

TRACER STUDY OF METHIONINE METABOLISM IN SHEEP

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Introduction

In many situations methionine is the first limiting amino acid in wool producing sheep (Buttery and Foulds, 1985). Information on the metabolic fate of methionine following administration of different doses is limited. In this report, labelled methionine (xMet) was used to study the metabolism of different sulphur-containing compounds in hay-fed sheep.

Materials and Methods

Three rumen and duodenal cannulated Norwegian sheep weighing 87.5 ± 3.5 kg were housed in metabolic cages. They were fed 1,180 g pasture hay once a day in the morning. For five consecutive days amino acid mixtures containing 5 or 15 g each of D,L-methionine, -lysine and -threonine were prepared in rumen fluid and administered via rumen cannula into each of the sheep at the time of feeding. On the fifth day the animals received a tracer dose of xMet (35 S-D,L, methionine) either intravenously (i.v.) or intraruminally (i.r.), simultaneously with the i.r. administration of the mixture of cold amino acids. In all experiments rumen, duodenal and blood samples were collected at hourly intervals for 2 h, then every 2 h for 8 h. Blood samples, urine and faeces were collected for 6 days. Samples were treated according to standard procedures and kept frozen prior to analyses. For amino acid analyses HPLC was used. Radioactivity was counted in a Hewlett Packard model 3310 liquid scintillation counter.

Flow rates of methionine into and out of rumen and plasma pools were calculated assuming non-steady state conditions (Shipley and Clark, 1972).

Results

Labelling of plasma proteins was much more rapid after i.v. than after i.r. xMet injection, while activity in rumen inorganic sulphate was highest

after i.r. xMet injection.

After i.v. as well as i.r. xMet injection, the load of cold methionine had little effect on activity (expressed as % of dose of xMet/l) in the following fractions: rumen amino acids and inorganic sulphate, duodenal amino acids, plasma proteins, amino acids and inorganic sulphate. This suggests greater flow of methionine out of the rumen, and greater incorporation of methionine into plasma proteins at the 15 g than at the 5 g dose level, and that the transfer rates were proportional to dose rates.

The estimated flow rates into and out of the plasma methionine pool were 0.10 ± 0.015 and 0.10 ± 0.013 g/day respectively at a daily dose level of 5 g, and 0.24 ± 0.002 and 0.25 ± 0.005 g/day at a daily dose level of 15 g methionine.

Following i.r. injection of xMet, there was substantial labelling of amino acids in duodenum, and of proteins and amino acids in blood plasma.

After i.v. injection of xMet, incorporation of label into plasma proteins was rapid and remained at a level higher than 90 % of the labelled dose.

After i.r. injection of xMet, most of the label in plasma was initially present in inorganic sulphate, presumably due to degradation of methionine in the rumen. After 24 h most of the label was present in plasma proteins, and this fraction increased to about 70 % of the injected dose at 50 h post i.r. injection for both dose levels.

Excretion of activity in faeces and urine was followed for 6 days. Following i.v. xMet injections, the percentages of the dose recovered were about equal for urine and faeces, while i.r. xMet injections resulted in higher excretion with the urine than with faeces. The total amount excreted increased with increasing dose, but total amount of activity excreted was much higher after i.r. than after i.v. xMet injection.

Discussion

The results agree with earlier work, indicating

that substantial amounts of orally administered methionine leaves the rumen either as free amino acid or incorporated into rumen microbes (Bird and Moir, 1972; Champredon et al., 1973; Hidiroglou and Zarkades, 1976).

The percentage of plasma label present in proteins after 24 h was similar for the 5 and 15 g dose (42%), as was the excretion of activity in urine (46 vs 52%).

In an associated study, the change in average plasma concentrations of an amino acid with dose administered *i.r.* suggested that the requirement for methionine in the feed was met by 10-15 g/d (Cottle and Velle, 1989). The patterns of labelling suggest that one requirements are met, the excess methionine is excreted in both faeces and urine, with greater degradation occurring in the rumen.

In the present study the sheep requirement for methionine at the duodenum was probably about 4.3 g/d (0.15 g/kg^{0.75}/d, Lewis and Mitchell, 1976). Following the 5 and 15 g doses about 0.7 and 3.5 g/d respectively of methionine flowed to the duodenum intact. Thus 3.6 and 0.8 g/d of the daily requirement respectively had to be met from microbial protein (or amino acid absorbed across the rumen or synthesized in tissue). The estimated microbial supply to methionine on this diet was 1.1 g/d, assuming microbial supply equals 7.8 xMet and 2% of microbial protein consists of methionine. The requirements for methionine were covered by methionine outflow and microbial methionine following a 15 g dose of methionine. The 5 g dose did not meet requirements (assessed from plasma concentrations), suggesting that the transfer of methionine to plasma via the rumen or synthesis of methionine from inorganic sulphate was limited.

There is some indication that differential metabolism of D- and L-methionine may occur in the rumen. D,L-methionine gave only 63% of the growth on microbial cells *in vitro* compared with

that from L-methionine (Kahlon et al., 1975). Thus there are unknown errors associated with using ³⁵S-D,L-methionine to trace the metabolism of DL-methionine.

(Key Words: Methionine, Sheep, Metabolism)

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