

# Effect of supplement strategy on splanchnic net fluxes of ammonia and urea in dairy cows fed fresh grass

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## ABSTRACT

Different strategies for supplementation of grass with rolled barley and soya hulls were investigated as tools for improving the nitrogen efficiency of dairy cows under restricted zero-grazing (47% grass DM) conditions. Three multicatheterized cows were subjected to 3 experimental treatments in a Latin square design. Treatments were: SAM, soya hulls fed am and barley fed pm; BAM, barley fed am and soya hulls fed pm; SBPM barley and soya hulls fed pm. The total daily ration was the same for all treatments. The arterial concentrations, net portal fluxes and net hepatic fluxes of ammonia, urea-N, oxygen and glucose were not affected by treatments. The cows had higher net portal flux of ammonia than transfer of urea-N to the gut although the diet had a low CP content (12.4%). Efficient recycling of nitrogen to the rumen combined with decreased nitrogen intake is in theory a promising strategy for improved nitrogen efficiency in ruminants. However, better understanding of factors affecting nitrogen recycling in ruminants is necessary for a successful utilization of this strategy.

KEY WORDS: ammonia, urea, nitrogen recycling, dairy cows

## INTRODUCTION

Approximately 25% of the nitrogen input to a dairy herd is recovered in milk and meat. The aim of the present study was to investigate the effect of three supplement strategies in a zero-grazing system on splanchnic net fluxes of ammonia and urea and splanchnic energy metabolism in dairy cows.

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## MATERIAL AND METHODS

Three Holstein dairy cows (BW 620, 580, and 624 kg; milk yield 20, 7, and 21 kg/d; parity 1, 2, and 3; DIM 251, 255, and 294 d, respectively) fitted with permanent indwelling catheters in the mesenteric artery, mesenteric vein, hepatic portal vein, and hepatic vein as well as a ruminal cannula were used. The diet (composition % of DM; fresh grass harvested daily, 47; maize silage, 16; soya hulls, 18; rolled barley, 18; barley straw, 1) was fed according to three different supplement strategies: soya hulls a.m. and barley p.m. (SAM), barley a.m. and soya hulls p.m. (BAM), and both barley and soya hulls fed p.m. (SBPM). Cows were milked at 6 a.m. and 4 p.m. Barley and soya hulls were fed at 7 a.m. and 3.30 p.m. Fresh clover grass was harvested 7.30 a.m. and fed 8 a.m.

Treatments were arranged in a Latin square with 1 wk periods and blood, rumen fluid, urine and faeces were collected over a 24 h period on the last day of each experimental period. Continuous infusion of p-aminohippuric acid (pAH,  $31 \pm 3$  mmol/h) into the mesenteric vein was initiated at 5 am and 16 sets of blood samples (20 ml) were obtained simultaneously from the artery, portal, and hepatic catheters at 1.5 h intervals starting at 6 a.m.

The oxygen content in whole blood was determined by an ABL520 (Radiometer, Copenhagen, Denmark) and blood glucose was determined on fresh blood using an Accu-Check compact (Roche Diagnostics, Mannheim, Germany). The packed cell volume was determined by centrifugation of micro capillary tubes and plasma was separated by centrifugation at  $3.000 \times g$ , at  $4^\circ\text{C}$  for 20 min. Previously described methods were used to analyse pooled plasma samples for ammonia (Rhine et al., 1998), urea (Marsh et al., 1965), and pAH (Harvey and Brothers, 1962). Ammonia in ruminal fluid was analysed by the same method as used for plasma.

Twenty-four h treatment means were analysed by ANOVA by the GLM procedure of SAS (SAS Inst. Inc., Cary, NC, USA). The model included the effect of cow, period, and treatment. Values in the text are given as means of three cows  $\pm$  SEM.

## RESULTS AND DISCUSSION

The experiment was conducted in a period with abnormal spring weather from May 20 to June 10, 2004. The grass had a lower content of CP (14.3% of DM) than expected and the total ration was therefore low in CP (12.4% of DM). The DMI ( $14 \pm 2$  kg/d) did not differ ( $P=0.50$ ) between treatments. The ruminal ammonia concentration was in average  $1.7 \pm 0.6$  mmol/l and lower than the expected optimal level for microbial growth (3.5 to 6 mM; Parker et al., 1995).

None of the arterial variables tested were affected by treatment ( $P \geq 0.10$ ; Table 1). In agreement with the low CP content in the diet, the plasma urea-N concentrations were low, but the arterial ammonia concentrations were relatively high in the present study compared with literature data (Reynolds et al., 2003).

Table 1. Arterial packed cell volume and concentrations of pAH, ammonia, urea-N, oxygen and glucose in dairy cows with 3 different supplement strategies in a zero-grazing system (SAM: soya hulls fed a.m. and barley fed p.m.; BAM barley fed a.m. and soya hulls fed p.m.; SBPM soya hulls and barley fed p.m.). Mean of three cows  $\pm$  SEM

Item	Treatment			SEM	P
	SAM	BAM	SBPM		
<i>Proportion of whole blood</i>					
packed cell volume	0.237	0.240	0.239	0.003	0.83
<i>Mmol/l of plasma</i>					
p-aminohippurate	0.10	0.09	0.08	0.01	0.66
ammonia	0.51	0.51	0.46	0.01	0.10
urea-N	2.10	2.16	2.76	0.55	0.70
<i>Mmol/l of whole blood</i>					
oxygen	4.92	4.79	4.87	0.04	0.25
glucose	3.6	3.5	4.2	0.3	0.41

The portal and hepatic blood flow as well as the net portal and net hepatic fluxes of ammonia, urea-N, glucose and oxygen did not differ between treatments ( $P>0.10$ ; Table 2). The net portal flux of ammonia accounted for 22% of the

Table 2. Portal and hepatic blood flow as well as net portal and net hepatic fluxes of ammonia, urea-N, oxygen and glucose in dairy cows with 3 different supplement strategies in a zero-grazing system (SAM: soya hulls fed am and barley fed pm; BAM barley fed am and soya hulls fed pm; SBPM soya hulls and barley fed pm). Mean of three cows  $\pm$  SEM

Item	Treatment			SEM	P
	SAM	BAM	SBPM		
<i>Blood flow, l/h</i>					
portal	1218	1217	1074	105	0.62
hepatic	1561	1561	1613	130	0.95
<i>Net portal flux, mmol/h</i>					
ammonia	201	134	219	21	0.18
urea-N	-51	-160	-142	38	0.30
glucose	214	162	57	65	0.40
oxygen	-1502	-1429	-1349	171	0.83
<i>Net hepatic flux, mmol/h</i>					
ammonia	-143	-84	-157	49	0.61
urea-N	179	156	142	69	0.96
glucose	841	835	730	96	0.70
oxygen	-1823	-1598	-2187	258	0.43

nitrogen intake. Based on the expected digestibility of ingested N this value is in the lower range of previously reported values, which is in agreement with the

low nitrogen intake in the present study (Lapierre and Lobley, 2001). Despite an expected negative nitrogen balance in the rumen (AAT/PBV system) the negative net portal flux of urea accounted for only 64% of net portal flux of ammonia. The estimated salivary urea flux was negative (-23 mmol/h; net hepatic flux of urea - urinary urea excretion - portal urea uptake). The negative value is probably reflecting sampling and/or analytical errors, but does indicate that the salivary contribution with urea to the lumen of the gut was low under the present experimental conditions.

## CONCLUSIONS

Dairy cows fed a grass-based diet with low nitrogen content had a higher net portal flux of ammonia than urea-N transfer to the gut. These observations indicate that although the ruminant system in principle has the potential to be highly nitrogen efficient we need to know more about nitrogen metabolism of dairy cattle in order to increase the overall nitrogen efficiency of dairy operations.

## REFERENCES

- Harvey R.B., Brothers A.J., 1962. Renal extraction of para-aminohippurate and creatinine measured by continuous in vivo sampling of arterial and renal-vein blood. *Ann. N. Y. Acad. Sci.* 102, 46-54
- Lapierre H., Lobley G.E., 2001. Nitrogen recycling in the ruminant: A review. *J. Dairy Sci.* 84, Suppl. 1, E223-E236
- Marsh W.H., Fingerhut B., Miller H., 1965. Automated and manual direct methods for the determination of blood urea. *Clin. Chem.* 11, 624-627
- Parker D.S., Lomax M.A., Seal C.J., Wilton J.C., 1995. Metabolic implications of ammonia production in the ruminant. *Proc. Nutr. Soc.* 54, 549-563
- Reynolds C.K., Aikman P.C., Lupoli B., Humphries D.J., Beever D.E., 2003. Splanchnic metabolism of dairy cows during the transition from late gestation through early lactation. *J. Dairy Sci.* 86, 1201-1217
- Rhine E.D., Sims G.K., Mulvaney R.L., Pratt E.J., 1998. Improving the berthelot reaction for determining ammonium in soil extracts and water. *Soil Sci. Soc. Amer. J.* 62, 473-480