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Effect of the grass silage particle size offered as TMR on the ruminal thiamine concentration in high-yielding dairy cows*

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ABSTRACT

The thiamine concentration was studied in solid and fluid rumen digesta of three Holstein cows, fitted with rumen cannulae and fed grass silage-based TMR with three particle sizes (25, 11 and 5.5 mm). The decrease of the particle size led to an increase of the thiamine concentration in solid ($P<0.05$) and fluid rumen digesta. Thiamine concentration was increased (R^2=0.56, $P<0.001$) as digestible organic matter intake increases. The pH (mean 5.81) did correlate negatively (R^2=0.21 to 0.26, $P<0.001$) to the thiamine concentration of rumen. Thiamine concentration of the rumen digesta did reflect in great extent the changes of rumen conditions.

KEY WORDS: thiamine, particle size, rumen digesta, dairy cows

INTRODUCTION

About 90% of ruminal thiamine is synthesized by microorganisms (Breves et al., 1981), which provide a sufficient thiamine amount to the ruminant animal. The thiamine requirement for the high yielding cows can be higher (Girard, 2000), due to the high thiamine excretion through milk and the high concentrate level offered, which can impair the rumen digestion. Feed particle size (PL) can also influence the rumen digestion. The reduction of PL of grass silage (GS) from 25 to 5.5 mm increased the feed intake (Zebeli et al., 2003; Tafaj et al., 2004) while the rumen conditions were slightly impaired. As ruminal thiamine content is a result of microbial activity, we hypothesised that the effects of PL of GS on rumen milieu can be also reflected in the thiamine content. The results of the same

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experiment about the digestible organic matter intake (DOMI), pH and ruminal concentration of SCFA, diaminopimelic acid (DAPA) and protozoa (Zebeli et al., 2003; Tafaj et al., 2004) are also correlated to the thiamine concentration.

MATERIAL AND METHODS

Animals, diets and sample collection

The study was carried out on three Holstein cows (BW 640 kg, 63 DIM, 36 kg milk/d) fitted with ruminal cannulae (100 mm i.d.). Grass silage (GS) (13.2% CP, 46.5% NDF, degradation rate of DM [DM-k_d] 0.087) that was harvested at 25mm theoretical length provided the coarse GS. By re-cutting the ensiled GS in a forage harvester the PL levels of 11 and 5.5 mm were obtained. The GS was offered as TMR (15.1% CP, 35% NDF, DM-k_d 0.105) with a concentrate (C) mixture (18.7% CP, 19.3% NDF, DM-k_d 0.127) and hay (H) (GS:H:C= 45:5:50). Cows were fed *ad libitum* twice daily at 08.00 and 16.00 h. After 11-d adaptation, the digesta samples were collected from different rumen digesta compartments (D) in 4 sampling times (T: 1 h prior, 2 h, 7 h and 11h after the morning feeding). Each sampling was repeated over two days. Solid digesta was taken from dorsal and ventral rumen according to Tafaj et al. (2001), pooled, manually squeezed and filtered through cloth bags (SRF-S). Free liquid (FRL) was collected from ventral rumen by a vacuum pump. The samples for thiamine analyses were kept frozen at -20°C for no longer as six months.

Thiamine analysis

Thiamine was extracted out of the digesta by acidic and enzymatic digestion according to Analytical Methods Committee (1999), modified. Analysis was performed by HPLC, using a reversed-phase column and fluorescence detection, after the precolumn derivatization of thiamine to thiochrome using $K_3[Fe(CN)_6]$.

Statistical analyses

The effects of PL and D and PL×D on thiamine concentration were tested using a model for double (D, T) repeated measures (PROC MIXED, SAS, 8.2). The relationships among various parameters were tested by PROC REG, SAS, 8.2.

RESULTS AND DISCUSSION

Mean value and range of parameters studied are shown in Table 1. Reducing the PL of GS led to an increase ($P<0.05$) of thiamine concentration in the SRF-S, while that in FRL was numerically higher than 11 and 25 mm PL. The SRF-S showed higher ($P<0.05$) thiamine concentration than FRL presumably due to the differences in the concentration of microorganisms and substrate in different digesta compartments (Table 2).

Table 1. Mean and range of the parameters studied

Factor	N	Mean	SD	Range
Thiamine, µg/g	214	0.64	0.337	0.13 – 1.67
DOMI, g/kg $W^{0.75}$	27	118.3	6.50	107.7 – 128.9
SCFA, mmol/l	213	136.8	22.67	77.2 – 189.6
pH-Value	213	5.81	0.40	5.12 – 6.97
Protozoa number, 10^5 /ml	214	7.03	4.94	0.94 – 21.8
DAPA /N digesta, mg/g	27	9.79	2.95	5.35 – 22.44

Table 2. Thiamine concentration (µg/g) at different digesta compartments (D), particle size (PL) of grass silage and sampling times (T: 1h prior, 2, 7 and 11 h after the feeding) (Lsmeans) (n=6)

Sampling time, h	SRF-S				FRL				Significance
	25 mm	11 mm	5.5 mm	SEM	25 mm	11 mm	5.5 mm	SEM	P<0.05
1 h prior	0.53^{a}	0.65^{b}	0.93^{b}	0.065	0.26	0.26	0.49	0.083	PL, D
2 h	0.58^{a}	0.85^{ab}	0.99^{b}	0.084	0.33	0.29	0.49	0.098	PL•D, D
7 h	0.60^{a}	0.92^{b}	0.64^{b}	0.074	0.34	0.31	0.39	0.090	PL•D, D
11 h	0.69^{a}	1.16^{a}	0.89^{b}	0.071	0.33	0.48	0.31	0.095	PL, D

SRF-S - digesta particle-associated fluid; FRL= free ruminal fluid; ^{ab} significant differences among PL levels within the digesta compartment

Table 3. Relationship between thiamine concentration (Y, µg/g) and various parameters (X)

Parameters	Digesta	Relationship	R^2	Significance
DOMI, g/kg $W^{0.75}$	SRF-S	Y = -41.5 + 0.69 (± 0.175) X – 0.003 (± 0.0007)X^2	0.56	***
	FRL	Y = -21.4 + 0.36 (± 0.180) X – 0.002 (± 0.0007) X^2	0.28	n.s.
SCFA, mmol/l	SRF-S	Y = -0.48 + 0.009 (± 0.0010) X	0.33	***
	FRL	Y = 0.1+ 0.002 (± 0.0007) X	0.10	**
pH-Value	SRF-S	Y = 3.56 – 0.48 (± 0.071) X	0.26	***
	FRL	Y = 1.39 – 0.16 (± 0.039) X	0.21	***
Protozoa No. 10^5 /ml	SRF-S	Y = 0.63 + 0.02 (± 0.005) X	0.10	***
	FRL	Y = 0.46 – 0.039 (± 0.0146) X + 0.002 (± 0.0009) X^2	0.10	**
DAPA/N Digesta, mg/g	SRF-S	Y = 0.49 + 0.03 (± 0.015) X	0.13	*
	FRL	Y = 0.44 – 0.006 (± 0.0156) X	0.01	n.s.

* P<0.05; **P<0.01; ***P<0.001; R^2= determination coefficient; DOMI= digestible organic matter intake; SCFA=short-chain fatty acids; DAPA= 2.6-diaminopimelic acid

The ruminal thiamine concentration increased for PL of 11 mm with time after feeding. This trend was different for the PL of 5.5 mm. With increasing the DOMI the concentration of thiamine increased more in SRF-S (R^2=0.56) as in FRL (R^2=0.28). This relationship is also consistent with the positive relationship between the SCFA and the thiamine concentration in SRF-S (R^2=0.33), presumably due to a better availability of energy for the ruminal microbial synthesis. This assumption was also partially supported by the positive relationship of thiamine concentration to the DAPA and protozoa concentration in SRF-S (Table 3). Breves et al. (1981) found a positive relationship between the flow of thiamine and DOMI (R^2=0.87) as well as the microbial nitrogen (R^2=0.85) in duodenum. The relationship between the thiamine concentration and the ruminal pH was negative and clearly higher on 11 (R^2=0.77; $P<0.001$) and 5.5 mm (R^2=0.58; $P<0.001$) than on 25 mm (R^2=0.12; $P<0.05$). The low pH values (5.1 to 5.5) were observed 1 h after the morning feeding, only in SRF-S. Alves de Oliveira et al. (1996) found also a negative relationship between pH (range 5.17 to 7.01) in the fluid digesta of RUSITEC system and the thiamine concentration.

CONCLUSIONS AND IMPLICATIONS

The reduction of PL of GS from 25 to 11 and 5.5 mm led to an increase of thiamine concentration, mainly in SRF-S. The negative relationship of thiamine concentration to pH need to be considered only for the pH range studied. The higher thiamine concentration of SRF-S compared to FRL did reflect the compartmental differences of microbial activity. The ruminal thiamine concentration reflected in a great extent the changes of rumen conditions. Further studies are needed to prove in which extent the ruminal thiamine concentration can be used as a reliable indicator for studying the microbial activity in the rumen.

REFERENCES

Alvez de Oliveira L., Jean-Blain C., Durix A., Komisarczuk-Bony S., Durier C., 1996. Use of a semi-continuous culture system (Rusitec) to study the effect of pH on microbial metabolism of thiamine (vitamin B_1). Arch. Anim. Nutr. 49, 193-202

Analytical Methods Committee, 1999. Determination of thiamine and riboflavin in pet foods and animal feedingstuffs. Analyst 125, 353-360

Breves G., Brandt M., Höller H., Rohr K., 1981. Flow of thiamin to the duodenum in dairy cows fed different rations. J. Agr. Sci. 96, 587-591

Girard C.L., 2000. A new look at the requirements of high-producing dairy cows for B-complex vitamins. In: P.C. Garnsworthy, J. Wiseman (Editors). Recent Advances in Animal Nutrition. Nottingham, University Press, pp. 89-107

Tafaj M., Maulbetsch A., Junck B., Steingaß H., Drochner W., 2001. Research note: A method for studying of local differences in ruminal fermentation in dairy cattle. Arch. Anim. Nutr. 54, 341-347

Tafaj M., Zebeli Q., Junck B., Drochner W., 2004. Influence of particle size of grass silage fed as TMR on chewing activity and rumen fermentation on high-yielding dairy cows. Proc. Soc. Nutr. Physiol. 13, 76

Zebeli Q., Tafaj M., Junck B., Drochner W., 2003. Einfluss der Partikellänge der Grassilage einer TMR auf die Verdaulichkeit bei Hochleistungskühen. VDLUFA-Kongress, pp. 43-44