Short communication: Substituting dry distillers grains with solubles and rumen-protected amino acids for soybean meal in late-lactation cows' diets based on corn silage or ryegrass silage

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ABSTRACT

Excess protein in dairy cattle diets increases production costs and contributes to environmental pollution. The objective of the present study was to evaluate the effect of feeding dry distillers grains with solubles (DDGS) supplemented with rumen-protected Lys and Met in place of solvent-extracted soybean meal on the performance of late-lactation cows. Two experiments were carried out, with each using 24 late-lactating dairy cows distributed among 4 pens. In trial 1, corn silage was the main forage source. Control (HP1) total mixed ration (TMR) contained 16.3% crude protein (CP) with soybean meal as the main protein source. Treatment TMR (LP1) had 13.7% CP when soybean meal was replaced with DDGS and rumen-protected Lys and Met. Forage in trial 2 was ryegrass silage; control TMR (HP2; 15.4% CP) contained soybean meal and rumen-protected Met, whereas treatment TMR (LP2; 13.8% CP) contained DDGS and rumen-protected Lys and Met. Trials were analyzed as crossover design using the MIXED procedure of SAS (SAS Institute Inc., Cary NC) with cow as sampling unit and pen as the experimental unit. Treatments were similar in dry matter intake (21.0 and 20.4 kg/cow per day for HP1 and LP1, respectively) and milk yield (20.7 and 20.5 kg/cow per day for HP1 and LP1, respectively) during trial 1. Milk composition was similar between treatments, averaging 4.22, 3.73, 4.54, and 9.15, respectively, for fat, protein, lactose, and solids nonfat. Milk urea nitrogen decreased from 17.2 mg/dL for HP1 to 9.93 mg/dL for LP1. In trial 2, no significant differences were observed for dry matter intake (21.4 and 20.9 kg/cow per day for HP2

and LP2, respectively), milk yield (28.1 and 26.6 kg/d for HP2 and LP2, respectively), fat yield (0.99 vs. 0.92 kg/d for HP2 and LP2, respectively), protein yield (0.94 vs. 0.86 kg/d for HP2 and LP2, respectively) and lactose yield (1.37 vs. 1.28 for HP2 and LP2, respectively). Milk urea nitrogen decreased from 9.88 mg/dL with HP2 to 6.39 mg/dL with the LP2 treatment. Milk N efficiency tended to be higher for LP treatments in trial 1, but not in trial 2. Low milk urea N suggested nitrogen losses to the environment may be lower when cows were fed diets based on DDGS in both trials. The studies indicated that DDGS with rumen-protected Lys and Met could substitute solvent-extracted soybean meal in low-protein corn silage- and ryegrass silage-based diets for late-lactation dairy cows averaging 20.6 or 27.4 kg of milk/d, respectively.

Key words: corn silage, dry distillers grains plus solubles, rumen-protected amino acids, ryegrass silage

Short Communication

Solvent-extracted soybean meal (SSBM) price volatility in recent years increased interest for alternative protein sources, such as dry distillers grains with solubles (DDGS). Dry distillers grains with solubles contains less CP and higher RUP and Met concentrations but is limited in Lys when compared with SSBM (NRC, 2001). This combination of factors limits RDP intake, microbial protein synthesis in the rumen, and, ultimately, AA supply in the duodenum of high-producing cows. For that reason, complete replacement of SSBM with DDGS can be challenging in early-lactation cows diets, but the goal may be achieved in diets for late-lactation cows in which protein intake and RUP and RDP balance may be less demanding (NRC, 2001).

As economic and environmental pressures mount on dairies to reduce protein inputs and N waste, a necessary corresponding increase in N efficiency hinges on precise protein feeding. Rumen-protected amino acids

Received March 20, 2015.

Accepted July 10, 2015.

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(**RPAA**) are supplemented to deliver essential AA that may be limiting in the duodenum (Colmenero and Broderick, 2006; Lee et al., 2012a). However, discrepancies exist among studies using RPAA at different levels of CP on cow performance when supplementing rumen-protected Met (**RPM**) in the diets (Leonardi et al., 2003; Broderick et al., 2008), rumen-protected Lys (**RPL**) (Paz and Kononoff, 2014), or when using RPL, RPM, and rumen-protected His (Lee et al., 2012b). Two of these studies used a negative balance of MP (Broderick et al., 2008; Lee et al., 2012a,b). Those discrepancies may be the result of forage and feed sources and their interactions influencing AA availability in the small intestine at different production levels (Brito et al., 2006). Therefore, in the current study, 2 experiments were conducted to determine whether late-lactation cows' performance could be maintained when SSBM was replaced with DDGS supplemented with RPL and RPM in low-protein, corn silage- or ryegrass silage-based diets.

Site, Animals, and Management

Both studies were conducted at the Louisiana State University Agricultural Center Southeast Research Station, located in Franklinton, Louisiana. Animal care and handling were conducted according to Louisiana Agricultural Center Institutional Animal Care and Use Committee Guidelines (IACUC protocol #AE2008–09). For each study, 24 Holstein cows were randomly distributed among 4 pens (northeast, northwest, southeast, and southwest) in the research freestall. Each pen (16.5 × 8.5 m) was enclosed by 6 electronic Calan gates

(American Calan, Northwood, NH). Cows were allowed 3 wk of adaptation period for gate training followed by 2 periods of 3 wk each in a crossover design. Each period was comprised of 14 d of treatment adaptation and 7 d for sampling and data recording. Each pen had access to a water trough equipped with flow meter (Recordall model 40, Badger Meter, Milwaukee, WI). Dietary treatments were mixed twice daily using a Super Data Ranger (American Calan) at 0700 and 1300 h. Cows were milked twice daily at 0500 and 1500 h.

Trial 1 was conducted between December 16, 2010, and January 27, 2011, and cows averaged (means ± SD) 334 ± 43 DIM and 22.2 ± 3.8 kg of milk/d immediately before the experimental period. Trial 2 was carried out between June 17, 2011, and July 29, 2011, with cows averaging 244 ± 55 DIM and 30.7 ± 4.8 kg of milk/d. Three fans (150 cm in diameter) on each side of the experimental barn and automatic water soakers were used to attenuate heat stress during trial 2. Soakers were turned off during d 19, 20, and 21 of each period for a concurrent study that required manure collection from the floor.

Diets

Individual feed composition is presented in Table 1. Concentrates and mineral supplements were mixed for each treatment (Table 1) to simplify management. Concentrate mixes contained SSBM (**HP**) or DDGS (**LP**) as main protein sources. Treatment premixes were prepared in batches sufficient for a trial. Premix LP contained RPL, RPM, and DDGS (Table 2). A similar amount of DDGS was added in trial 1 as HP

Table 1. Nutrient composition of feed ingredients used by trial

Ingredient (% of DM, unless otherwise noted)	DM (% as fed)	CP	NDF	ADF	Ca	P	K	Mg
Trial 1								
Corn silage	30.3	8.47	41.0	23.9	0.16	0.26	1.05	0.16
Bermudagrass hay	91.9	8.26	70.9	40.4	0.33	0.29	1.33	0.37
Dry distillers grains with soluble	91.5	27.8	30.3	16.0	0.03	0.86	0.93	0.23
HP1 mix ¹	90.0	25.9	8.50	4.60	2.07	0.69	1.6	0.47
LP1 mix ¹	89.6	13.3	9.37	3.77	2.66	0.61	1.01	0.51
Whole cottonseed	93.2	24.9	58.2	42.4	0.11	0.72	1.03	0.30
Trial 2								
Ryegrass silage	30.4	15.0	56.1	38.6	0.64	0.35	2.52	0.19
Dry distillers grains with soluble	86.8	26.8	32.3	18.6	0.02	0.92	1.09	0.26
HP2 mix ¹	89.9	12.7	18.3	11.5	0.91	0.36	1.15	0.29
LP2 mix ¹	89.8	9.95	18.5	11.5	0.97	0.33	0.94	0.27
Whole cottonseed	92.9	26.2	45.7	39.0	0.10	1.16	0.72	0.27

¹Concentrate mixes contained dry ground corn, dry distillers grains with solubles, soybean meal, soyhulls, and minerals. Trial 1 treatments: (1) HP1 = formulated to contain 16.5% CP and 340 g of MP balance with soybean meal with no addition of RPAA; (2) LP1 = formulated to contain 13.5% of CP and –50 g of MP balance with dry distillers grains plus solubles supplemented with rumen-protected Lys (RPL, AminoShure-L, Balchem, New Hampton, NY) and rumen-protected Met (RPM, Metaspart, Adisseo, Antony, France). Trial 2 treatments: (1) HP2 = formulated to contain 15.4% CP and 169 g/d of MP balance with soybean meal supplemented with RPM; (2) LP2 = formulated to contain 13.5% CP and –59 g/d of MP balance with dry distillers grains plus solubles supplemented with RPL and RPM.

Table 2. Ingredients and nutrient contents of diets in trial 1 and trial 2 containing different levels of CP

Item	Trial 1		Trial 2	
	HP1 ¹	LP1 ¹	HP2 ¹	LP2 ¹
Ingredient (% of DM)				
Bermuda grass hay	11.1	11.1	—	—
Corn silage	43.5	43.5	—	—
Ryegrass silage	—	—	41.0	41.0
Cottonseed, whole with lint	7.30	7.30	6.70	6.70
Corn grain, ground, dry ²	16.2	16.2	32.5	32.5
Dry distillers grain plus soluble ²	0.00	13.5	0.00	4.00
Soybean meal (48% CP) ²	17.0	3.50	7.03	0.00
Soybean hulls ²	0.00	0.00	7.50	10.2
AminoShure-L ^{3,4}	0.00	0.30	0.00	0.25
Metaspart Dry ^{4,5}	0.00	0.10	0.10	0.18
Dry distillers grains plus soluble ⁴	2.10	1.70	2.10	2.10
Calcium carbonate ²	0.80	0.80	0.34	0.34
Sodium bicarbonate ²	0.00	0.00	0.90	0.90
SE Sta. Mineral ^{2,6}	1.50	1.50	1.25	1.25
Salt ²	0.50	0.50	0.58	0.58
Nutrient content, means ($\pm$ SD)				
DM (% as fed)	65.1 ($\pm$ 0.26)	64.9 ($\pm$ 0.20)	50.8 ($\pm$ 0.01)	50.6 ($\pm$ 0.01)
CP (% of DM)	16.3 ($\pm$ 0.61)	13.7 ($\pm$ 0.20)	15.4 ($\pm$ 0.002)	13.8 ($\pm$ 0.004)
NDF (% of DM)	35.8 ($\pm$ 1.12)	36.7 ($\pm$ 1.09)	33.0 ($\pm$ 0.59)	33.9 ($\pm$ 0.47)
ADF (% of DM)	20.8 ($\pm$ 0.72)	21.0 ($\pm$ 0.63)	22.5 ($\pm$ 0.41)	23.0 ($\pm$ 0.33)
Ca (% of DM)	0.62 ($\pm$ 0.12)	0.72 ($\pm$ 0.10)	0.74 ($\pm$ 0.003)	0.73 ($\pm$ 0.002)
P (% of DM)	0.53 ($\pm$ 0.01)	0.50 ($\pm$ 0.01)	0.40 ($\pm$ 0.02)	0.41 ($\pm$ 0.02)
K (% of DM)	1.23 ($\pm$ 0.13)	1.09 ($\pm$ 0.10)	1.59 ($\pm$ 0.02)	1.47 ($\pm$ 0.02)
Mg (% of DM)	0.28 ($\pm$ 0.01)	0.28 ($\pm$ 0.02)	0.26 ($\pm$ 0.002)	0.24 ($\pm$ 0.001)
MP balance ⁷ (g/d)	395	6.0	-12	-24
RDP balance ⁷ (g/d)	80	-315	43	-283
RUP balance ⁷ (g/d)	464	7.0	-16	-31
NE _L balance ⁷ (Mcal/d)	7.1	6.1	4.6	9.7
Lys ⁷ [g/d (%)]	181 (6.47)	149 (6.38)	171 (6.41)	181 (6.72)
Met ⁷ [g/d (%)]	51 (1.81)	49 (2.09)	56 (2.13)	63 (2.44)
His ⁷ [g/d (%)]	62 (2.24)	51 (2.16)	58 (2.22)	57 (2.14)
Lys:Met ratio	3.55:1	3.04:1	3.05:1	2.87:1

¹Trial 1 treatments: (1) HP1 = formulated to contain 16.5% CP and 340 g of MP balance with soybean meal with no addition of RPAA; (2) LP1 = formulated to contain 13.5% of CP and -50 g of MP balance with dry distillers grains plus solubles supplemented with rumen-protected Lys (RPL, AminoShure-L, Balchem, New Hampton, NY) and rumen-protected Met (RPM, Metaspart, Adisseo, Antony, France). Trial 2 treatments: (1) HP2 = formulated to contain 15.4% CP and 169 g/d of MP balance with soybean meal supplemented with RPM; (2) LP2 = formulated to contain 13.5% CP and -59 g/d of MP balance with dry distillers grains plus solubles supplemented with RPL and RPM.

²Feedstuffs were mixed weekly and stored in bins.

³AminoShure-L contained rumen-protected Lys (Balchem).

⁴Feedstuffs were premixed and stored for the entire study.

⁵Metaspart dry contains rumen-protected Met (Adisseo).

⁶SE Sta. Mineral = Southeast Research Station Mineral; contained Ca = 20%; P = 3%, Mg = 7%; K = 6%; S = 3%; Co = 15 mg/kg; Cu = 650 mg/kg; I = 50 mg/kg; Mn = 1,200 mg/kg; Se = 18 mg/kg; Zn = 3,700 mg/kg; vitamin A = 300 kIU/kg; vitamin D = 30 kIU/kg; and vitamin E = 1.5 kIU/kg, manufactured by Kentwood Co-op (Kentwood, LA).

⁷Values of MP, RDP, and RUP balance and AA availability in the duodenum were estimated using the NRC (2001) model.

premix, whereas in trial 2 HP premix contained DDGS plus RPM (Table 2). Rumen-protected Lys and RPM were added to adjust for AA availability in the duodenum according to the NRC (2001) model (3:1 Lys:Met ratio).

Each dietary treatment was randomly assigned to 2 pens: northeast and southwest or northwest and southeast. Diets were assessed using the NRC (2001) model.

Trial 1. Diets contained approximately 55% forage as corn silage and bermudagrass hay (Table 2). Two treatments were used.

1. Control TMR (**HP1**) targeted 16.5% CP and 340 g of MP balance containing SSBM.
2. Treatment TMR (**LP1**) containing DDGS supplemented with RPL (AminoShure-L, Balchem, New Hampton, NY) and RPM (Metaspart, Adisseo, Antony, France) and targeted 13.5% of CP and -50 g of MP balance. Rumen-protected Lys had 50% RUP and 24.3% MP-Lys bioavailability according to the product's label. Rumen-protected Met had an estimated absorption of 50% through the rumen (according to

the product's label) in the form of 2-hydroxy-4-(methylthio) butanoic acid product.

Bermudagrass hay was harvested, stored, and ground as described in McCormick et al. (2011). Corn (Dekalb 6786 Roundup Ready; Monsanto, St. Louis, MO) was harvested with a John Deere Forage Harvester 3975 pull-type equipped with corn processor (John Deere, Moline, IL) and stored in a single trench silo.

Trial 2. In trial 2, diets contained 41% forage as ryegrass silage (Table 2).

1. Control TMR (**HP2**) targeted 15.6% CP and 169 g/d of MP balance using SSBM plus RPM (Metasmart, Adisseo).
2. Treatment TMR (**LP2**) targeted 13.6% CP and -59 g/d of MP balance using DDGS plus RPL (AminoShure-L 2nd gen, Balchem) and RPM (Metasmart). AminoShure-L 2nd gen (Balchem) was reported (label) to contain 80% RUP and MP-Lys bioavailability of 30.4%.

Water was added in similar amounts to both treatments during TMR mixing to reach 50% moisture. Ryegrass was conditioned at cutting with a flail-type mower centerline (Kuhn model FC-300G disc cutter, Kuhn North American, Brodhead, WI), wilted for 2 d, picked up, and chopped with a John Deere Forage Harvester 3975 pull-type equipped with silage head for ryegrass pickup and stored in a bag silo.

Sampling, Laboratory Analysis, and Data Collection

Milk yield was measured as described in Moreira et al. (2009). Milk was sampled on d 16 to 18 of each period according to Moreira et al. (2009). Milk samples were analyzed at the Mid-South Dairy Records DHIA (Springfield, MO) for fat, true protein, and SNF by near-infrared reflectance spectroscopy (Bentley 2000, Bentley Instruments, Chaska, MN), for SCC in a flow cytometer (Bentley Somacount 300), and for MUN by colorimetric enzymatic assay (Bentley Chemspect 150). Milk components were calculated according to morning and evening weights. Energy-corrected milk was calculated according to Tyrrell and Reid (1965).

Water flow was recorded once daily after morning feeding to determine group drinking water intake per pen. Forage, TMR, and refusal were sampled daily, stored frozen at -20°C, and composited weekly for analyses and for DMI estimation. Daily forage samples were taken from the conveyor as silages were loaded to the Data Ranger. Weekly fresh forage samples were

dried in a forced-air, 60°C drying oven for 72 h for dietary DM adjustments.

Cottonseed, DDGS, grain mixes, and treatment premixes (Table 1) were sampled weekly and dried in forced-air, 60°C drying oven for 72 h. Feed samples were processed and analyzed for total N, Kjeldahl, P, NDF, ADF, and minerals as described in Moreira et al. (2009). Actual feed composition, DMI, and milk yield and composition were used in the NRC (2001) model to estimate MP, RDP, and RUP balances, as well as AA availability in the duodenum.

Cows were weighed for 2 consecutive days, immediately after morning milking, and at the beginning and end of each experimental period. Body condition was scored by 3 individuals at the beginning and end of experimental periods (Wildman et al., 1982).

Statistical Analysis

Variables were analyzed using the MIXED procedure of SAS (SAS Institute, 2003). Milk yield, milk composition, and DMI were averaged over time to obtain one observation per cow in each pen or period. The analyses used cow as sampling unit and pen (sequence) as experimental unit, as recommended for pen trials by St-Pierre (2007). Previously mentioned variables, ADG and BCS, were analyzed with treatment, period, and sequence as fixed effects. Pen within sequence and pen within sequence by treatment were set as random effects. Free water intake was analyzed using pen as experimental unit including treatment, period, and sequence as fixed effects, with pen within sequence included as a random effect. Kenward-Roger denominator degrees of freedom adjustment was used for all variables. Values reported are LSM. Significance was declared at $P \leq 0.05$ and a trend was reported if $0.05 < P \leq 0.10$.

Nutrient Composition

Nutrient contents of feeds and diets are summarized in Tables 1 and 2. Substitution of SSBM for DDGS decreased CP content and MP, RDP, and RUP balances in both trials. In trial 1, RPAA supplementation helped narrowing the duodenal flow gap for Lys (181 and 149 g/d as well as 6.47 and 6.38% of MP, respectively, for HP1 and LP1) and for Met (51 and 49 g/d as well as 1.81 and 2.09%, respectively, for HP1 and LP1). In trial 2, estimated Lys availability in the duodenum was 171 (6.41% of MP) and 181 g/d (6.72% of MP), and Met was 56 (2.13% of MP) and 63 g/d (2.44% of MP) for HP2 and LP2, respectively (NRC, 2001).

Table 3. Dry matter and free water intake, milk production and composition, and changes in BW and BCS for late-lactation Holstein cows fed diets containing soybean meal or dry distillers grain supplemented with rumen-protected Lys and Met in corn silage-based diets (trial 1)

Item	Treatment ¹		SEM	Effect ($P > F$)		
	HP1	LP1		Treatment	Sequence	Period
DMI (kg/d)	21.0	20.4	0.59	0.535	0.458	0.069
Milk yield (kg/d)	20.7	20.5	1.18	0.922	0.455	0.184
Fat (%)	4.18	4.25	0.23	0.730	0.885	0.737
Fat (kg/d)	0.85	0.85	0.04	0.989	0.574	0.164
Protein (%)	3.74	3.71	0.10	0.789	0.902	0.785
Protein (kg/d)	0.77	0.75	0.03	0.734	0.371	0.147
Lactose (%)	4.55	4.53	0.08	0.799	0.229	0.589
Lactose (kg/d)	0.95	0.93	0.07	0.820	0.406	0.280
SNF (%)	9.18	9.12	0.12	0.659	0.309	0.439
SNF (kg/d)	1.91	1.87	0.11	0.784	0.393	0.218
4% FCM (kg/d)	21.0	20.9	0.92	0.972	0.483	0.094
ECM (kg/d)	23.3	23.1	1.01	0.911	0.452	0.104
MUN (mg/dL)	17.2	9.93	0.71	0.004	0.404	0.718
Milk yield/DMI	0.98	1.01	0.06	0.721	0.562	0.730
Milk N/N intake	0.22	0.27	0.01	0.077	0.638	0.236
Water intake (L/d)	69.0	64.0	3.33	0.094	0.999	0.015
ADG (kg/d)	1.82	1.77	0.31	0.905	0.625	0.295
BCS change (units/period)	0.14	0.10	0.06	0.707	0.972	0.059

¹Treatments: (1) HP1 = formulated to contain 16.5% CP and 340 g of MP balance with soybean meal with no addition of rumen-protected AA; (2) LP1 = formulated to contain 13.5% of CP and -50 g of MP balance with dry distillers grains plus solubles supplemented with rumen-protected Lys (AminoShure-L, Balchem, New Hampton, NY) and rumen-protected Met (Metaspart, Adisseo, Antony, France).

Animal Performance

Animal performance is summarized in Table 3 for trial 1 and Table 4 for trial 2. No significant difference was found for DMI and milk yield between treatments

in both trials. Similarly, substitution of SSBM for DDGS did not significantly affect DMI or milk yield in Christen et al. (2010).

No difference was found between treatments for milk yield and milk components in both trials, except for

Table 4. Dry matter and free water intake, milk production and composition, and changes in BW and BCS for late-lactation Holstein cows fed diets containing soybean meal supplemented with rumen-protected Met or dry distillers grains supplemented with rumen-protected Lys and Met in ryegrass silage-based diet (trial 2)

Item	Treatments ¹		SEM	Effect ($P > F$)		
	HP2	LP2		Treatment	Sequence	Period
DMI (kg/d)	21.4	20.9	0.65	0.573	0.896	0.932
Milk yield (kg/d)	28.1	26.6	1.31	0.386	0.734	0.143
Fat (%)	3.46	3.45	0.06	0.904	0.791	<0.001
Fat (kg/d)	0.99	0.92	0.05	0.288	0.819	0.374
Protein (%)	3.31	3.22	0.06	0.393	0.994	0.274
Protein (kg/d)	0.94	0.86	0.04	0.205	0.807	0.270
Lactose (%)	4.70	4.68	0.08	0.820	0.493	0.673
Lactose (kg/d)	1.37	1.28	0.07	0.360	0.671	0.118
SNF (%)	9.01	8.90	0.10	0.488	0.478	0.482
SNF (kg/d)	2.44	2.23	0.13	0.254	0.706	0.927
4% FCM (kg/d)	25.6	23.8	1.31	0.277	0.800	0.707
ECM (kg/d)	26.1	24.1	1.38	0.262	0.751	0.712
MUN (mg/dL)	9.88	6.39	0.23	0.008	0.629	0.255
Milk yield/DMI	1.32	1.28	0.05	0.469	0.788	0.069
Milk N/N intake	0.28	0.30	0.01	0.242	0.963	0.441
Water intake (L/d)	118	112	6.46	0.473	0.409	0.223
ADG (kg/d)	-0.21	-1.02	0.17	0.079	0.923	<0.001
BCS change (units/period)	-0.02	0.10	0.03	0.119	0.492	<0.001

¹Treatments: (1) HP2 = formulated to contain 15.4% CP and 169 g/d of MP balance with soybean meal supplemented with rumen-protected Met (Metaspart, Adisseo, Antony, France); (2) LP2 = formulated to contain 13.5% CP and -59 g/d of MP balance with dry distillers grains plus solubles supplemented with rumen-protected Lys (AminoShure-L, Balchem, New Hampton, NY) and rumen-protected Met (Metaspart, Adisseo).

MUN. Milk N efficiency tended to be higher for LP (0.22 vs. 0.27; $P \leq 0.08$) in trial 1 with corn silage-based diets, but not when ryegrass silage-based diets were fed in trial 2 (0.28 vs. 0.30; $P \geq 0.24$). Conversely, ADG and BCS were similar between treatments in trial 1, but LP2-fed cows tended ($P \leq 0.08$) to lose more weight than HP2-fed cows in trial 2, although both groups of cows maintained BCS. Combined, the results suggest that MP may have been sufficient when DDGS plus RPM and RPL replaced SSBM in trial 1, but overall efficiency may have been influenced by forage source, as performance in trial 2 appeared to be partially supported by mobilized body protein.

In contrast to our study, milk yield was lower when cows were fed corn silage-based diet compared with diets containing alfalfa hay or a mix of both when protein levels were plentiful, but protein content varied among treatments (16.9 vs. 18.3 and 18.7% CP, respectively; Kleinschmit et al., 2007). Apparent recovery of AA for milk protein synthesis had higher efficiency when cows were fed diets with a negative balance of MP compared with diets with a neutral balance, but the uptake of AA at the mammary gland was not due to RPM and RPL supplementation (Lee et al., 2015).

Considering optimum MUN values range from 8.5 to 11.5 mg/dL (Kohn et al., 2002), MUN appeared to corroborate milk yield and components yield in both trials. Treatments at (LP1 = 9.93 mg/dL, trial 1; and HP2 = 9.88 mg/dL, trial 2) or above (HP1 = 17.2 mg/dL) recommended MUN range appeared to adequately support production. Conversely, cows in LP2 had very low MUN (6.39 mg/dL), but milk yield and milk protein yield differences were not significant in trial 2. Several low-MP balance studies resulted in linear decrease in MUN (Leonardi et al., 2003; Broderick et al., 2008; Lee et al., 2012b). When 13.5% CP diets without RPAA were fed to mid-lactation dairy cows, MUN was 7.7 mg/dL (Colmenero and Broderick, 2006). No effect on milk yield was found for cows on late-lactation fed either a high-CP, low-RUP diet and low-CP, high-RUP diet similar to our study (Kalscheur et al., 1999). Our data suggests that late-lactation cows fed a low-CP diet with RPAA will maintain milk production compared with high-protein diets without AA supplementation.

Income over feed cost per cow per day was calculated based on feed costs and income from milk at the time of the experiments. For trial 1, income over feed cost was \$4.36 for HP1 and \$4.64 for LP1, with a slightly greater return (\$0.30) for the diet containing DDGS with RPL and RPM with corn silage-based diets. In contrast, return was nearly \$1 lower for LP2 than HP2 (HP2 = \$3.61; LP2 = \$2.79) in ryegrass silage-based diets (trial 2).

In conclusion, substituting soybean meal with dry distillers grains adjusted for RPL and RPM maintained performance in late-lactation cows fed corn silage- and ryegrass silage-based diets containing marginal MP levels in short-term studies. Our results suggest that dietary protein levels can be reduced for late-lactation cows without detriment to production and potentially reducing ammonia emissions to the environment. Further investigation is necessary to elucidate the role of marginal dietary protein levels on cows' homeorhesis and AA utilization efficiency diets based on corn silage and ryegrass silage.

ACKNOWLEDGMENTS

The authors are grateful to Adisseo (Antony, France; Daniel Luchini) and Balchem (New Hampton, NY; Michael de Veth) for the donation of the amino acid supplements Metaspart and Aminosure-L, respectively. We are also grateful to Bill Barber, Doug McKean, Dennis Morgan, Shannon Forbes, Tara Martin, and the rest of the Louisiana State University Agricultural Center Southeast Research Station (Franklinton, LA) personnel for their support of this research.

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