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Regulation of Dietary Protein Solubility Improves Ruminal Nitrogen Metabolism In Vitro: Role of Bacteria–Protozoa Interactions

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Abstract: Precision dietary interventions (e.g., altering proportions of dietary protein fractions) has significant implications for the efficiency of nutrient use in ruminants, as well as lowering their environmental footprint, specifically nitrogen (N) emissions. Soluble protein (SP) is defined as the protein fraction that is rapidly degraded in the rumen (e.g., non-protein N and true protein), and our previous study found that regulating SP levels could improve N efficiency in *Hu* sheep. Thus, the present study was conducted to explore in vitro how protein fractions with different SP levels modulate the rumen microbial community and its association with N metabolism. Four dietary treatments with different SP proportions and similar crude protein (CP) content (~14%) were formulated (% of CP): 20 (S20), 30 (S30), 40 (S40) and 50 (S50). Results showed that NH_3-N content increased with increasing SP levels at 4, 12 and 24 h; TVFA, acetate, propionate and valerate were higher in S30 and S40 ($p < 0.05$) and had quadratic effects ($p < 0.05$). Moreover, dry matter digestibility (DMD) and N digestibility (ND) were all decreased with S20 and S50 ($p < 0.05$). The S30 and S40 treatments increased the abundance of Bacteroidetes and *Prevotella* (*Prevotella_ruminicola*) but decreased the abundance of Firmicutes and Proteobacteria ($p < 0.05$). Bacterial pathways related to amino acid and fatty acid metabolism also were enriched with S30 and S40. The abundance of *Entodinium* was increased with S30 and S40 and had a positive correlation with *Prevotella*, and these two genera also played an important role in N metabolism and VFA synthesis of this study. In conclusion, bacterial and protozoal communities were altered by the level of SP (% of CP), with higher SP levels (~50% of CP) increasing the microbial diversity but being detrimental to rumen N metabolism.

Keywords: dietary interventions; soluble protein; nitrogen metabolism; rumen bacteria; ciliate protozoa

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1. Introduction

With the rapid development of the modern economy, the demand for high-protein food sources is increasing year by year, and beef, mutton, and dairy products (especially ruminant products) seem to be the first choice for most consumers [1,2]. However, ruminant production is one of the main drivers of global environmental degradation, and the contribution to environmental pollution is much greater compared with non-ruminants [3,4].