

# Screening for effects of plant extracts and active compounds of plants on dairy cattle rumen microbial fermentation in a continuous culture system

M. Busquet<sup>a</sup>, S. Calsamiglia<sup>a,\*</sup>, A. Ferret<sup>a</sup>, C. Kamel<sup>b</sup>

<sup>a</sup> *Departament de Ciència Animal i dels Aliments, Universitat Autònoma de Barcelona, Facultat de Veterinària (Edifici V), 08193 Bellaterra, Spain*

<sup>b</sup> *Axiss France, 01205 Bellegarde-sur-Valserine Cedex, France*

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## Abstract

Eight dual-flow continuous culture fermenters were used to study effects of plant extracts (Experiment 1) and active compounds of plants (Experiment 2) on rumen microbial fermentation. Each experiment consisted in two replicated periods of 9 days. Fermenters were fed 95 g dry matter (DM)/day in three feedings of a 600 g/kg (DM basis) alfalfa hay and 400 g/kg concentrate (178 g/kg crude protein, CP; 325 g/kg neutral detergent fibre, NDF diet), and maintained at constant temperature (38.5 °C), pH 6.4, and solid (0.05/h) and liquid (0.10/h) dilution rates. Both experiments included a negative control with no extract (CTR) and a positive control with monensin (MON). Treatments in Experiment 1 were: *Trigonella foenum graecum*, *Juniperus oxycedrus*, *Syzygium aromaticum* (CLO), *Anethum graveolens*, *Zingiber officinale*, and *Melaleuca alternifolia*. Treatments in Experiment 2 were: benzyl salicylate, anethol, carvacrol (CAR), cinnamaldehyde (CIN), eugenol, and D-carvone. During the adaptation period (i.e., days 1 through 7), samples for ammonia N and volatile fatty acid (VFA) con-

*Abbreviations:* ANE, anethol; ADF, acid detergent fibre; BEN, benzyl salicylate; CAD, cade oil; CAR, carvacrol; CIN, cinnamaldehyde; CLO, clove bud oil; CP, crude protein; CTR, control; CVO, D-carvone; DIL, dill oil; DM, dry matter; EUG, eugenol; FEN, fenugreek; GIN, ginger oil; LPep, large peptide; MON, monensin; NDF, neutral detergent fibre; S.E.M., standard error of the mean; SPepAA, small peptide plus aminoacid; TAN, tungstic acid soluble N; TCAN, trichloroacetic acid soluble N; TEA, tea tree oil; VFA, volatile fatty acids

\* Corresponding author. Tel.: +34 935811495; fax: +34 935811494.

E-mail address: [sergio.calsamiglia@uab.es](mailto:sergio.calsamiglia@uab.es) (S. Calsamiglia).

centrations were collected 2 h after feeding. On days 8 and 9, samples for VFA (2 h after feeding), and large peptide (LPep), small peptide plus amino acid (SPepAA), and ammonia N concentrations (0, 2, 4, 6 and 8 h after feeding) were also collected. During the adaptation period of Experiments 1 and 2, total VFA and ammonia N concentrations were not affected by treatments. During the first 6 days of fermentation in Experiments 1 and 2, MON resulted in lower acetate and higher propionate proportions compared with CTR. However, these differences disappeared after day 6. On days 6 and 7, CLO in Experiment 1 resulted in lower acetate, and higher butyrate, proportions compared with CTR. On day 7, the proportion of acetate was lower in CIN in Experiment 2 compared with CTR. After the adaptation period, CLO resulted in lower acetate, and higher propionate, proportions compared with CTR. The LPep N concentration was higher in CLO compared with CTR, suggesting that CLO reduced peptidolytic activity of rumen microorganisms. In Experiment 2, the LPep N concentration was lower in CAR, and MON resulted in lower SPepAA N concentrations and higher ammonia N concentrations compared with CTR, suggesting that MON stimulated deamination activity of rumen microorganisms. Results indicate that ruminal microbes may adapt to additives within 7 days. However, some plant extracts modified rumen microbial fermentation patterns and may allow manipulation of ruminal fermentation under current commercial practices.

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## 1. Introduction

Additives that modify rumen fermentation, such as organic acids, yeasts, enzymes and antibiotics, have been used to optimise performance in dairy and beef cattle production systems (Martin et al., 1999; Russell and Houlihan, 2003). Use of ionophore antibiotics improves performance and health in beef and dairy cattle (Chalupa, 1988; McGuffey et al., 2001). However, the use of antibiotics as feed additives in dairy cows is banned in the EU due to the fearing of appearance of residues in milk (Russell and Houlihan, 2003). For this reason, attention has recently shifted to natural antimicrobials as a safe means of modifying ruminal fermentation.

Plant extracts, including their essential oils, have been used for centuries in traditional medicine, for industrial applications, and as food preservatives due to their antimicrobial activity (Davidson and Naidu, 2000), and because most of them are considered safe for human consumption in the EU (Decision 1999/217/EC). The antimicrobial activity of plant extracts is attributed to a number of secondary metabolites, such as saponins (present in extracts of *Trigonella foenum graecum*, fenugreek, or *Yucca schidigera*), terpenoids (such as carvacrol, carvone, thymol, or terpinen-4-ol) and phenylpropanoids (such as cinnamaldehyde, eugenol, or anethol) compounds, present in the essential oil fraction of many plants (Helander et al., 1998). The effects of some of these plant extracts and their constituents on rumen fermentation have been documented (Hristov et al., 1999; Evans and Martin, 2000; Cardozo et al., 2004). However, there is limited information on effects of other plant extracts, or secondary plant metabolites, on rumen microbial fermentation, their mechanism of action, and optimal doses to improve the efficiency of nutrient utilization.

The objective of this study was to evaluate effects of several plant extracts (Experiment 1) and active compounds of plants (Experiment 2) on protein degradation and rumen microbial fermentation profile in a dual flow continuous culture system.

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