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Invited commentary

Utilization of essential amino acids synthesized in the intestinal microbiota of monogastric mammals

The amino acid supply of ruminants eating poorly digestible low-protein diets depends on the microbial activities in their forestomachs. However, it is insufficiently understood whether and to what extent microbially synthesized essential amino acids (EAA) might be used to support growth and protein homeostasis of the monogastric host. In various monogastric mammals such as rats (Torrallardona *et al.* 1996*a*, 1996*b*), pigs (Metges *et al.* 1996; Backes *et al.* 2002; Torrallardona *et al.* 2003*a*, 2003*b*), man (Metges *et al.* 1999*a*, 1999*b*; Millward *et al.* 2000) and now also rabbits, as a paper in the present issue of *British Journal of Nutrition* demonstrates (Belenguer *et al.* 2005), microbial synthesis of EAA in the gastrointestinal tract (GIT) and their utilization have been demonstrated. Experimental evidence comes from the '^{15}N labelling paradigm' (Metges, 2000), which involves the administration of an ^{15}N-labelled inorganic N source (e.g. $^{15}NH_4Cl$, ^{15}N[urea]) as a precursor of microbial amino acid-N. Since lysine and threonine do not undergo transamination in mammalian tissues (Torrallardona *et al.* 1996*a*), the appearance of ^{15}N-labelled lysine and threonine in the host tissues indicates their microbial origin. Enrichment of ^{15}N-labelled lysine and threonine is very low (below 0.1 atom% excess ^{15}N) and its detection requires a highly sensitive and precise mass spectrometric technique (e.g. Metges *et al.* 1996; Metges & Petzke, 1997; Belenguer *et al.* 2005).

With regard to the biological significance of the intestinal microbial amino acid contribution there are still open questions because the values given in the literature so far are based on many assumptions. First, it would be important to know whether microbial amino acids contribute to the amino acid homeostasis of the host in a net way. This would require that the material utilized to produce microbial EAA be of no further value for the body. Quantification is further complicated by the constant N recycling in the gut (Fuller & Reeds, 1998; Metges *et al.* 1999*a*) and the broad variety of N sources used by the microbiota in the GIT (Metges & Loh, 2003). On the other hand, it has been shown that carbohydrates that cannot be degraded by mammalian enzymes are used to produce microbial amino acids utilized by the host (Torrallardona *et al.* 2003*b*).

Second, it is still not certain at which intestinal sites microbial amino acid absorption occurs, and there are differences in the various monogastric species (Metges, 2000). Further, quantification of the microbial amino acid contribution relies on the product–precursor relationship. However, the nature of the pool of microbial lysine absorption to be sampled is unclear (e.g. lumenal microbial protein, microbial peptides or free amino acids, bacteria adherent to the mucus overlaying the villi; Metges, 2000). Among monogastric animals (pig, rabbit, rat, mouse) and man there are distinct differences in the anatomical and physiological properties of the intestine (Metges & Loh, 2003). Rats are coprophagic, which also occurs in the young pig if not prevented by housing in metabolic cages. As compared with man, mice and rats have large caeca relative to the overall size of their GIT. One of the major differences between the GIT microbiota in man and pigs is the high number of bacteria in the porcine small intestine and stomach (pars oesophagea). Torrallardona *et al.* (2003*a*,*b*) demonstrated by digesta exchange experiments that the majority of microbial lysine is absorbed in the small intestine of the pig. Earlier results in pigs with ileo-rectal anastomosis also suggest absorption of microbial amino acids by the small intestine (Metges *et al.* 1996). That small intestinal absorption might be the major route for microbial amino acids also in man comes from studies with ileostomates (Metges *et al.* 1999*a*). The ratio of ^{15}N enrichment in plasma free lysine and ileal microbial lysine in ileostomates was higher than the ^{15}N enrichment in plasma free lysine and faecal microbial lysine in subjects with an intact GIT. In contrast, it appears that rats cannot utilize microbial amino acids directly via absorption from sites of synthesis in the GIT, and incorporation of microbial amino acids in body proteins entirely relies on coprophagy (Torrallardona *et al.* 1996*b*). Whether this is because of no or very low microbial amino acid synthesis in the small intestine, or low proteolytic activity in the lower parts of the small intestine, is unclear. Belenguer *et al.* (2005), in their study in the present issue, compared the ^{15}N enrichment of lysine incorporated in liver protein of rabbits prevented to re-ingest caecotrophes by neck collars with that of unrestricted control animals, concluding that 97 % of microbial lysine utilization in the rabbit originates from the caecotrophy process and thus direct intestinal absorption is very low. Whether uptake of caecotrophes is crucial for the EAA supply for growth in rabbits remains to be determined.

Third, it is incompletely understood whether different dietary or physiological conditions affect the availability of microbial amino acids to the host (Metges & Loh, 2003). Dietary constituents such as non-digestible oligosaccharides affect intestinal microbiota composition and thus perhaps also microbial amino acid synthesis. Also, low-protein diets known to alter amino acid utilization of the gut might influence systemic availability of microbial amino acids in pigs (Van Goudoever *et al.* 2000; Van der Schoor *et al.* 2001). We have recently shown that minipigs fed a diet low in lysine do not adapt by showing an enhanced availability of microbial lysine to the extrasplanchnic tissues (Backes *et al.* 2002). This is presumably because microbial lysine continues to be used for splanchnic protein synthesis with high priority.

In conclusion, the available evidence indicates that microbial lysine absorption in man and pigs is located in the small intestine, but requires re-ingestion of faecal or caecal microbial protein in rats and rabbits. Whether intestinal microbial EAA make a net addition to meet the metabolic EAA demand is still unclear.

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