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## Bioavailability of lysine from hydroxymethyl lysine

### Abstract

Twelve mature sheep were used as a ruminant model to estimate the bioavailability of lysine in hydroxymethyl lysine (HML) compared with a commercial product of rumen-protected lysine (RPL; LysiPEARL, Kemin Industries, Inc.) with known availability. The sheep were fed a diet with a forage to concentrate ratio similar to that of dairy diets. Following a control period in which plasma lysine was measured when sheep received no supplemental lysine, the sheep were provided 2 of 4 treatments during periods 2 and 3; treatments included RPL to provide 3 or 6 g/day of available lysine (actual amounts of product provided were based on the manufacturer's data related to ruminal escape and intestinal availability) and 3 or 6 g/day of lysine provided as HML. Blood samples were collected at the end of each feeding period at 3 hours after feeding. Both HML and RPL significantly increased plasma lysine concentrations. By comparison with plasma lysine concentrations when known amounts of bioavailable lysine were provided as RPL, the bioavailability of lysine in HML was estimated to be 94%. Results indicate that HML may be an effective means of supplementing lysine to dairy cattle.; Dairy Day, 2010, Kansas State University, Manhattan, KS, 2010; Dairy Research, 2010 is known as Dairy Day, 2010

### Keywords

Dairy Day, 2010; Kansas Agricultural Experiment Station contribution; no. 11-129-S; Report of progress (Kansas Agricultural Experiment Station and Cooperative Extension Service); 1039; Dairy; Lysine bioavailability; Rumen-protected lysine; Sheep

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# Bioavailability of Lysine from Hydroxymethyl Lysine

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## Summary

Twelve mature sheep were used as a ruminant model to estimate the bioavailability of lysine in hydroxymethyl lysine (HML) compared with a commercial product of rumen-protected lysine (RPL; LysiPEARL, Kemin Industries, Inc.) with known availability. The sheep were fed a diet with a forage to concentrate ratio similar to that of dairy diets. Following a control period in which plasma lysine was measured when sheep received no supplemental lysine, the sheep were provided 2 of 4 treatments during periods 2 and 3; treatments included RPL to provide 3 or 6 g/day of available lysine (actual amounts of product provided were based on the manufacturer's data related to ruminal escape and intestinal availability) and 3 or 6 g/day of lysine provided as HML. Blood samples were collected at the end of each feeding period at 3 hours after feeding. Both HML and RPL significantly increased plasma lysine concentrations. By comparison with plasma lysine concentrations when known amounts of bioavailable lysine were provided as RPL, the bioavailability of lysine in HML was estimated to be 94%. Results indicate that HML may be an effective means of supplementing lysine to dairy cattle.

**Key words:** lysine bioavailability, rumen-protected lysine, sheep

## Introduction

Ruminant microbial protein provides a well balanced source of amino acids for dairy cows, but the amount of amino acids is often not sufficient for high-producing cows. Lysine is considered a limiting amino acid for dairy cows, especially when they are fed corn-based diets. Amino acids can be degraded by rumen microorganisms, and this has led ruminant nutritionists to investigate methods to protect lysine from ruminal degradation such that lysine can be made available in the small intestine for absorption. Protected forms of lysine, however, not only should be protected from ruminal degradation, but also should be available for intestinal absorption. Little research exists on hydroxymethyl lysine (HML) as a source of lysine for dairy cattle, although one study reported a lack of response to HML and suggested that this might be the result of the low pH of the diet (corn silage) leading to degradation of the HML. The goal of this research was to study the intestinal availability of the lysine from HML using sheep as a model by monitoring plasma lysine concentrations compared with observations of a commercial product of known intestinal availability.

## Experimental Procedures

Twelve mature black-faced ewes (77.4 kg) were housed in a large pen (6 x 12 meters) at the Kansas State University Sheep and Goat Center. They were limit-fed (1.6 kg/day, dry matter basis) individually twice daily (7 a.m. and 7 p.m.) for 3 consecutive periods (7 days each). The diet comprised 45.5% alfalfa hay, 44.7% rolled corn, 4.1% soybean meal, 5.1% molasses and 0.57% salt, and it contained 15.4% crude protein and 23.4% neutral detergent fiber (NDF; dry matter basis). During meals, sheep were placed in individual feeding pens (18 inches wide) for 30 minutes.

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The first period was a control period with no lysine supplementation, and blood samples were collected at the end of this period to serve as baseline measurements. The second and third periods were treatment periods. Diets were mixed with 3 or 6 g/day of available lysine from a commercial product of rumen-protected lysine (**RPL**; LysiPEARL, Kemin Industries, Inc.; 21% of product weight was intestinally available lysine) or with HML in amounts that contained 3 or 6 g/day of total lysine. Hydroxymethyl lysine was manufactured in our lab as described in U.S. Patent No. 4,073,945 by the reaction of lysine-HCl with formaldehyde (1.32 moles formaldehyde per mole lysine) in an aqueous solution containing calcium hydroxide (1 mole calcium hydroxide per mole of lysine). Formaldehyde was added with the temperature of the solution near 15°C, then the mixture was stirred for 3 hours, cooled to 5°C, collected on filter paper, and rinsed with cold water. The product was dried in a 55°C oven for 24 hours and then in a vacuum oven at 55°C for an additional 24 hours.

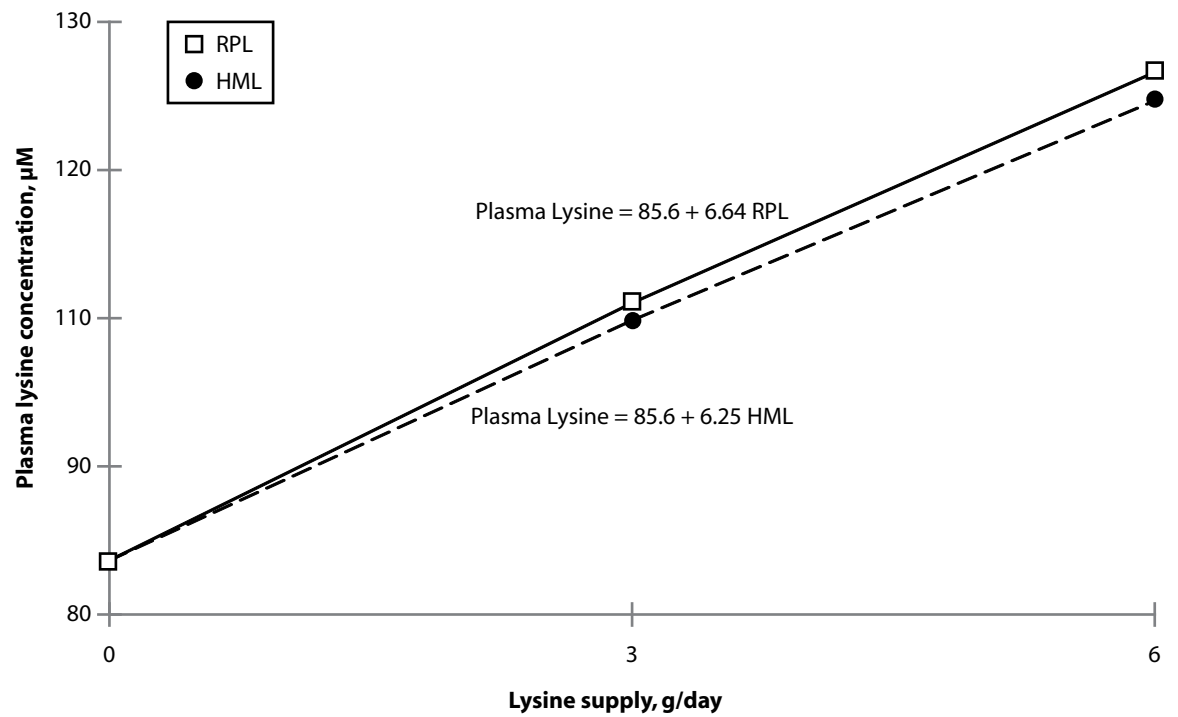
On the final day of each period, blood samples (10 mL) were collected by jugular venipuncture at 3 hours after the morning feeding. Blood samples were placed on ice directly after collection, transferred to the lab, and then centrifuged at  $1,000 \times g$  for 15 minutes. Plasma was transferred to microcentrifuge tubes and frozen at  $-20^{\circ}\text{C}$  pending later analysis of amino acids by HPLC.

## Results and Discussion

A linear increase of plasma lysine was detected after either HML ( $P < 0.01$ ) or RPL ( $P < 0.01$ ) supplementation, but no statistical difference ( $P = 0.78$ ) was detected between the slopes of the regression lines of lysine concentration plotted against HML and RPL supplementation levels (Figure 1). The relative bioavailability of lysine from HML compared with RPL was 94% (not statistically different from 100%), calculated by dividing the slope for HML by that for RPL. Because the amounts of RPL were based on amounts of available lysine, the 94% availability for lysine in HML can be considered a true availability value for HML. Similar conclusions were drawn when plasma lysine as a percentage of total plasma amino acids was used as the response criteria, which strengthens our conclusions.

Hydroxymethyl lysine is a chemical derivative of lysine that has the amino groups chemically attached to a reactive hydroxymethyl group such that the lysine is unavailable to ruminal microbes. The acidic pH of the abomasum, however, releases lysine from the complex and hence the lysine becomes available for intestinal absorption. Because the product is acid labile, acidic diet ingredients such as silages should be considered when mixing with the product because they might release the lysine from HML before feeding.

In conclusion, our results show that HML can be used as an effective source of lysine for ruminant animals and may be an effective product for dairy cattle diets.



**Figure 1: Effect of hydroxymethyl lysine (HML) and rumen protected lysine (RPL) on plasma lysine concentrations of sheep.**

The RPL was provided as amounts of available lysine, whereas HML was provided in amounts of total lysine. Both sources increased ( $P < 0.01$ ) plasma lysine concentrations. Slopes were similar between lysine products ( $P = 0.78$ ).