

# AjiPro<sup>®</sup>-L bioavailability validation with the isotope dilution technique

To balance for amino acids, you must have absolute confidence in the sources of accuracy in your formulation. While expensive compared to commodities on a per pound basis, the **value of rumen-protected amino acid products lies in their precision**, giving you confidence that your formulation meets the amino

acid requirements of your cow, whether it be in lactation or transition. In other words, confidence in your rumenprotected amino acid product allows you confidence in your formulation.

#### AjiPro<sup>®</sup>-L is nutritionists' no. 1 choice for rumen-protected

Lysine, and with good reason. Its 64% bioavailability specification has been verified by multiple in-vitro, in-situ, and in-vivo trials. A recent trial performed by Dr. Mark Hanigan of the Animal Science Department of Virginia Tech University further validates AjiPro®-L's specifications with an in-vivo technique common in nutrition, the isotope dilution technique.

Elements, such as Carbon, exist in several isotopes – the same number of electrons and protons, but differing number of neutrons. Stable isotopes such as Carbon 12 (<sup>12</sup>C) and Carbon 13 (<sup>13</sup>C) exist in nature in set ratios. By introducing a known quantity of amino acids with a known isotope ratio to a cow and sampling its blood plasma over time, one can calculate the quantity the native amino acids by knowing what the new dilution rate is and therefore calculating the pools size. Figure 1: Basic Principle of isotope dilution. Final ratio of isotopes can be used to calculate original pool size.



Since isotopes behave similarly in a metabolic sense, this technique **creates an internal standard of AA measurement in cow**s. This allows for great precision when measuring amino acid concentrations. Another in-vivo method, the plasma dose response technique, lacks this standard and instead relies on direct measurement of amino acids in the blood plasma. In addition, the plasma dose response technique favors quick-release products that may "overload" the splynic system, flooding blood plasma with amino acids, and may not reflect natural feeding conditions. **The blood plasma technique may falsely give greater absorption values to quick-release products, compared to products with slower release patterns**.

### Using this technique, Dr. Hanigan's lab was able to assess the true bioavailability of AjiPro®-L.

- $\rightarrow$  All cows were fed a base TMR.
- Treatment consisted of top dressing 333.5 g of AjiPro3G on a daily basis.
- → 6 cows were used in a 6x7 Latin Square design, during which each cow saw each treatment one time for a total of 6 replicates per treatment.
- → Positive controls (a mixture of essential AA and sodium caseinate), were infused abomasally. The AA from the EAA mix was expected to be 100% available and those from casein, greater than 95% available.
- Bioavailability of RPAA is defined as grams of intestinal AA absorption/gram of consumed AA in each product. Derived plasma AA entry rates for each treatment (g AA absorbed/d) were regressed on the total intake (diet plus infused) of that AA for each diet (negative control, positive controls, and each RPAA) to estimate entry rates by ingredient, or in the case of the base TMR, for the entire diet.
- → The resulting bioavailability predictions were corrected for initial use of AA by the gut during first pass through the splanchnic tissues using an estimate of 7% loss as described by Estes et al. 2018.

## Table 1. Ajinomoto Rumen Protected Lysine Results

Treatment		Plasma appearance*
EAA Lys		102.0
EAA Met		105.0
EAA His		97.4
Casein Lys		99.8
Casein Met		104.0
AjiPro®-L 3rd Gen	Plasma appearance	64.1
	Corrected Bioavailability**	68.9
	Specification value	64.0

\* Grams amino acid absorbed into blood per gram fed AA

\*\* Predicted intestinal availability corrected for 7% loss during first pass

#### **References:**

Estes, K. A., R. R. White, P. S. Yoder, T. Pilonero, H. Schramm, H. Lapierre, and M. D. Hanigan. 2018. An in vivo stable isotope-based approach for assessment of absorbed amino acids from individual feed ingredients within complete diets. J. Dairy Sci. 101:7040–7060



Learn about AjiPro<sup>®</sup>-L at www.AjiPro-L.com and contact technical representatives for more information.

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