Available online at www.sciencedirect.com
ScienceDirect
Livestock Science 111 (2007) 181–192
LIVESTOCK SCIENCE
www.elsevier.com/locate/livsci

Review article

Urinary purine derivates excretion as an indicator of in vivo microbial N flow in cattle: A review

B.M. Tas *, A. Susenbeth

Institute of Animal Nutrition and Physiology, Faculty of Agricultural and Nutritional Science, Christian Albrechts University of Kiel, D-24118, Kiel, Germany

Received 5 January 2007; received in revised form 9 May 2007; accepted 10 May 2007

Abstract

Microbial protein flow to the duodenum may be regarded as the most important and sensitive indicator to optimise rumen metabolism in high-yielding dairy cows. In this review, the methodology and the sources of variation to estimate the duodenal microbial N flow with urinary excretion of purine derivatives (PD) as a non-invasive method is discussed. The urinary PD excretion was linearly related with the amount of purine bases (PB) infused in the abomasum or duodenum, but the recovery of PB in urine differed between experiments. The main sources of variation in the relationship between microbial N flow and urinary PD excretion are dietary contribution of nucleic acids to duodenal flow, varying N:purine ratio in duodenal digesta, differences in intestinal digestibility of nucleic acids and infused PB, and endogenous contribution of PD to urinary excretion. The recycling of PD to the rumen is negligible, and does not explain the incomplete urinary recovery of PD. A large proportion of the total PD is excreted as allantoin in urine. In some experiments this proportion was constant, whereas in others it varied with diet or physiological state of the animal. The excretion of PD in milk is not a suitable indicator of microbial N flow, due to mammary purine catabolism to uric acid and due to the strong positive correlation between milk allantoin excretion and milk yield. Instead of total urine collection, the molar ratio between urinary PD and creatinine can be used to estimate microbial N flow. However, a substantial between-animal variation in this ratio was found, and effects of changes in energy balance of dairy cows on urinary creatinine excretion should be determined. The urinary excretion of total PD and of allantoin provided lower estimates of duodenal microbial N flow than with measurements in the omasum or duodenum, but they closely reflected the changes observed with these measurements.

Keywords: Purine base; Purine derivative excretion; Microbial N; Allantoin; Cattle

1. Introduction

More than half of the high energy and protein requirements of high-yielding dairy cows is supplied by volatile fatty acids (VFA) produced by rumen microbes and by the flow of microbial protein to the duodenum. To increase energy intake and optimise rumen fermentation, diets are fed with a minimum of structural carbohydrates and with high amounts of non-structural carbohydrates. A large proportion of non-structural carbohydrates is rapidly degraded by rumen microbes, and this increases VFA production and microbial protein

* Corresponding author. Tel.: +49 431 880 2538; fax: +49 431 880 1528.
E-mail address: tas@aninut.uni-kiel.de (B.M. Tas).

1871-1413/$ - see front matter © 2007 Elsevier B.V. All rights reserved.
doi:10.1016/j.livsci.2007.05.010

synthesis, alters VFA molar proportions, reduces rumen pH, and increases VFA absorption (Dijkstra, 1994; Bannink et al., 2006). These diets with a low structural carbohydrate content lead to increased passage rates, increased efficiency of microbial protein synthesis and hence lead to increased supply of metabolizable energy and intestinal digestible protein (e.g. Bach et al., 2005; Firkins et al., 2006). However, a minimum of structural carbohydrates is required to maintain rumen function and to prevent rumen acidosis (Allen, 1997; Zebeli et al., 2006). Microbial protein flow to the duodenum may be regarded as the most important and sensitive indicator to optimise rumen metabolism in high-yielding dairy cows.

The measurement of microbial protein flow in vivo requires surgically cannulated animals, which is expensive, increases animal care concerns, and may affect dry matter intake and milk yield. Urinary excretion of purine derivatives is a non-invasive method that has the potential to be used as an on-farm method. The urinary excretion of purine derivatives (PD: allantoin, uric acid, hypoxanthine, and xanthine) appears to be a reliable method to estimate the microbial N flow to the duodenum. The principle is that the duodenal flow of nucleic acids and their derivatives is mainly of microbial origin, which are to a large extent digested and absorbed in the small intestine, purine bases are catabolized to their PD, and excreted in the urine (Topps and Elliot, 1965). Therefore, microbial N flow can be estimated from the quantitative excretion of PD in urine.

This paper aims to review the opportunities of urinary PD excretion to estimate microbial N flow in cattle, with reference to sheep. This review focuses on the methodology and the sources of variation to estimate the duodenal microbial N flow with urinary excretion of PD. The duodenal microbial N flow estimated with urinary PD excretion is compared with in vivo measurements, and relationships with flow of purine bases (PB) are determined. The method for a quantitative estimation of microbial N flow based on urinary PD excretion is described and its sensitivity and accuracy are discussed.

2. Microbial N flow to the duodenum

2.1. Nucleic acids flow

Microbial protein is synthesized in the rumen. Rumen microbes contain a high content of nucleic acids, composed of nucleotides. Each nucleotide consists of a pentose sugar, a phosphate group, and a nitrogenous heterocyclic base (either a purine or a pyrimidine). The main source of duodenal flow of nucleic acids is from microbial origin (Kanjanapruthipong and Leng, 1998). Other sources of nucleic acids and derivatives are the diet, saliva and cells from rumen epithelium. In general, the nucleic acid content in feed is lower than in rumen microbes. Free nucleic acids and derivatives are almost completely degraded in the rumen (McAllan and Smith, 1973; Storm et al., 1983), but a significant proportion of 0.15 of the duodenal flow of RNA was not from microbial origin. Dietary nucleic acids can be protected by association with resistant cell structures. The rumen degradability of PB was for a number of feeds high and the rumen escape low (Perez et al., 1996a; Djouvinov et al., 1998), although a substantial proportion of PB of fish meal, rich in nucleic acids, could escape the rumen and contribute to duodenal flow with 0.25 to 0.34 g/g (Perez et al., 1996b; Djouvinov et al., 1998). In continuous culture fermenters, the dietary contribution to purine flow was low with 0.017 mol/mol, and was not significantly affected by diet (Calsamiglia et al., 1996). In sheep, dietary purines accounted for 0.13 to 0.27 of the total duodenal purine flow (Perez et al., 1997a). In heifers, estimated with ^{15}N ammonium sulfate continuously infused in the rumen, Vicente et al. (2004) found a substantial contribution of 0.34 of non-microbial PB to the duodenal flow, whereas Hristov et al. (2005) found a lower contribution of 0.035 in the liquid digesta phase and of 0.197 in the solid digesta phase. Moreover, the contribution of these dietary purines to urinary PD excretion was small (Hristov et al., 2005). Although data on the contribution of non-microbial PB to the duodenal PB flow are scarce and vary, this contribution may lead to an overestimation of the duodenal microbial N flow if not corrected for. Sheep saliva contains a significant concentration of allantoin and uric acid (Chen et al., 1990a). However, allantoin incubated in vitro with rumen fluid was degraded (Chen et al., 1990a), and disappeared to a large extent after 48 h of incubation (Prasitkusol et al., 2002). No effect of allantoin infused in the rumen of sheep or steers on urinary PD excretion was found (Chen et al., 1990a). Similarly, intrajugular infusion of allantoin increased slightly the concentration of allantoin in saliva, but did not affect the urinary PD excretion (Surra et al., 1997). The magnitude of this salvage pathway is discussed later. Microbes associated with the rumen epithelium are well adapted to degrade epithelial debris, including nucleic acids (Chen et al., 1990a). Therefore, the contribution of saliva and epithelial cells to the duodenal flow of nucleic acids and derivatives is negligible. Although dietary nucleic acids might escape rumen degradation, the duodenal flow of nucleic acids is predominantly of microbial origin.