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## Ruminal Microbiome Manipulation to Improve Fermentation Efficiency in Ruminants

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Rumen microbiome manipulation to enhance fermentation efficiency and mitigate emissions: mechanisms, interventions, and applicability. The rumen microbiome orchestrates the conversion of fibrous feeds into short-chain fatty acids (SCFAs), microbial protein, and gaseous byproducts; targeted manipulation can improve fibrolysis, redirect hydrogen toward propionate, and reduce methane and ammonia losses, thereby increasing feed efficiency and sustainability in ruminant systems [1–4]. Practical strategies integrate microbial ecology, hydrogen flow, pH stabilization, and nitrogen synchronization to optimize energy capture while limiting environmental emissions from enteric fermentation and nitrogen excretion [3–6].

#### Rumen ecology and intervention targets

The rumen houses bacteria, archaea, protozoa, fungi, and phages forming a resilient, diet responsive consortium in which fibrolytic bacteria such as Fibrobacter succinogenes, *Ruminococcus albus*, and R. *flavefaciens*, alongside cellulolytic protozoa and anaerobic fungi, drive plant cell wall degradation [1,7–13]. Fermentation outputs like SCFAs, H2, CO2, and ammonia—are coupled to host absorption and pH homeostasis, linking pathway stoichiometry to milk and meat production; because methanogenesis competes with propionate as a terminal H2 sink, redirection of reducing equivalents underpins many efficiency and mitigation approaches [3–5,14].

#### Enhancing fibrolysis and SCFA yield

Incomplete utilization of plant cell walls is common on forage-based diets due to biochemical barriers and limited rumen retention, elevating nutrient loss and methane per unit intake [8–10,14]. Direct-fed microbials (DFM) including lactate-utilizers and yeasts (e.g., *Saccharomyces cerevisiae*, *Aspergillus oryzae*) stabilize rumen pH, scavenge oxygen at feed particle surfaces, provide growth factors, and stimulate cellulolytics, improving fiber digestion and performance at comparatively low cost [15–23]. Exogenous fibrolytic enzymes (EFE) add complementary cellulase/xylanase activities that release reducing sugars and cleave linkages impeding microbial attachment; outcomes depend on aligning enzyme spectra and dose with diet pH, temperature, and substrates, given frequent co-activities and variable in vivo responses [24–26].

## Redirecting hydrogen: decreasing methanogenesis, increasing propionate

Hydrogenotrophic methanogens reduce CO2 using H2 derived from bacteria, protozoa, and fungi; many methanogens are embedded in feed-particle biofilms or occur as endosymbionts within protozoa, complicating direct inhibition [4,30–31]. Ionophores such as monensin and lasalocid suppress H2-producing Gram-positive bacteria and shift fermentation toward propionate while modulating deamination and biohydrogenation; effects can be transient due

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to adaptation, stronger on starch-rich diets, and subject to regulatory limits, so on-farm strategies often emphasize alternatives that preserve SCFAs and productivity [32–34].

#### Plant secondary compounds: tannins, saponins, essential oils

Plant secondary compounds (PSC) modulate microbial communities without antibiotic resistance concerns, though responses vary by chemistry, dose, diet, and adaptation [35–39]. Condensed tannins (CT) can bind proteins/carbohydrates and disrupt methanogen–protozoa associations; tanniferous legumes and browse (e.g., Leucaena, Prosopis) reduce methane and ammonia and often increase propionate on forage diets, with efficacy influenced by CT structure and co-metabolites such as hydrolyzable tannins and mimosine [41–44]. Saponins are protozoacidal via sterol-binding but may show transient responses if deglycosylated to sapogenins by rumen bacteria [37,48–50]. Essential oils (EO) rich in phenylpropanoids and terpenes can suppress methanogenesis via membrane disruption and mixture synergism, but dose, palatability, encapsulation, and diet interactions must be managed to preserve fiber degradability [36,39–40].

#### Propolis, plant oils, and chitosan

Bee propolis, containing flavonoids and isoflavones, has reduced methane while increasing digestibility and total SCFAs in vitro and in vivo, indicating a shift from methane to microbial protein and SCFAs, with practicality where local supply is strong [53–58]. Dietary fats and plant oils up to about 6% of dry matter can suppress protozoa and methanogens and act as hydrogen sinks via biohydrogenation, with effects contingent on fatty acid profile, inclusion form, and forage to concentrate ratio [59–60]. Chitosan, a biodegradable polycationic polysaccharide, preferentially inhibits Gram-positive bacteria, often increasing propionate and lowering methane effects strongest at lower pH with some grain inclusion and can shift biohydrogenation toward more unsaturated milk fatty acids and cis-9, trans-11 CLA [61–64].

### Managing pH and acidosis risk

High loads of rapidly fermentable carbohydrate predispose to subacute or acute acidosis, suppressing cellulolytics and impairing performance; buffers including sodium bicarbonate, magnesium oxide, and calcium magnesium carbonate stabilize pH and have improved milk fat in high-starch diets, while malate stimulates lactate-utilizers such as *Selenomonas ruminantium* to convert lactate to SCFAs [66–71].

## Maximizing ruminal microbial protein synthesis

Microbial protein supplies roughly 50–90% of amino acids to the small intestine; maximizing it reduces nitrogen waste and reliance on expensive true protein, especially on low-protein forages [72–74]. Energy supply and synchronization with degradable nitrogen drive microbial protein yield; urea molasses blocks, slow-release urea, and legume supplementation improve capture, and reduced methane formation often coincides with higher microbial protein due to improved hydrogen economy and redox balance [73–75].

## Curtailing proteolysis and ammonia emissions

Excess ruminal proteolysis, peptidolysis, and deamination elevate ammonia beyond microbial demand, increasing urinary urea and environmental loading; strategies include formulating with rumen-undegradable protein, synchronizing fermentable carbohydrate with degradable nitrogen, using slow-release nonprotein nitrogen, and employing CT to protect protein in the rumen while improving post-ruminal amino acid supply [41–43,72–75].

## Product quality and co-benefits

Manipulating biohydrogenation through tannins, essential oils, chitosan, and selected vegetable oils can increase polyunsaturated fatty acids and cis-9, trans-11 CLA in milk and meat by limiting terminal saturation or altering key bacterial groups, provided fiber digestibility and energy balance are maintained [59–64]. These quality gains can accompany

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methane and nitrogen mitigation when dosing and diet matching are carefully managed [59–64].

#### **On-farm integration**

Heterogeneity across animals, diets, and microbial adaptation explains variable responses; stacked, complementary levers tailored to local feeds are more reliable than single additives, particularly in forage-first and smallholder systems where tanniferous legumes/browse, yeast based DFM, substrate-matched fibrolytic enzymes, and synchronized nonprotein nitrogen often deliver robust gains in efficiency and emissions intensity [8,20,24,26,41–44].

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