*Escherichia coli oppE, met, trp, his, pro* mutant (strain SS3240), defective in tripeptide uptake and resistant to triornithine ($orn_3$), was transformed with a plasmid library of strain $B_14$ chromosomal DNA, and plated out on minimal medium supplemented with methionine, proline, histidine, and the tripeptide L-lysyl-tryptophanyl-lysine. Colonies which appeared on the selective plates were then tested for $orn_3$ sensitivity and tryptophan prototrophy, to confirm that the recombinant DNA clones carried gene(s) involved with oligopeptide transport. Plasmid DNA was extracted from four transformants possessing the appropriate phenotype (i.e. $orn_3$-sensitivity and tryptophan auxotrophy) and used to retransform SS3240. All plasmid DNA preparations transformed SS3240 to an *oppE*$^+$ phenotype, and one of the plasmid clones (pANS1000) was selected for further examination. A 4.0 kilobase *Bam*HI-*Pst*I fragment from pANS1000 was subcloned in pBluescript SK$^+$ to generate pANS1001, and *E. coli* SS3240/pANS1001 transformants were shown to possess an *oppE*$^+$ phenotype. Tri-alanine ($Ala_3$) uptake rates were measured using procedures described by Payne and Bell [1] and determined to be 51.4, 7.2, 29.2 and 54.1 nmol $Ala_3$ $min^{-1}$ (mg protein)$^{-1}$ for cultures of *E. coli* SS320 (*oppE*$^+$), SS3240 (*oppE*), SS3240/pANS1000 (*oppE*$^+$), and SS3240/pANS1001 (*oppE*$^+$), respectively. Nucleotide sequence analysis of pANS1001 identified an open reading frame which possesses considerable homology (~59% identity) with the RprX protein of *Bacteroides fragilis*, which belongs to a family of histidine protein kinase receptors (e.g. EnvZ and PhoA) found in a number of eubacteria. The RprX protein modulates the expression of genes encoding outer membrane porins such as OmpF and OmpC [2], which are known to influence peptide uptake in *E. coli* [3]. Our findings to date suggest that *P. ruminicola* possesses a gene or genes with functions analogous to the *oppE* locus in *E. coli*, and that OppE-like gene products might be involved in coordinating porin expression.

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## HYDROGEN TRANSFER

### Alternative hydrogen sink pathways in hindgut fermentation.

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Besides methanogenesis (M), non assimilatory sulphate reduction and/or reductive acetogenesis (RA) have been identified as major pathways of metabolic hydrogen disposal in hindgut fermentation for a number of animals, based mainly on stoichiometry of metabolic hydrogen recovery [1]. These alternative hydrogen sinks to methanogenesis do not function in the rumen, although the bacteria capable of using them have been isolated.

This report summarizes experiments in our laboratory over the past ten years re-

lated to attempts to induce RA in the rumen in vitro and in vivo, and understanding the relative importance of RA and M in the rabbit caecum.

The affinity for $H_2$ of RA is lower than that of M but the importance of RA in the hindgut has been related to the higher amounts of free amino acids, mucins and bile salts, inducive and/or inhibitory for RA and M respectively. Batch incubations of sheep rumen contents with these compounds added did not stimulate RA although selective inhibition of M was demonstrated, accompanied however by increased propionate production [2,3]. Addition of *Peptrostreptococcus productus* ATCC 35244 [4] or methanol [5] together with 2-bromoethane sulfonic acid (BES) to sheep rumen contents *in vitro* increased acetate production through RA. Such experiments *in vivo* are excluded however because of the very fast adaptation of the rumen to BES [5]. Use of a similar combination with a bacteriocin-like inhibitor of M resulted in similar results in vitro and in vivo (Nollet *et al*, in preparation). Also, direct introduction of frozen and thawed cattle hindgut contents to a sheep rumen did not affect rumen fermentation stoichiometry in vitro [5].

RA is a major characteristic of the caecal fermentation in young suckling rabbits, producing significant amounts of VFA with little or no M. Subject to a litter effect, RA is replaced gradually and partially by M with the increasing intake of solid feed [6]. However, caecal RA was clearly present in non-fasted rabbits only [7]. In contrast to the sheep caecum [8], monensin increased non fasted rabbit caecal M *in vitro*, accompanied by a decrease in butyrogenesis. Depression of M was observed with BES (Piattoni *et al*, in preparation). The results suggest that rabbit caecal RA is inhibited by monensin, although such an effect was not apparent from viable counts of bacteria active in RA. It is suggested that caecal bacteria capable of RA are different in sheep and rabbits.

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## Characteristics of $H_2/CO_2$ metabolism in acetogenic bacteria from the human colon.

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In the human colon, the fermentation of substrates which are not absorbed in the upper- digestive tract, leads to the production of short chain fatty acids (mainly acetate, propionate and butyrate) and