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Enterolactone production and its correlation among body fluids in cows fed flax meal

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Introduction and Objective

- Flax (*Linum usitatissimum*) is the richest source of the plant lignan secoisolariciresinol diglucoside (SDG; Prasad, 1997)
- SDG is converted in secoisolariciresinol (SECO) under the action of the microbial enzyme glycosidases in the intestine of human (Saarinen et al., 2002) and SECO is converted mainly into enterodiol (ED) and enterolactone (EL; Clavel et al., 2006)
- In ruminants, conversion of flax lignans into EL occurs mainly in the rumen (Gagnon et al., 2009) .
- There are linear increases in EL concentration of plasma, milk and urine of cows when flax hulls are placed in the rumen (via cannula) (Gagnon et al., 2009).
- In ruminant animals, importance of the activity of ß-glucuronidase, which enhances absorption of mammalian lignans in non-ruminant mammals, is unknown.
- The objectives were to investigate the relationships between concentrations of EL in milk and other body fluids when cows are fed increased proportions of flax meal (FM) in the diet.

Materials & methods

- Eight lactating Holstein cows with a ruminal fistula were allotted to a replicated 4 x 4 Latin square design with four 21 d periods.
- Four (4) diets were fed for *ad libitum* intake: 0 (CON), 5% (5FM), 10% (10FM) and 15% FM (15FM) in the dietary dry matter (15FM).
- On day 20:
  - ✓ Ruminal contents were collected before (0 h), and 2, 4, and 6 h after feeding (samples pooled for 2, 4 and 6 h),
  - ✓ Samples of blood were collected 6 h post-feeding.
- On day 21:
  - ✓ Samples of urine were collected 2 h post-feeding.
  - ✓ Samples of milk were collected from am and pm milkings.
- Lignans in ruminal fluid, plasma, urine and milk samples were hydrolysed, extracted and analyzed as described by Gagnon et al. (2009). Determination of EL was performed using an EIA kit (Cayman Chemical, Ann Arbor, MI, USA).
- Data were analyzed as to a replicated 4 x 4 Latin square design using PROC MIXED of SAS. The Spearman's correlation test was used to determine strength of the relationships among samples with the CORR procedure of SAS.

Table 1. Ingredient and chemical composition of the experimental diets

	CON	5FM	10FM	15FM
Ingredient, g/kg DM				
Grass silage	315	316	317	314
Corn silage	292	291	290	290
Ground maize grain	211	202	192	189
Soybean meal (48% CP)	108	74.5	41.4	20.4
Beet pulp	34.3	29.6	25.0	16.1
Calcium carbonate (35% Ca)	5.5	5.5	5.4	5.5
Flax meal	0	47.9	95.3	141.0
Protein supplement	17.4	17.4	17.9	8.5
Minerals and vitamins	16.4	16.3	16.3	15.5
Chemical				
DM, %	37.7	37.9	37.6	38.1
CP, % of DM	17.0	17.4	17.6	17.9
Ether extract, % of DM	2.44	2.41	2.34	2.41
NDF, % of DM	28.4	28.6	29.5	29.6
ADF, % of DM	18.3	18.5	19.2	19.3
NE_L, KJ/kg of DM	6.65	6.61	6.61	6.61

Results

✓ Concentrations of EL in urine, ruminal fluid (RF), milk and plasma increased linearly ($P<0.01$) with higher FM concentration.

✓ Correlation coefficients were statistically significant for all tested combinations except for correlation between EL concentration in urine and RF at 0 h.

✓ Correlation coefficients between EL concentration in RF 2 h post-feeding and EL in milk, EL in RF before and post-feeding, EL in plasma and urine, and EL in plasma and milk were, respectively, 0.76, 0.75, 0.64, and 0.61.

Discussion

✓ Concentration of EL in milk presented the highest correlation with EL concentration in ruminal fluid at 2 h post-feeding, suggesting that the conversion of plant into the mammalian lignan EL in the rumen is responsible for the increased EL concentration in milk.

✓ Data may indicate that EL is absorbed directly through the rumen wall and that the concentration of EL in milk depends directly on the concentration of EL in ruminal fluid.

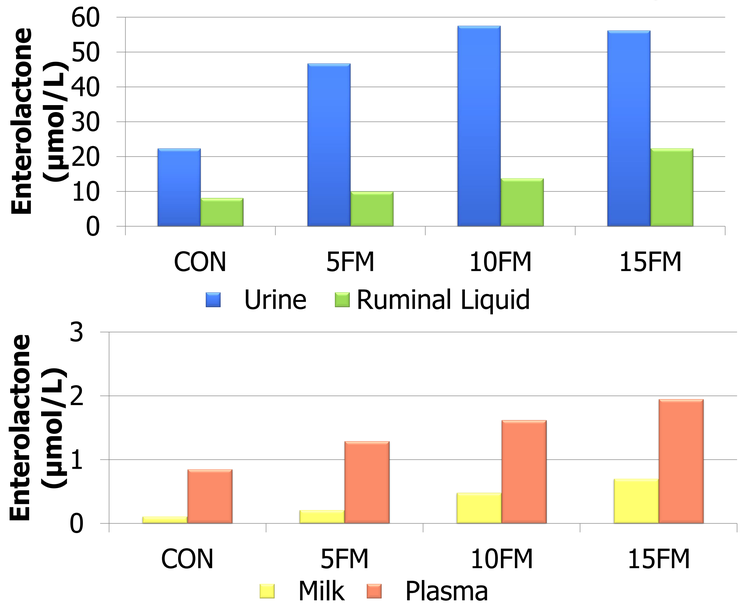

Figure 1. Concentration of enterolactone (µmol/L) in urine and ruminal fluid (a) and milk and plasma (b) in Holstein cows fed no flax meal (CON) or 5 (5FM), 10 (10FM) and 15% (15FM) flax meal in the diet.

Variables	EL milk	EL plasma	EL urine	EL RF 0 h	EL RF 2 h
EL milk	-				
EL plasma	0.61‡	-			
EL urine	0.48¶	0.64‡	-		
EL Rum 0 h	0.56‡	0.40¶	0.30†	-	
EL rum 2 h	0.76‡	0.55‡	0.39¶	0.75‡	-

Table 2. Spearman's correlation coefficients between EL concentrations of milk, plasma, urine and ruminal fluid (RF) of Holstein cows fed no flax meal or 5, 10 and 15% flax meal in the diet. Significance level: †$P<0.10$; ¶$P<0.05$; ‡$P<0.001$.

Conclusions

- As EL has antioxidant properties, increased production of EL in the rumen resulting from flax supplementation may contribute to better health of animals and quality of milk.

References

Clavel et al. 2006. Anaerobe 12:140-147; Gagnon et al. 2009. Br. J. Nutr. 102:1015-1023; Prasad, K. 1997. Mol. Cell. Biochem. 168:117-121; Saarinen et al. 2002. J. Chromatogr. B 777:311-319.

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