

AA ( $\mu\text{mol/L}$  basis) to determine the degree of elevation of plasma Lys in response to treatment. Relative to A2G, estimated bioavailability of A3G was determined using the slope-ratio assay technique. Plasma Lys was greatest ( $P \leq 0.05$ ) for 150 g/d A2G ( $119.8 \pm 3.2 \mu\text{mol/L}$ ) and 150 g/d A3G ( $123.8 \pm 3.5 \mu\text{mol/L}$ ) when compared with 0 g/d Lys ( $110.7 \pm 3.3 \mu\text{mol/L}$ ). The slope for A3G treatment was numerically greater ( $0.009$ ;  $r^2 = 0.92$ ) when compared with the slope for A2G treatment ( $0.006$ ;  $r^2 = 0.93$ ) when expressing the concentration of plasma Lys relative to total AA. Calculated bioavailability of A3G was 146.7% of the bioavailability of A2G. To our knowledge, this is the first study to measure relative bioavailability of RPL products in a TMR to simulate on-farm exposure when TMR is fed  $1 \times /\text{d}$ .

**Key Words:** lysine, bioavailability, rumen-protected

**T213 Characteristics of a rumen-protected lysine product. 1: Bioavailability of the third-generation AjiPro-L.** M. Miura<sup>\*1</sup>, A. Haruno<sup>1</sup>, H. Sato<sup>1</sup>, S. Shimizu<sup>1</sup>, M. Nakamura<sup>1</sup>, Y. Miyazawa<sup>1</sup>, T. Fujieda<sup>1</sup>, and I. Shinzato<sup>2</sup>, <sup>1</sup>Research Institute for Bioscience Products & Fine Chemicals, Ajinomoto Co. Inc., Kawasaki, Kanagawa, Japan, <sup>2</sup>Ajinomoto Heartland Inc., Chicago, IL.

A new generation of a rumen protected lysine product, the third-generation AjiPro-L (A3G, Ajinomoto Co. Inc.), was recently developed. Because the particle size of A3G is smaller than the previous generation product (the second-generation AjiPro-L, A2G), it was expected that A3G has higher intestinal digestibility than A2G due to an increased surface area while the rumen protection of A3G might be compromised. This study was conducted to evaluate the bioavailability of A3G by examining the rumen escape and the fecal excretion of the product. Four dry cows (BW 650–720 kg), fistulated both ruminally and duodenally and fed a corn silage based diet (DMI 13–14 kg/d), were used. Ruminal escape of A2G and A3G was determined by the duodenal digesta collection method with using highly protected L-arginine as a control marker (HP-Arg; Robinson et al., 2011). A2G or A3G was ruminally administered along with HP-Arg and duodenal digesta was collected every 2 or 3 h for 48–54 h. The digesta was heated with hot water to dissolve fat-containing A2G or A3G particles and homogenized to extract free Lys and Arg. Changes of Lys and Arg concentrations in duodenal digesta were plotted to calculate the area under the curve (AUC). The proportion of AUC of Lys to Arg was defined as the rate of ruminal escape of A3G, based on in situ rumen protection of HP-Arg (>90%). Fecal excretion of A2G or A3G was measured by the 72 h-fecal collection method (Miura et al., 2015). Feces collected individually after ruminal ingestion of the products were homogenized with hot water to extract Lys from A2G or A3G particles in feces. Amounts of Lys excreted in feces were calculated by analyzing free Lys concentration in the extract. Statistical differences were tested by a one-way ANOVA. Ruminal escape of A3G ( $68 \pm 7\%$  of initial, mean  $\pm$  SE) was not different from A2G ( $73 \pm 6\%$ ,  $P = 0.60$ ), but fecal excretion of A3G ( $18 \pm 3\%$ ) was significantly lower than that of A2G ( $32 \pm 4\%$ ,  $P < 0.05$ ). These results indicated that A3G can deliver more bioavailable Lys to the absorption site of dairy cows than A2G, thanks to improved intestinal digestibility due to an increased surface area of A3G.

**Key Words:** rumen-protected lysine, dairy cow, bioavailability

**T214 Characteristics of a rumen protected lysine product. 2: Handling properties of the third-generation AjiPro-L in feeding practices.** M. Miura<sup>\*1</sup>, A. Haruno<sup>1</sup>, M. Tanida<sup>1</sup>, Y. Miyazawa<sup>1</sup>, T. Fujieda<sup>1</sup>, and I. Shinzato<sup>2</sup>, <sup>1</sup>Research Institute for Bioscience

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Our results of in vivo animal tests showed that the third-generation AjiPro-L (A3G, Ajinomoto Co. Inc.) has higher bioavailability than the previous generation product (the second-generation AjiPro-L, A2G) mainly due to an increased intestinal digestibility which is attributed to the smaller particle size of A3G compared with A2G. Because the smaller particle size may affect the handling properties such as mixing homogeneity in feeds, durability at TMR mechanical mixing, and stability in TMR contact, however, the following studies were conducted to evaluate those properties of A3G. To examine the mixing homogeneity in feeds, A3G was added into a grain mix at 4.0 wt. % by using an auger mixer. The mix was transported by truck for 100 miles and unloaded into a feed bin. Four sub-samples of the mix (1 kg) were collected from a discharger of the bin. A3G particles in the mix were separated, weighed, and calculated for the inclusion rate. Statistical differences ( $P < 0.05$ ) were tested by a one way-ANOVA. Mean inclusion rates of A3G in the mix before and after the transportation and subsequent discharge from the bin were  $4.3 \pm 0.2\%$  and  $3.8 \pm 0.1\%$ , respectively (mean  $\pm$  SD,  $P < 0.05$ ). Assuming the general CV of such feed mixing test is about 5%, those 2 rates fell within an acceptable range of the theoretical inclusion rate  $\pm$  2 SD. Next, in situ ruminal protection of A3G after TMR mechanical mixing was evaluated. A3G (1 g) were weighed and placed into a Dacron Bag. The bags were mixed with TMR by the Data Ranger (American Calan, Inc.) and were collected. Then the bags were placed in the rumen for 24 h. Comparing with intact A3G (Control), in situ protection of A3G was not affected by TMR mechanical mixing ( $P = 0.09$ ). Lastly, a loss of Lys from A3G after contact with TMR was evaluated by measurements of Lys released from A3G into TMR after extraction with water at 0, 6, 12, 18, and 24 h. When A3G was mixed with TMR, about 5% of Lys in A3G was released into TMR within the first 6 h but no further Lys was released during the following 18 h. These results demonstrate that handling properties of A3G are not compromised regardless of reduction in the particle size.

**Key Words:** rumen-protected lysine, handling, stability

**T215 Impact of tannins and grazing schedule on ruminal inoculum activity of dairy cows: Evaluation using the in vitro gas-production technique.** C. A. Pozo<sup>\*1</sup>, J. L. Repetto<sup>2</sup>, G. V. Kozloski<sup>1</sup>, M. Cuffia<sup>3</sup>, A. Ramirez<sup>2</sup>, and C. Cajarville<sup>2</sup>, <sup>1</sup>Departamento de Zootecnia, Universidade Federal de Santa Maria, Santa Maria, RS, Brazil, <sup>2</sup>Facultad de Veterinaria, Universidad de la República, San José, Uruguay, <sup>3</sup>Facultad de Agronomía, Universidad Nacional del Litoral, Esperanza, Santa Fe, Argentina.

The aim of this study was to evaluate the effects of dietary Acacia mearnsii tannin extract (TE) and managing the grazing schedule, on in vitro fermentative activity of the rumen inoculum (RI) collected from dairy cows. The trial was conducted with 9 Holstein cows averaging  $197 \pm 12$  d in milk, in a triplicate  $3 \times 3$  Latin square design, through three 22-d experimental periods. The treatments consisted of morning grazing and afternoon TMR (AM), morning grazing and afternoon TMR added with 15 g/kg of TE (AMt), or morning TMR and afternoon grazing (PM). Cows were fed twice a day at 0700 and 1600 h, having access to TMR or pasture during 5 h. After 20 d of adaptation, RI of each cow was collected twice a day, 4 h after the morning and afternoon meal. The pH was measured and the fermentative activity of the RI was estimated through a 96-h in vitro gas-production assay. The substrates used were 2 whole-crop oat silages incubated in triplicate. Data were fitted to a simple exponential model with lag. Statistical analysis was carried out using the PROC MIXED of SAS, where treatments were compared