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## Intestinal ciliates and their endosymbionts from the cockroach hindgut: evolutionary aspects

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was positive ( $r=0.65$   $P<0.01$ ). Values declined from  $14.4 \pm 2.79$  (0h) to  $6.8 \pm 1.98 \mu\text{g g}^{-1}$  rumen fluid (16h). The abundance of Eucarya increased when the cows were returned to the normal diet, reaching  $40.3 \pm 5.5 \mu\text{g RNA per g rumen fluid}$  at 72h. Archaea decreased following the induction of acidosis, from  $1.4 \pm 0.4$  to  $0.4 \pm 0.07 \mu\text{g g}^{-1}$  rumen fluid at 0 and 72h respectively. The population encompassed by the families *Methanoplanaceae*, *Methanomicrobiaceae* and *Methanocorpusculaceae* declined rapidly during the first 16h. ( $0.54 \pm 0.07$  to  $0.04 \pm 0.01 \mu\text{g g}^{-1}$ ) and did not recover. In contrast the *Methanobacteriaceae* increased after the perturbation (24.3% of the total Archaea at 0h and 79.8% at 72h). These values were positively correlated with Eucaryal biomass ( $r = 0.7$   $P<0.01$ ). *Desulfovibrio* sp. SSU rRNA accounted for an average  $1.2\% \pm 0.59$  of the total SSU rRNA abundance. There was a strong positive correlation between the total amounts of *Desulfovibrio* and *Methanobacteriaceae* SSU rRNA ( $r=0.51$   $P<0.01$ ) throughout the observation period.

**Intestinal ciliates and their endosymbionts from the cockroach hindgut: evolutionary aspects.** AHAM Van Hoek, TA Van Alen, VSI Sprakel, JHP Hackstein, GD Vogels (*Department of Microbiology and Evolutionary Biology, University of Nijmegen, NL-6525 ED Nijmegen, The Netherlands*)

Anaerobic ciliates occur in the hindgut of many cockroach species, but only in those cockroaches that also are host to intestinal methanogens. Such ciliates contain, without exception, endosymbiotic methanogenic bacteria [1]. Although all of these

anaerobic ciliates seem to be related to *Nyctotherus* sp. behavioural studies (e.g. galvanotaxis) and the morphology of the methanogenic endosymbionts suggest the presence of different protists in the various cockroach species. The rDNA genes of individual ciliates and their methanogenic endosymbionts were analysed in order to study the symbioses. DNA sequencing and restriction analysis of PCR-amplified rDNA genes showed that the ciliates isolated from the various cockroach hosts did differ significantly. The endosymbiotic methanogenic bacteria of these ciliates, also, proved to be different. Thus, in contrast to the situation in aphids with their eubacterial endosymbionts [2], the phylogeny of the anaerobic ciliates and their cockroach hosts [3] does not match. Since the methanogenic endosymbionts also differ substantially, it seems reasonable to conclude that the symbioses between methanogenic bacteria, ciliates, and cockroaches evolved more than once.

1. Hackstein JHP, Stumm CK (1994) *Proc Natl Acad Sci USA* 91, 5441-5445
2. Moran NA (1996) *Proc Natl Acad Sci USA* 93, 2873-2878
3. Kambhampati S (1995) *Proc Natl Acad Sci USA* 92, 2017-2020

## MICROBIAL GENETICS

**Transformation of *Butyrivibrio fibrisolvens* strains with pBH<sub>1</sub> plasmid.** J Kopečný<sup>1</sup>, K Fliiegerová<sup>1</sup>, K Gregg<sup>2</sup> (<sup>1</sup>*Institute of Animal Physiology and Genetics, Czech Academy of Sciences, Prague 10, Uhořinives, 104 00, Czech Republic;* <sup>2</sup>*Institute of Biotechnology, University of New England, Armidale, NSW 2351, Australia*)