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Studies on the biotin flow at the duodenum of dairy cows fed differently composed rations

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Introduction

It is well established that rumen microbes are able to synthesize B-vitamins, but knowledge of the effects of feed composition on net vitamins output is scarce. One of the B-vitamins of interest for ruminants is biotin. However, literature data on biotin flow at the duodenum vary considerably (NRC, 2001; Santschi et al., 2005).There is evidence from *in vivo* studies in steers that the portion and the type of grain in the diet influence the biotin metabolism in the rumen, resulting in changes in the duodenal flow of the vitamin (Miller et al., 1986). The objective of the study reported herein was to measure the biotin flow at the proximal duodenum of dairy cows either receiving pure hay- or mixed roughage/concentrate-rations with corn silage or grass silage, the latter being supplemented with high amounts of wheat- or corn-based concentrate, and to prove if there will be a correlation to the degree of microbial activity in the rumen.

Materials and Methods

Two dry (Diet 1) and four lactating (Diets 2 to 4) dairy cows, each fitted with cannulae in the rumen and in the proximal duodenum were used in four trials. On dry matter basis Diet 1 consisted of 8.9 kg grass hay, Diet 2 of 8.9 kg corn silage and 2.0 kg protein-mixture and Diets 3 and 4 of 7.4 and 7.3 kg grass silage and 10.0 kg concentrate mixture containing 87% wheat or corn grain respectively, 11% soybean meal and 2% oil. Mineral-vitamin premix in the concentrates was free of biotin. Diets were given restrictively to avoid feed refusals and administered in two equal sized meals. Concentrate of Diets 3 and 4 was given in four meals.

Each four-week experimental period (one per diet) consisted of two weeks for adaptation to the diet, one week during which ruminal fluid for three days (180 min. after the start of morning feeding) was collected for the determination of pH, ammonia and short chain fatty acids (SCFA) and one week for duodenal sample collection according to Rohr et al (1979). To estimate digesta flow at the duodenum, Cr_2O_3 mixed with wheat flour was used (Rohr et al., 1984) and microbial N in the duodenal chyme was determined by near infrared spectroscopy (NIRS) as described by Lebzien and Paul (1997). Biotin in feedstuffs and duodenal digesta was measured applying a microbiological method (Frigg and Brubacher, 1976).

Results

The rumen fermentation data are shown in Table 1.

Table 1. umen fermentation parameters 180 min after the start of morning feeding (means and standard deviations)

	Grass hay	**Corn silage + concen-trate**	**Grass silage + wheat based concentrate**	**Grass silage + corn based concentrate**
Diet	**1**	**2**	**3**	**4**
(n)	(6)	(12)	(9)	(9)
pH	6.74 [a] ± 0.18	6.20 [b] ± 0.15	5.47 [d] ± 0.26	5.82 [c] ± 0.39
NH_3-N (mg/100ml)	10.8 [b] ± 0.4	12.8 [b] ± 3.3	6.3 [c] ± 3.0	17.8 [a] ± 6.2
SCFA (mmol/l)	80.9 [d] ± 15.6	96.6 [c] ± 8.3	143.8 [a] ± 18.4	134.7 [b] ± 16.2
molar proportion (%)				
acetate	71.7 [a] ± 1.6	68.0 [a] ± 4.6	56.9 [b] ± 2.7	59.0 [b] ± 3.2
propionate	16.4 [c] ± 0.9	16.3 [c] ± 2.0	28.1 [a] ± 5.3	22.7 [b] ± 3.3
butyrate	8.1 [c] ± 1.1	11.1 [b] ± 2.3	15.4 [a] ± 3.1	14.3 [a] ± 1.1
acetate : propionate	4.4 : 1[a]	4.2 : 1[a]	2.1 : 1[c]	2.7 : 1[b]

a > b > c > d in the same line: $p < 0.05$

The pH value decreased and short chain fatty acid concentration increased with an increasing concentrate portion in the diet which was more pronounced in combination with the wheat than with the corn grain based concentrate. Ammonia concentration was significantly lower with Diet 3 and significantly higher with Diet 4 as compared to Diets 1 and 2. Acetate : propionate ranged from 2.1 : 1 (Diet 3) to 4.4 : 1 (Diet 1). Daily means for the intake of organic matter (OM), crude protein (CP) and biotin, as well as the average amounts of OM fermented in the rumen (FOM) and of microbial nitrogen (MN) and biotin at the duodenum are summarized in Table 2.

The percentage of FOM was significantly higher when feeding the concentrate rich diet with wheat grain (Diet 3) as compared to Diet 1 or 4. Efficiency of microbial protein synthesis (g MN/kg FOM) increased with increasing DM intake.

Table 2. Intake of organic matter (OM), crude protein (CP) and biotin, OM fermented in the rumen (FOM)[1] as well as flow of microbial nitrogen (MN) and biotin at the proximal duodenum

Diet	Grass hay 1	Corn silage + concentrate 2	Grass silage + wheat based concentrate 3	Grass silage + corn based concentrate 4
(n)	(2)	(4)	(3)	(3)
Intake				
OM (kg/d)	8.33	10.27	16.12	16.29
CP (g/d)	1188	1412	2849	2646
Biotin (mg/d)	2.55	2.35	4.49	4.20
(mg /kg DM)	0.29	0.22	0.25	0.24
FOM[1] (kg/d)	5.94 ± 0.09	7.83 ± 0.39	13.13 ± 0.32	11.59 ± 0.65
(% of OM intake)	71.3^{b} ± 1.06	76.2^{ab} ± 3.76	81.5^{a} ± 2.02	71.1^{b} ± 3.94
Flow at the duodenum				
MN (g/d)	120.0 ± 9.5	174.6 ± 32.3	395.1 ± 17.3	356.1 ± 37.2
(g/kg FOM)	20.2^{b} ± 2.2	22.3^{b} ± 3.6	30.9^{a} ± 1.9	30.5^{a} ± 1.6
Biotin (mg/d)	1.84 ± 0.62	4.37 ± 0.41	5.89 ± 1.35	6.16 ± 0.93
(mg/kg FOM)	0.31^{b} ± 0.10	0.56^{a} ± 0.05	0.45^{ab} ± 0.09	0.53^{a} ± 0.05
(µg/g MN)	15.4^{ab} ± 6.7	25.0^{a} ± 3.6	14.9^{b} ± 2.8	17.3^{b} ± 1.1
Biotin flow – biotin intake (mg/d)	-0.71	2.02	1.40	1.96

a > b: $p < 0.05$
[1]FOM = OM-intake – (OM flow at the duodenum – microbial OM) where microbial OM = microbial N x 11.8 (Schafft 1983)

Biotin intake per kg DM was roughly the same for the four diets (0.22 – 0.29 mg/kg DM). In consequence the biotin intake depended primarily on the level of feed intake. Biotin flow at the duodenum was lowest with Diet 1. Consequently, the biotin balance calculated as difference between flow at the duodenum and intake was negative in the hay fed cows (Diet 1) and positive in association with the other three diets.

Discussion

The amounts of biotin determined in the present investigations leaving the rumen may give some hints as to ruminal biotin degradation/utilisation and ruminal microbial net synthesis in cows supplied with different amounts of dietary biotin and differently composed feed rations entailing large variations in ruminal fermentation conditions. In agreement with other studies (Miller et al., 1986; Zinn et al., 1987), the positive biotin balances observed in our study clearly confirm biotin synthesis by rumen microbes. In contrast Santschi et al. (2005) could not observe a net biotin synthesis in the rumen. Biotin flow at the duodenum was not significantly ($p > 0.05$) related to the amounts of consumed biotin (r = 0.63) but to the amount of organic matter fermented in the rumen (r = 0.85) and to the amount of synthesised microbial protein (r = 0.84). More duodenal biotin per kg FOM was measured in cows supplied with the mixed diets. These results agree with earlier observations in fistulated steers (Miller et al., 1986), and may be associated with the shift of propionate production from biotin-dependent randomising to the biotin-independent acrylate pathway when easily fermentable carbohydrates are supplied with the concentrate. Overall mean daily biotin flow was 0.48 ± 0.11 mg/kg FOM. Taking a mean daily biotin requirement of 6 mg, given by NRC (2001) for a dairy cow (650 kg LW) with 35 kg fat corrected milk, into account it can be calculated, that about 12.5 kg OM have to be fermented.

Summary

Dairy cows fitted with permanent cannulas in the dorsal rumen and in the proximal duodenum were fed differently composed diets, and the biotin flow at the proximal duodenum was measured. The diets (on DM basis) consisted of 8.9 kg grass hay (Diet 1), 8.9 kg corn silage plus 2.0 kg concentrate (Diet 2), or 7.3 and 7.4 kg grass silage plus 10.0 kg concentrate (Diets 3 and 4). The concentrate in Diets 3 and 4 contained 87% wheat and corn grain, respectively. The duodenal flow of biotin was not significantly related to biotin intake, but to the amount of fermented organic matter (FOM) and the amount of microbial protein (r = 0.85 and r = 0.84), irrespective of the composition of the diet fed. Mean daily biotin flow was 0.48 ± 0.11 mg/kg FOM without any systematic effect of diet composition.

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