

Toxic Effects of Fatty Acids on Ruminal Protozoa and Their Methanogenic Endosymbionts

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Effects of supplemental fats on ruminal fermentation have been reported that include decreased fiber fermentation, decreased ruminal microbial populations (especially protozoa) and decreased methane emission. We proposed that specific fatty acids are toxic to methanogenic bacteria, which are endosymbionts in *Entodinium spp.* protozoa. The loss of the endosymbionts results in either reduced competitive advantage or protozoal death. To measure effects of specific fatty acids on ruminal fermentation, protozoal numbers and their endosymbionts, two in vitro experiments with a split-plot design and two blocks were conducted. Substrates were barley or alfalfa with treatments of none, stearic, oleic, linoleic or linolenic acid (2% by weight). With barley substrate, rate of gas accumulation was greater ($P < .10$) with C18:0 than the unsaturated fatty acids (.18 vs $.16 \pm .01$ mL/h). Epifluorescence micrographs showed decreased methanogenic endosymbionts without decreased *Entodinium spp.* numbers. In contrast, with alfalfa substrate, no effect of fatty acid addition was noted on gas accumulation or methanogenic endosymbionts. Therefore, the association between *Entodinium spp.* and their methanogenic endosymbionts was not obligate for protozoal survival and growth with the barley substrate but may have been obligate with the alfalfa substrate.

Key Words: Barley, Alfalfa, Ruminal Fermentation, Protozoa, Epifluorescence