Does Mechanical Mixing of TMR Compromise Protection Efficacy of Rumen-Protected Lysine Products?

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INTRODUCTION
- Rumen-protected Lys (RPL) products claim some degree of protection from rumen degradation.
- Most research has focused on ruminal bypass rate and intestinal digestion.
- Little information is available regarding the impact of mechanical mixing on the protection or coating material of commercially available RPL products during routine feeding management.

OBJECTIVE
To determine whether mechanical mixing of TMR compromises the ruminal protection efficacy of RPL products.

MATERIALS AND METHODS

Experimental Design
- Six commercial RPL products: AminoShure-L, LysiPEARL, MEGAMINE-L, MetaboLys, USA Lysine, AjiPro-L.
- 2 x 4 factorial arrangement of treatments in a randomized completely block design:
  - Treatments (Trt): with or without mechanical mixing (MIX vs. CON)
  - Time: ruminal incubation (Time): 0, 6, 18, and 24 h
- Each load of TMR and corresponding cannulated cow used as combined blocking factor.

Experimental Procedures
- RPL product (1± 0.03 g) weighed into Dacron bag (5 cm × 10 cm, 50 µm porosity) and considered the experimental unit.
- Super Data Ranger (SDR) with paddle mixer used to simulate TMR mixer.
- Three loads of 350 kg of TMR was mixed and Dacron bags were either distributed into loaded SDR to mix with TMR for 6 min (MIX) or placed with the same load of TMR in a barrel for 6 min (CON). Triplicates of bags were used in each combination of product, treatment, load, and incubation time.
- Bags from the same load of TMR (both MIX and CON) were incubated in one of three cannulated cows simultaneously.
- At each incubation time, bags from cows and bags with undigested products were sequentially processed by hand-washing in 3 barrels of cold water, paper patting to remove dripping water, hanging on clothing line (> 24 h) to air-dry, and extracting Lys from residues left in bags.

Lys extraction procedure:
- Dried residues removed from bag put into 50 mL conical.
- To residues, 10 mL 3 N HCl + 2 mL 95% ethanol was added.
- Samples melted in oven at 90°C for 60 min.
- Samples cooled in ice-water for 10 min for re-solidification of fat.
- Liquid portion was removed below fat layer and brought to 100 mL volume.
- Solution filtrated through 0.45 µm filter.

Lys concentration analyzed by ACQUITY UPLC® system Lys analysis performed by Ajinomoto Bio-Fine Research Laboratories with all samples blindly labeled.

Lys disappearance rate was calculated as:

\[
\frac{(\text{Lys content in RPL product, mg} - \text{Lys in RPL product residue, mg}) \times 100}{\text{Lys content in RPL product, mg}}
\]

Statistical Analysis
- MIXED procedure of SAS (v. 9.2) with mechanical mixing treatment and incubation time and their interaction as fixed effect and each Dacron bag as random term. PDIFF option was used for mean separation. Significance was declared at \( P < 0.05 \).

RESULTS

Table 1. Physicochemical characteristics of RPL products and Lys recovery rate from acid hydrolysis

<table>
<thead>
<tr>
<th>RPL</th>
<th>AminoShure-L</th>
<th>LysiPEARL</th>
<th>MEGAMINE-L</th>
<th>MetaboLys</th>
<th>USA Lysine</th>
<th>AjiPro-L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lys, % as fed basis</td>
<td>53.6</td>
<td>38.2</td>
<td>19.4</td>
<td>28.5</td>
<td>53.6</td>
<td>40.4</td>
</tr>
<tr>
<td>Specific gravity, g/cm³</td>
<td>1.124 ± 0.0005</td>
<td>1.112 ± 0.001</td>
<td>1.089 ± 0.002</td>
<td>1.075 ± 0.0008</td>
<td>1.116 ± 0.0008</td>
<td>1.121 ± 0.0009</td>
</tr>
<tr>
<td>Recovery rate*, %</td>
<td>92.6 ± 3.5</td>
<td>88.7 ± 2.2</td>
<td>81.5 ± 2.1</td>
<td>91.0 ± 0.9</td>
<td>98.4 ± 1.2</td>
<td>90.5 ± 1.6</td>
</tr>
<tr>
<td>Particle size, % of wt</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&gt; 4.75 mm</td>
<td>-</td>
<td>-</td>
<td>84.7</td>
<td>-</td>
<td>-</td>
<td>5.2</td>
</tr>
<tr>
<td>3.35 – 4.75 mm</td>
<td>1.0</td>
<td>-</td>
<td>15.0</td>
<td>-</td>
<td>-</td>
<td>93.5</td>
</tr>
<tr>
<td>2.36 – 3.35 mm</td>
<td>-</td>
<td>-</td>
<td>0.3</td>
<td>-</td>
<td>-</td>
<td>1.3</td>
</tr>
<tr>
<td>1.18 – 2.36 mm</td>
<td>35.7</td>
<td>26.1</td>
<td>-</td>
<td>73.4</td>
<td>9.8</td>
<td>-</td>
</tr>
<tr>
<td>0.6 – 1.18 mm</td>
<td>1.2</td>
<td>46.7</td>
<td>-</td>
<td>26.2</td>
<td>63.1</td>
<td>-</td>
</tr>
<tr>
<td>0.3 – 0.6 mm</td>
<td>-</td>
<td>23.8</td>
<td>-</td>
<td>0.4</td>
<td>25.5</td>
<td>-</td>
</tr>
<tr>
<td>&lt; 0.3 mm</td>
<td>-</td>
<td>3.4</td>
<td>-</td>
<td>-</td>
<td>1.6</td>
<td>-</td>
</tr>
</tbody>
</table>

*Recovery rate was calculated as Lys content in 100 mL 0.2 N HCl buffer solution (after acid hydrolysis) ÷ 100/Lys content of original samples

CONCLUSIONS
- Mechanical mixing moderately increased ruminal Lys loss rate of MetaboLys, LysiPEARL, and USA Lysine at incubation times of 0-12 h, indicating compromised ruminal protection.
- LysiPEARL and USA Lysine had faster and greater ruminal Lys loss, indicating a faster bypass rate is required to ensure post-ruminal Lys availability.

LIMITATION OF THE STUDY
- The diversity of physical characteristics of RPL products (Table 1) may result in different chewing damage and ruminal kinetics that could influence potential to be ruminated and passage rate of each product. These factors could not be accounted for in the design of this in situ study.

Figure 1. Lys disappearance rate of RPL products during a 24-h period

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