AN EXOGENOUS PROTEASE INCREASES ENZYMIC ACTIVITIES, MICROBIAL NUMBERS AND FIBER DEGRADATION BY MIXED RUMINAL MICROORGANISMS IN CONTINUOUS CULTURE.

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The effects of pH and addition of a protease mixture on the fermentation characteristics of a total mixed ration (TMR) were investigated in a dual-flow continuous culture apparatus, using a 4 x 4 Latin Square design with a 2 x 2 factorial arrangement of treatments. The diet (DM basis) consisted of 30% alfalfa hay, 30% corn silage and 40% rolled corn. The silage and the grain were milled fresh, mixed with the alfalfa and treated with the enzyme (1.5 µL/g feed) daily. Ruminal fluid was collected 2 h post-feeding from 3 lactating dairy cows fed a TMR. Fermenters were fed 80 g DM/d in equal portions every 12 h. Treatments were control (C) and enzyme-treated (T) TMR at either high pH (HC and HT) or low pH (LC and LT). The pH was altered by diluting the artificial saliva to 60% of its original composition. Enzymic activities and total and cellulolytic bacterial numbers were determined on the liquid phase of the fermenter contents, 6 h post-feeding. Fiber degradation was determined from the outflow residues. Enzyme addition increased ($P < 0.05$) xylanase, xylosidase, endoglucanase, and protease activities (608 vs. 750; 0.48 vs. 0.80; 82 vs. 112; and 1.2 vs. 7.5 units for C and T, respectively), whereas it tended ($P < 0.12$) to increase exoglucanase and glucosidase activities (0.8 vs. 1.4; and 4.7 vs. 5.9 units). However, enzyme did not affect ($P = 0.18$) arabinofuranosidase activity (5.2 vs. 6.8). Total microbial numbers (expressed as Log_{10}) were increased ($P < 0.05$) at low pH (9.13 vs. 9.36) but enzyme had no effect ($P = 0.13$). Cellulolytic bacteria were reduced ($P < 0.02$) at low pH (3.91 vs. 2.79), with no effect ($P = 0.88$) of enzyme. Low pH reduced ($P < 0.001$) NDF, ADF, and cellulose degradation. NDF degradation was increased ($P < 0.01$) by enzyme addition (20% vs. 27%) but ADF was unaffected ($P < 0.20$), resulting in an increase ($P < 0.001$) in hemicellulose degradation. It is speculated that the enzyme removed structural barriers present in the feed, allowing a more rapid colonization of the fiber by ruminal microorganisms.