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Comparative effects of multiple sources of rumen-protected methionine on milk production and serum amino acid levels in mid-lactation dairy cows

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ABSTRACT

The dairy industry demand for rumen-protected methionine (RP-Met) supplements has been competitive because of the constant emergence of new products. To evaluate performances, our study was designed to characterize the production response of 3 RP-Met supplements in mid-lactation dairy cows. Twelve multiparous Holstein cows were used in a replicated 4 × 4 Latin square design with 21-d treatment period. Treatments included control [(basal) diet based on corn silage and alfalfa haylage, supplemented with 0.025% of ration DM of lysine (Lys; Ajipro)] or 1 of 3 RP-Met supplements [Smartamine M (SMM), Mepron M85 (MM85), or Novimet (NVM)]. For RP-Met groups, Met and Lys were supplemented to the basal diet at 0.03 and 0.20% of ration DM, respectively. Treatments had no effect on DMI or milk yield. Treatment did not modify milk fat or lactose concentration; however, milk protein content was elevated with SMM, relative to control or NVM (3.30% vs. 3.24 or 3.24%, respectively; $P < 0.05$). Milk fat, protein, and lactose yield were not modified by treatments. Treatments tended ($P = 0.12$) to affect milk urea nitrogen. Serum Met concentration increased for SMM compared with control, MM85, or NVM (27.3 μM vs. 21.2, 23.3, or 22.7 μM, respectively; $P < 0.001$). Similarly, supplementation of SMM reduced the serum Lys:Met ratio (4.5:1) compared with control (5.2:1), MM85 (5.1:1), or NVM (5.2:1) ($P < 0.05$). Treatment did not modify the serum levels of all other EAA. We conclude that SMM increased circulating Met and milk protein content more effectively than NVM or MM85.

Key words: dairy cow, lysine, methionine

INTRODUCTION

Ruminal synthesized microbial protein and RUP, together with endogenous protein, contribute to the metabolizable protein (**MP**) requirement for maintenance, growth, pregnancy, and lactation in dairy cows. The NRC (2001) stated that the AA profile of RUP in corn-based feeds and soybean products is not balanced compared with microbial protein, which has similar AA composition with milk. As a result, the typical United States diets base on corn silage and alfalfa haylage supply inadequate methionine (Met) and lysine (Lys), which limits milk production performance in dairy cows (NRC, 2001).

For decades, supplementation of rumen-protected (**RP**) AA has been a common method to ameliorate the AA deficiency in dairy cow diets. These RP-AA products are designed to protect AA from ruminal fermentation and to make AA available in the small intestine. Lara et al. (2006) demonstrated that RP-AA supplementation can increase duodenal AA flow and absorption to improve milk production. Previous studies have observed that the addition of RP-Met and RP-Lys can elevate DMI, milk yield (**MY**), and yields and percentage of milk protein and fat (Donkin et al., 1989; Socha et al., 2008). Also, supplementation of RP-Met and RP-Lys to a low CP diet elevated MY and milk protein yield, in a similar manner as a high CP diet (Piepenbrink et al., 1996).

Among the technologies used in the industry, encapsulation of AA with polymeric compounds has been proven to effectively deliver AA postruminally (Rogers et al., 1987). Specifically, Smartamine M (Adisseo Inc., Alpharetta, GA) employs a pH-sensitive copolymer of stearic acid and 2-vinylpyridine-co-styrene, whereas Mepron M85 (Evonik Inc., Kennesaw, GA) uses a combination of ethylcellulose and stearic acid. In a meta-analysis, these RP supplements have been reported to exhibit extensive bioavailability in the postruminal gastrointestinal gut (Patton, 2010). Novimet, a new product by Innovad Inc. (Essen, Belgium), also claims to have a SFA coating. The dairy industry can benefit from the comparative evaluation of currently available RP-Met supplements on milk production.

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Therefore, the objective of this study was to compare the production performance of Smartamine M, Mepron M85, and Novimet in modifying milk production and serum AA concentrations in mid-lactation dairy cows.

MATERIALS AND METHODS

Experimental Design and Animal Management

Experimental procedures were approved by the Institutional Animal Care and Use Committee at West Virginia University (Morgantown). Twelve multiparous Holstein dairy cows were enrolled in a study completed at the West Virginia University Animal Science Farm (Morgantown, WV). Cows were housed in a free-stall barn and trained to access the feed from Calan gate feeders (American Calan Inc., Norwood, NH). All cows were selected for this study based on DIM and MY and blocked into 3 squares by DIM. At the initiation of the experiment, the mean ± SD of BW, DIM, and MY for the cows were 602 ± 46 kg, 174 ± 18 d, and 31.0 ± 4.5 kg/d, respectively. Cows were randomly assigned to 1 of 4 dietary treatments in a replicated 4 × 4 Latin square with 21-d treatment period. Treatments included control [basal diet supplemented with 0.025% of ration DM of Lys (Ajipro; Ajinomoto Heartland Inc., Chicago, IL)] or 1 of 3 RP-Met supplements [Smartamine M (**SMM**), Mepron M85 (**MM85**), or Novimet (**NVM**)]. For RP-Met groups, Met and Lys (Ajipro) were provided to the basal diet at 0.03 and 0.20% of ration DM, respectively. The amounts of RP-Met and RP-Lys supplemented were calculated by dividing the amounts of Met and Lys by bioavailability of AA in the products, respectively.

The basal diet (% of DM) was composed of 37% corn silage, 21% alfalfa haylage, and 42% concentrate mix and was formulated to meet requirements of energy, protein, minerals, and vitamins for dairy cows with a corresponding 632 kg of BW, 170 DIM, 36 kg of milk/d, 3.7% milk fat, and 2.95% milk true protein (Table 1; NRC, 2001). The diet was mixed once daily at 0800 h and fed twice daily at 0900 and 1800 h. Access to feed was blocked from 0700 to 0900 h to allow for orts and feed offered to be weighed. Feedstuffs including corn silage, alfalfa haylage, grain mix, and dairy pellets were weighed and mixed in a Triolet 1–700 TMR mixer (Trioliet Inc., Oldenzaal, the Netherlands). Cows were fed at 110% of expected intake and had free access to water. The products of RP-AA were weighed for each cow daily and mixed into individual TMR by hand before feeding. Cows were milked twice daily at 0800 and 1800 h.

Sample Collection and Analysis

Samples of ingredients and TMR were collected on a weekly basis and dried at 60°C for 48 h in a forced-air oven for storage and DM measurement. Based on DM contents of ingredients, TMR was adjusted weekly. At the end of the experiment, each dried feed ingredient sample was ground to pass a 1-mm screen of a Wiley mill (Thomas Scientific, Swedesboro, NJ), and wet chemistry analysis was performed by the Rumen Fermentation Profiling Lab (Morgantown, WV). Samples were analyzed for ash by combustion at 550°C overnight (method 942.05; AOAC International, 1995). Neutral detergent fiber and ADF contents were determined using an Ankom 200 Fiber Analyzer (Ankom Technology Corp., Macedon, NY). Heat-stable α-amylase and sodium sulfite treatments (Mertens, 2002) were used to obtain NDF. Ether extraction was performed according to AOAC International (1995) using a Soxtec Foss Tecator (Foss Analytical, Hillerød, Denmark). Crude protein content was analyzed according to AOAC International (1995) using an automated Tecator digestion system (Tecator Inc., Herndon, VA). Soluble protein was determined with a sodium borate, sodium phosphate buffer procedure (Roe and Sniffen, 1990). Starch content of feed samples was determined by the procedure of Smith (1969). Water-soluble carbohydrates were determined by the extraction procedure adapted from Deriaz (1961). Reducing sugars were determined with a spectrophotometer and potassium ferricyanide.

Milk yield was recorded at each milking starting at the evening milking on d 15 of each 21-d treatment period. Milk samples were collected at each milking starting at the evening milking on d 19 of each 21-d treatment period and stored at 4°C with 2-bromo-2-nitropropan-1,3 diol preservative. Milk samples were sent to Dairy One (Ithaca, NY) and were analyzed for content of fat, true protein, lactose, total solids, and milk urea nitrogen (**MUN**) with midinfrared spectroscopy.

Blood samples were collected (10 mL) via the coccygeal vein at 1100, 1400, and 1700 h on d 21 of each period using BD Vacutainer tubes containing clot activator (Becton, Dickinson and Co., Franklin Lakes, NJ). Blood samples were allowed to clot for 30 min in a heat box set at 37°C. Serum samples were centrifuged for 10 min at 3,400 × *g* at 4°C and were aliquoted into microcentrifuge tubes. Serum samples for each cow were pooled across all 3 collection time points. Composited samples were transported on dry ice box and stored at −80°C until analysis. The serum AA samples were processed using the EZ:faast GC-MS Free (Physiological) Amino Acid Analysis Kit (KG0–7166; Phenomenex, Torrence, CA) containing a norvaline internal standard for normalization. Serum AA were quantified using a Thermo Scientific TRACE 1310 series gas chromatograph coupled to a TSQ8000 Triple Quadrupole mass spectrometer (GC-MS/MS) equipped with an AI/AS 1310 autosampler (Thermo Fisher Scientific, Waltham, MA). Samples were derivatized and analyzed following the manufacturer's protocol as described by Badawy et al. (2008). Amino acids were measured by selected reaction, monitoring the best ions formed using autoSRM, and instrumentation and quantitation were handled by XCalibur (Thermo Fisher Scientific). A 5-point standard calibration curve of 26 AA (10 to 200 μ*M*) was made to calculate concentration (average $R^2 = 0.991$; average CV% = 4.84%).