyer (1972) and

TABLE 5. COMPARISON OF AVERAGE PERCENTAGES OF TIME SPENT IN VARIOUS ACTIVITIES AND AMOUNT OF WOOD CHEWED BY HORSES RECEIVING DIFFERENT TREATMENTS

Activity	Hay	Conc	Conc + Na_2CO_3	Conc
Eating feed	39.53[c]	3.37[d]	3.73	3.37
Drinking water	1.00	1.20	1.13	1.20
Chewing wood	2.13[e]	10.67[f]	8.10	10.67
Coprophagy	.77[e]	2.73[f]	1.13[e]	2.73[f]
Licking salt	.53	1.76	2.43	2.76
Searching	7.50[e]	13.13[f]	12.43	13.13
Lying down	3.50	5.07	3.83	5.07
Standing	44.67[a]	61.67[b]	66.90[c]	61.67[d]
Amount of wood chewed kg/day	.30[c]	1.06[d]	.97	1.06

[a]Significantly different (P<.01) from b within comparisons.

[c]Significantly different (P<.05) from d within comparisons.

[e]Significantly different (P<.10) from f within comparisons.

Reid *et al.* (1957). It was suggested by Reid *et al.* (1957) that under pH controlled conditions, acetate could be combining to form butyrate.

Mean cecal lactate concentrations (mg/100 ml) for the 12-hr collection periods were 8.68 ± 1.50, 214.96 ± 56.41 and 99.43 ± 29.7u for the hay, concentrate and concentrate plus Na_2CO_3 dietary treatments. As may be noted from the large standard errors, cecal lactate was very variable. Hungate (1966) reported a number of species *Lactobacillus* and *Streptococcus bovis* in the rumen which grown under acid conditions and are enriched by fermentable carbohydrates at low pH. These lactate-producing microorganisms also were accompanied by lactate-utilizing bacteria *Veillonella gazogenes* and *Peptostreptococcus elsdenii*, which produce propionate via decarboxylation and acrylate pathways. However, he further stated that the lactate fermenters were not always present in the rumen of cattle fed high concentrate diets. This may explain the considerable amount of variation noted in lactic acid accumulation in this experiment, since both lactate utilizers and fermenters have been isolated from the cecal fluid of ponies (Kern *et al.*, 1973; Alexander and Davies, 1963).

The horses fed the hay diet spent significantly more time (table 5) eating feed and significantly less time chewing wood, in coprophagy, in searching (for feed) and in standing than did the horses receiving the concentrate diet. The amount of wood chewed per day was significantly greater for the horses fed the concentrate diet. The Na_2CO_3-treated horses spent significantly less time in coprophagy and significantly more time standing than did those fed the concentrate alone. The Na_2CO_3 infusions appeared to reduce the amount of time spent chewing wood by the concentrate-fed horses but the difference was not significant. Willard *et al.* (1973) reported that ponies fed an all-concentrate diet spent more time chewing wood, in coprophagy and in licking salt than did the ponies receiving a hay diet. The difference in the amount of wood chewed by the horses fed the hay diet was significantly less than for those fed concentrate (table 6). During the last 5 days of the experimental periods, when the cecal infusions were being administered, there was a marked increase in the mean amount of wood chewed by the concentrate-fed horses. This increase was significantly less for the Na_2CO_3-infused horses than for those receiving only the concentrate diet plus the sham infusion.

Simple correlations of the animal activity data and the cecal parameters (table 7) measured revealed that the amount of time spent chewing wood was significantly related with cecal propionate (r = +.63), cecal lactate (r = +.87), cecal acetate (r = —.61) and the amount of time spent eating (r = —.77). A highly significant relationship (r = —.97) between cecal acetate and cecal propionate was found. Cecal lactate and cecal pH also were significantly correlated (r = —.63).

The results of this experiment indicate that type of diet fed to horses alters cecal fermentation and suggests that the changes observed may be related to feeding behavior. It may be possible that increased cecal acidity and a narrowed acetate-propionate ratio, as was noted with the horses fed the all-concentrate diet, influences the horse's desire to chew wood and practice coprophagy. Since the Na_2CO_3 infusions were not completely effective in stopping wood chewing, a more continuous method of

TABLE 6. COMPARISON OF AVERAGE AMOUNT OF WOOD CHEWED PER DAY (KG) BEFORE AND AFTER INFUSIONS WERE INITIATED

	Hay	*vs*	Conc	Conc	*vs*	Conc + Na_2CO_3
Before infusion	.34		.54	.54		.79
After infusion	.30		1.06	1.06		.97
Difference	.04[a]		.52[b]	-.52[a]		-.18[b]

[a]Significantly different (P<.01) from b within comparisons.

TABLE 7. COMPARISONS OF ANIMAL ACTIVITIES AND CECAL PARAMETERS

	Eating	Drinking	Chewing	Eating salt	Searching	pH	Acetate	Prop.	Butyr.
Drinking	-.01								
Chewing	-.77[a]	.47							
Eating salt	-.52	.04	.17						
Searching	-.81[a]	-.29	.50	.59[c]					
pH	.12	-.39	-.48	-.05	.02				
Acetate	.80[a]	-.23	-.61[c]	-.59[c]	.52	.12			
Propionate	-.75[b]	.24	.63[c]	.41	.46	-.13	-.97[a]		
Butyrate	-.50	.08	.16	.90[a]	.42	-.02	-.49	-.27	
Lactate	-.49	.59[c]	.87[a]	.01	.22	-.63[c]	-.24	-.23	.12

[a]Significantly different (P<.01).
[b]Significantly different (P<.05).
[c]Significantly different (P<.10).

Na_2CO_3 addition or a stronger buffer may be needed. Also, when assessing the probable causes of wood chewing in horses, the amount of fiber in the diet and the degree of intestinal fill provided by the diet should be investigated.

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