



Contents lists available at ScienceDirect

## Animal Feed Science and Technology

journal homepage: [www.elsevier.com/locate/anifeedsci](http://www.elsevier.com/locate/anifeedsci)



### Using microbial fatty acids to improve understanding of the contribution of solid associated bacteria to microbial mass in the rumen

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#### ARTICLE INFO

##### Article history:

Received 17 October 2007

Received in revised form 19 September 2008

2008

Accepted 24 September 2008

##### Keywords:

Microbial markers

Odd and branched chain fatty acids

Purine bases

Rumen bacteria

#### ABSTRACT

This study sought to distinguish liquid-(LAB) and detached (SAB<sub>1</sub>) and undetached (SAB<sub>2</sub>) solid-associated bacteria through their fatty acid (FA) and purine base (PB) profiles. Fatty acids and PB were also evaluated as internal microbial markers for estimating microbial biomass associated with rumen particles. Four merino rams fitted with rumen cannulae and fed dehydrated alfalfa pellets provided rumen contents. In 3 consecutive weeks, rumen contents were collected and samples of LAB and SAB<sub>1</sub>, total rumen content (TRC), washed rumen particles (WRP) and rumen particles after SAB<sub>1</sub> extraction (ERP) were obtained and analysed for PB and FA. The SAB<sub>2</sub> biomass composition was estimated from the non-NDF organic matter (OM) remaining in ERP. The concentration of total SAB biomass in particles was estimated using both PB and odd and branched-chain fatty acids (OBCFA). Concentrations of PB and

**Abbreviations:** CP, crude protein; ERP, extracted rumen particles; FA, fatty acids; LAB, liquid associated bacteria; NAN, non-ammonia N; NDFom, neutral detergent fibre not assayed with a heat stable amylase and expressed exclusive of residual ash; OBCFA, odd and branched-chain fatty acids; OM, organic matter; PB, purine bases; SAB<sub>1</sub>, detachable solid associated bacteria; SAB<sub>2</sub>, undetachable solid associated bacteria; TRC, total rumen content; WRP, washed rumen particles.

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OBCFA were highly correlated among the different rumen fractions. Marked differences between LAB and SAB populations occurred with LAB having higher PB content, lower FA content and a higher proportion (g/100 g fatty acids) of OBCFA than did SAB. The chemical composition of SAB<sub>1</sub> and SAB<sub>2</sub> was similar, except for the 15% higher crude protein content of the latter. The concentration of OBCFA (mg/g microbial OM) did not differ between bacterial fractions. The PB/OBCFA ratio (mg/mg) was higher in LAB (2.08) than in SAB (0.94). The ratio between branched-chain and odd-linear-chain FA was higher in LAB (2.26) than in SAB (1.46). Extraction of PB and OBCFA from WRP with our SAB detachment procedure was 61% and 31%, respectively. Estimated SAB<sub>1</sub> and total SAB biomass (mg OM/g WRP) were 158 and 266, and 47 and 164, respectively, using PB and OBCFA as microbial markers. This study suggests that the OBCFA have potential as internal microbial markers in rumen ecosystem studies.

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