

URINARY METABOLITES OF CATTLE AT TWO LEVELS OF FEED INTAKE

H.J. Anzola¹, R.R. Scott¹, F.N. Owens² and R. Raney³

Story in Brief

To estimate the nutritive status of cattle, urinary purine output might be used. Four dairy steers fitted with ruminal and duodenal cannulas were fed a concentrate diet at two levels of intake (1.5% and 2.0% of body weight). Total urine was collected and analyzed for creatinine and purine content. Urinary creatinine excretion varied widely between animals and day to day, with a residual coefficient of variation over 36%. This renders its concentration useless as an index of daily urine output in cattle. Purine excretion was only 6% of duodenal RNA-N flow. This can be partly attributed to oxidation of purines to uric acid and allantoin plus incomplete hydrolysis of wet urine samples. Future research is needed to ascertain consistent urinary metabolite ratios which might be employed to predict the nutrient status of cattle.

(Key Words: Cattle, Urine, Creatinine, Purines, Intake)

Introduction

Nucleic acids in the urine are derived from microbial synthesis in the rumen whereas creatinine is derived from muscle metabolism. As daily creatinine output in urine by humans is relatively constant, concentrations of other urinary metabolites relative to creatinine can be used to assess total excretion and, thereby, health status. For ruminants, if creatinine output were constant, the purine/creatinine ratio in urine might be used as an index of the amount of nitrogen flowing to the small intestine because purine excretion should be proportional to microbial protein synthesis in the rumen. Excretion of nucleic acid derivatives in urine has been found to be correlated with the flow of nucleic acids to the small intestine (Topps and Elliot, 1965). Duodenal microbial nitrogen flow, in theory, is directly related to the protein synthesis in the rumen (McAllan, 1982). Therefore a relationship between urinary nucleic acids and duodenal microbial nitrogen flow would be expected. In buffalo calves, the variation in urinary creatinine was found to be large between animals and also from day to day within animals (Chetal et al., 1975). They concluded that urinary creatinine cannot be used as an index in nutrient balance studies or for evaluating ruminant nutrition in general. The purpose of this study was to relate urinary metabolites with feed intakes and to determine if creatinine excretion was constant for cattle.

Materials and Methods

Four steers fitted with duodenal and ruminal cannulas were confined in metabolic crates for total urine collection. They were fed

¹Graduate Assistant ²Regents Professor ³Lab Technician

a 15% crude protein 80% concentrate diet at two levels of intake (1.5% and 2.0% of body weight) in a cross-over design. Chromic oxide was used as an indigestible marker. Duodenal and fecal samples were collected over a period of 6 days. Dry matter, ash, chromic oxide, and purine-nitrogen on feed, duodenal, and fecal samples were analyzed. Urinary creatinine was measured with a Beckman II creatinine analyzer and purine analyses were conducted using the Zinn and Owens (1986) procedure on lyophilized urine samples.

Results and Discussion

Mean creatinine flow (g/d) was similar at both levels of intake (2.45 ± 0.92 and 2.58 ± 1.03 for 2.0% and 1.5% intake levels), while urinary purine concentrations were 0.40 and 0.33 g N/day. Hence, RNA to creatinine ratios at the two intake levels were 0.18 and 0.15 (Table 1). Averaged across all animals diets and dates, and expressed proportionally to weight, urinary creatinine outputs were 29.6 and 31.2 mg/kg^{.75}/day at the 2.0% and 1.5% levels of feed intake. This creatinine output was lower than the 53.2 mg/kg^{.75}/d measured with lambs solely nourished by intragastric infusion (Hovell et al., 1983). In their experiment, daily creatinine excretion was not constant but varied between 2-d means with a 13.1% standard deviation of the overall mean. Creatinine flow in our study also varied from day to day (S.E. = 0.79 to 1.57; Table 2). These data agree with results of Chetal et al. (1975) who found large variability between individual buffalo and from day to day within each animal ($P < .01$). Willis (1978), found values of creatinine output, by gilts, ranged from 50 to 120 mg/kg^{.75}/day (S.E. 7.6), with a mean (85 mg/kg^{.75}/d) considerably higher than ours. In man, daily excretion of creatinine is so constant at 20 to 26 mg/kg that it is used as a base for measurement of other urinary constituents. Reasons for species differences are not clear. Van Niekerk et al. (1963) reported that starvation in sheep, similar to man, reduced urinary creatinine output. Concentrations of ruminal metabolites in urine also are variable. Topps and Elliot (1965) fed sheep a high energy diet and found urinary output of uric acid plus allantoin nitrogen was 544 + 260 mg/d. Urinary RNA flow in our trial (Table 1) was 0.40 ± 0.12 and 0.33

Table 1. Relationship between intake levels, urine metabolite flows, RNA/creatinine ratio and duodenal RNA-N recovered in the urine.

VARIABLES	-----INTAKE LEVEL %-----	
	2.0	1.5
Creatinine flow (g/day)	2.45 ± 0.92^a	2.58 ± 1.03^a
RNA flow (g N/day)	0.40 ± 0.12^a	0.33 ± 0.08^a
RNA/Creatinine ratio	0.18 ± 0.07^a	0.15 ± 0.06^a
% Duod RNA-N recovered in urine	6.30 ± 3.90^a	5.40 ± 1.90^a

Means in a row with different superscripts differ ($P < .05$)

Table 2. Urinary creatinine flow (g/day).

Steer	Weight,kg	Day		
		1	2	3
1	377	0.84	3.66	3.01
2	430	2.62	3.70	5.37
3	318	1.39	2.14	1.93
4	284	2.12	1.29	2.14
Mean		1.74	2.70	3.11
S.E.		0.79	1.19	1.57

S.E. = Standard Error.

+ 0.08 g N/d at the 2.0% and 1.5% intake levels. These values, as a fraction of purine-N at the duodenum, were only 6.3 ± 3.9 and 5.4 ± 1.9 at the 2.0% and 1.5% intake levels (Table 1). A simultaneous study with cattle grazing native range gave a mean allantoin/creatinine ratio of .265 which would calculate to equal 0.1 g N/day excreted as allantoin (Campbell and Pitts, 1986). Condon and Hatfield (1971) reported that 43% of ruminally produced nucleic acid N was excreted as purine derivatives in the urine. As only 6% of duodenal purine was recovered in urine as purine in our study, this suggests that purines were oxidized to uric acid and allantoin prior to excretion though composition varies with species. Incomplete hydrolysis of lyophilized urine samples with perchloric acid (Zinn and Owens, 1986) might also reduce nucleic acid recovery.

Because of the lability of creatinine, it is advisable to analyze fresh samples (Beckman 1978). Concentrated hydrochloric acid, diluted 1:1 with water, should be added to the total volume of urine to prevent nitrogen and urinary RNA losses. It is essential for the urine samples to be completely dry before analyzing for purines, as hydrolysis will be incomplete if the sample is wet (Zinn and Owens, 1986). One alternative to freeze drying would be to dry urine samples directly in screw-cap culture tubes in a forced air oven. Future research is needed to detect some consistent urinary metabolite to use as a baseline when calculating nutrient ratios for cattle as creatinine excretion in ruminants is quite variable.

Literature Cited

- Beckman. 1978. Creatinine Analyzer 2 Operating Manual.
 Campbell, G. and J. Pitts. 1986. Relationship between microbial nitrogen flow at the duodenum and the allantoin/creatinine ratio in urine from growing steers. Typed paper, Fall 1986. O.S.U.
 Chetal, V. et al. 1975. On the variation of urinary creatinine in buffalo calves and the effect of dietary protein intake on urinary creatinine, creatinine-nitrogen ratio and creatinine coefficient. J. Agric. Sci. 84:1-5.

- Condon, R.J. and Hatfield, E.E. 1971. Metabolism of exogenous nucleic acids by ovines. *Fed. Proc. Fed. Am. Soc. Exp. Biol.* 30, 402.
- Hovell, F.D. Deb. et al. 1983. The effect of changes in the amount of energy infused as volatile fatty acids on the nitrogen retention and creatinine excretion of lambs wholly nourished by intragastric infusion. *Brit J. of Nutr.* 50:331-343.
- McAllan, A.B. 1982. The fate of nucleic acids in ruminants. *Proc. Nutr. Soc.* 41:309-317.
- Topps, J.H. and R.C. Elliot. 1965. Relationship between concentrations of ruminal nucleic acids and excretion of purine derivatives by sheep. *Nature.* 205:498-499.
- Van Niekerk. et al. 1963. A study of some of the conditions affecting the rate of excretion and stability of creatinine in sheep urine. *J. Nutr* 79:373-380.
- Willis, G.M. 1978. Influence of protein intake, energy intake and stage of gestation on growth, reproductive performance, nitrogen balance, creatinine excretion of the gravid gilt. M.S. Thesis. Thesis Oklahoma State University, Stillwater.
- Zinn, R.A. and F.N. Owens. 1986. A rapid procedure for purine measurement and its use for estimating net ruminal protein synthesis. *Can. J. Sci.* 66:157.