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Satter L D & Slyter L L. Effect of ammonia concentration on rumen microbial protein production in vitro. *Brit. J. Nutr.* 32:199-208, 1974.
[Nutrition Institute, Agricultural Research Service, US Department of Agriculture, Beltsville, MD]

This paper showed that growth of ruminal bacteria was not stimulated by increasing ruminal ammonia concentrations above 2 mM. This led to prediction of when to expect benefit from addition of urea to ruminant diets. [The *SCI*® indicates that this paper has been cited in over 225 publications.]

Ammonia—How Much for Bacterial Feast or Famine?

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Urea is used extensively as a substitute for protein in ruminant diets, and in the early 1970s it was replacing as much as four million tons of soybean meal in the US. Urea can serve as a substitute for dietary protein because the bacteria in the ruminant forestomach (rumen) synthesize protein from ammonia produced through hydrolysis of urea. The extent to which urea can substitute for dietary protein depends on how much ammonia the bacteria can utilize for protein synthesis.

Rather strong differences of opinion existed regarding urea utilization by ruminants when we started our experiments at the Beef Cattle Research Branch, part of the Agricultural Research Service of the US Department of Agriculture in Beltsville, Maryland. It seemed something better was needed than the arbitrary recommendations in use at the time. The question that needed answering was what concentration of ammonia in ruminal contents was necessary to support maximum growth rate of ruminal bacteria.

The senior author's (LDS) first attempt at measuring microbial growth as a function of ammonia concentration relied on batch cultures of mixed ruminal bacteria. This did not work. There was no control over ammonia concentration in the media. In discussions with the coauthor, attention was turned to the use of continuous culture fermentation, a technique that Len Slyter had learned from Mike Wolin at the University of Illinois. This worked well for our purpose, and we were able to show that low concentrations of ruminal ammonia (5 mg NH_3-N/100 ml ruminal fluid, or 2 mM) would support maximum microbial growth and higher concentrations were without effect on bacterial yields. Since ruminal concentration of ammonia can range from barely detectable to 15 or 20 mM, depending upon dietary conditions, it was obvious that urea would be of little benefit with some dietary situations.

While our experimental results seemed clear enough, our conclusion that urea should not be included in ruminant diets when ruminal ammonia concentrations exceeded 2 mM was not embraced by everyone. The majority of people disagreeing with our conclusion felt we were well-intentioned but badly misguided. A few detractors may have doubted the former. Evidence obtained by others[1,2] in the ensuing years has resulted in a widely shared view that there are definite limits to the extent to which urea can be substituted for protein in ruminant diets.

We believe the primary impact of this paper was to focus attention on the issue that urea can function as a protein substitute but only under conditions of low ruminal ammonia concentrations. This paper, along with significant work by J.P. Hogan and R.H. Weston[3] and I.D. Hume *et al.*,[4] stimulated research that eventually became the basis for new approaches to calculating protein requirements for ruminants.[5,6]

It is very pleasing to have our work recognized as a *Citation Classic*. We both feel this is the most significant paper either of us has been involved with. This research questioned the dogma that prevailed in the feed industry at the time and resulted in numerous opportunities for the senior author to discuss the work at nutrition conferences in North America and Europe. This research was a major reason for the senior author being recognized with the American Feed Manufacturer's Award in 1977. The process of challenging entrenched thoughts was a lengthy one, and the time spent on communicating our research findings exceeded the time required to do the research.

1. **Schaefer D M, Davis C L & Bryant M P.** Ammonia saturation constants for predominant species of rumen bacteria. *J. Dairy Sci.* 63:1248-63, 1980. (Cited 35 times.)
2. **Odle J & Schaefer D M.** Influence of rumen ammonia concentration on the rumen degradation rates of barley and maize. *Brit. J. Nutr.* 57:127-38, 1987.
3. **Hogan J P & Weston R H.** The digestion of two diets of differing protein content but with similar capacities to sustain wool growth. *Aust. J. Agr. Res.* 18:973-81, 1967. (Cited 95 times.)
4. **Hume I D, Moir R J & Somers M.** Synthesis of microbial protein in the rumen. I. Influence of the level of nitrogen intake. *Aust. J. Agr. Res.* 21:283-96, 1970. (Cited 145 times.) [See also: **Hume I D.** Citation Classic. (Barrett J T, comp.) *Contemporary classics in plant, animal, and environmental sciences.* Philadelphia: ISI Press, 1986. p. 272.]
5. **Agricultural Research Council.** *The nutrient requirements of ruminant livestock.* Slough, England: Commonwealth Agricultural Bureaux, 1984. 40 p. Supp. 1.
6. **Subcommittee on Dairy Cattle Nutrition, National Research Council.** *Nutrient requirements of dairy cattle.* Washington, DC: National Academy Press, 1988. 157 p.

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