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BRIEF COMMUNICATION

Effect of atropine on milk protein composition of dairy cows

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Milk production by dairy cows and yield of the components, fat, protein and lactose are closely related to nutritional intake. The individual proteins in milk respond differently to a change in nutritional status, giving rise to significant differences in milk protein composition (DePeeters and Cant, 1992). For instance, Gray and Mackenzie (1987) noted a decrease in α -lactalbumin and increase in serum-derived proteins, serum albumin and immunoglobulin, in animals on restricted pasture for 10 days. Although β -lactoglobulin concentration decreased, the response was not significant. Treatment of concentrate-fed dairy cows with a single injection of atropine caused a similar, though temporary, decrease in milk yield, total protein and α -lactalbumin content of milk (Roets and Peeters, 1981). Changes in other milk proteins were not evaluated. The observed responses arise from the ability of atropine to rapidly and specifically depress plasma amino acids used for milk protein synthesis and provide a means of experimentally manipulating supply of substrates for milk protein synthesis. The present study was designed to evaluate the use of atropine in pasture-fed cows.

Lactating cows (n=12) were housed indoors in individual stalls and fed pasture *ad libitum*. Each cow was given a subcutaneous injection of saline or atropine (20 mg in saline) on separate days immediately following the morning (8 am) milking. The animals were milked at 4 hourly intervals until 12 h following injection and then normally at 8 am the following day. Blood was collected via the coccygeal vein just prior to, and 2 and 6 h after, injection. Plasma lactose was measured according to Stelwagen *et al.*, (1995). Milk was analysed for α -lactalbumin (Prosser *et al.*, 1992) and serum albumin, β -casein and β -lactoglobulin by specific ELISA. The ratio of γ -casein to β -casein was measured by FPLC and used as an index of endogenous plasmin activity in milk (Prosser *et al.*, 1995). Differences between saline and atropine over time were tested by ANOVA using repeated measures, with pre-treatment value as a co-variate.

Milk yield or composition before injection of saline was not different to that before atropine. Milk yield decreased significantly due to atropine (Table 1). Concentrations of α -lactalbumin decreased by 30% due to atropine injection, but were largely restored by the 8 am milking the day after treatment. The fall in α -lactalbumin, and hence lactose output, would explain the decrease in milk yield. Concentrations of β -lactoglobulin tended to increase with four-hourly milking

following saline or atropine, although the change in concentrations of β -lactoglobulin appeared to be less following atropine. We are unaware of similar effects of more frequent milking on β -lactoglobulin, or any other milk protein, so are unable to determine whether this is usual or to suggest a mechanism for how this would occur.

Concentrations of serum albumin and the ratio of γ/β -casein increased due to atropine, compared with saline. The γ/β -casein ratio was previously shown by us to reflect increased plasmin activity in milk. Plasmin, like albumin, is derived from serum (Prosser *et al.*, 1995). Gray and Mackenzie (1987), likewise observed increased serum proteins in milk of cows on restricted pasture, which they attributed either to their increased influx between mammary epithelial cells or a concentrating effect due to lower milk volume.

We have previously used lactose in plasma as an indicator of leakage of milk constituents out of, and blood constituents into, the gland (Stelwagen *et al.*, 1995). However, in the present study plasma lactose was not different in atropine compared with saline (data not shown), which strongly suggests the increase in serum-derived proteins was due to a concentrating effect of lower milk volume.

In conclusion, atropine offers a useful method for evaluating effects of substrate supply on milk composition. The advantage of this technique over restricting feed intake is that only plasma amino acids are depressed and the onset of changes are rapid (within 4-8 h) and readily reversible (generally by 12 h). This means that this treatment can be used to reduce basal amino acid concentrations and then add back (via intra-venous infusion) specific amino acids or combinations thereof to determine their relative importance in milk protein synthesis. The different response of individual mammary-synthesised proteins to atropine may reflect a different amino acid requirement for their synthesis, raising the possibility that individual milk proteins may be more susceptible to nutritional challenges than others. If so, this would have significant follow-on effects for processing milk, which is influenced by proportions of individual proteins in milk (Dalglish, 1992).

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TABLE 1: Milk yield and composition following subcutaneous injection of saline (sal) or atropine (atrp) into lactating cows (n=12).

Time from injection (h)	4		8		12		24		pooled SD	P atrp vs sal
	sal	atrp	sal	atrp	sal	atrp	sal	atrp		
Milk Yield (kg)	2.3	1.9	2.3	1.8	2.0	2.1	2.1	2.2	0.11	0.03
α -lactalbumin (mg/ml)	1.01	0.95	1.01	0.71	1.02	0.77	1.11	0.97	0.05	0.01
β -lactoglobulin (mg/ml)	4.4	4.0	5.3	5.1	6.2	4.8	3.8	4.8	0.50	0.49
β -casein (mg/ml)	8.5	9.4	8.4	9.1	8.2	11.5	10.2	12.1	0.38	0.001
BSA (mg/ml)	214	228	185	270	162	293	188	171	27	0.004
σ/β -casein ratio	0.31	0.34	0.30	0.38	0.32	0.39	NA	NA	0.02	0.001

Values are means adjusted for pre-treatment values.

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