Energy content of reduced-fat dried distillers grains with solubles for lactating dairy cows

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ABSTRACT

Eight Holstein and 8 Jersey multiparous, lactating cows were used to complete 56 energy balances to determine the energy content of reduced-fat dried distillers grains with solubles (RFDDGS). A repeated switchback design was used to compare treatments with and without RFDDGS. Diets consisted of 24.2% corn silage, 18.4% alfalfa hay, 6.94% brome hay with either 22.9% rolled corn or 14.8% soybean meal (control), or 8.95% rolled corn, 28.8% RFDDGS, and 0% soybean meal [Co-P; dry-matter (DM) basis]. The inclusion of RFDDGS did not affect DM intake, averaging 21.4 ± 0.53 kg of DM for all cows, but milk production tended to increase from 29.8 to 30.9 ± 1.46 kg/d for control and Co-P treatments, respectively. Milk fat percentage and energy-corrected milk did not differ between treatments, averaging $4.33 \pm 0.14\%$ and 34.1 kg/d, respectively. Milk protein was significantly decreased by the Co-P treatment (3.56 and $3.41 \pm 0.08\%$ for control and Co-P treatments), but protein yield was not affected. Milk energies were 1.40 Mcal/d greater with Co-P. Energy lost as methane was reduced by 0.31 Mcal/d with the addition of RFDDGS to the diet. Heat loss averaged 29.9 ± 0.55 Mcal/d and was not different between diets. Average energy retained as tissue energy was -2.99 ± 0.93 Mcal/d and did not differ between treatments. Intake of digestible and metabolizable energy were not different between the control and Co-P treatments, averaging 2.68 and 2.31 Mcal/kg of DM, respectively. The net energy of lactation values of control and Co-P diets were calculated to be 1.43 and 1.47 Mcal/kg of DM, respectively. These energy estimates suggest greater energy content of diets containing RFDDGS than diets containing a mixture of corn and soybean meal in lactating dairy cows.

Key words: dairy cow, energy balance, indirect calorimetry, reduced-fat dried distillers grains with solubles

INTRODUCTION

Dry distillers grains with solubles (DDGS), a by-product of ethanol production from corn grain, is most commonly produced in the midwestern United States and often included in dairy rations. In recent years, technology has been developed to remove a portion of the oil so that it may be used in biodiesel production. This process results in a reduced-fat dried distillers grains with solubles (RFDDGS; Berger and Singh, 2010). This RFDDGS has been used as a protein and energy source in lactating dairy cow diets, with fat concentrations low enough to reduce the risk of milk fat depression that may be associated with diets high in fat (Bauman and Griinari, 2003). The nutritional value of RFDDGS has not been investigated to the extent of full-fat DDGS, and the effects of RFDDGS on energy utilization of lactating cows has not yet been evaluated. When replacing forages, corn, soybean meal, and soy products, the inclusion of RFDDGS has been reported to have no effect on milk fat percentage (Castillo-Lopez et al., 2014), or to increase milk fat percentage with no negative effect on milk production (Mjoun et al., 2010). Given that the fat content is decreased, it is speculated that the energy content of RFDDGS is also less than DDGS. As a consequence, the determination of the energy value of diets containing RFDDGS will allow for more precise formulation of diets for lactating dairy cows. The objective of this study was to use total collection and indirect calorimetry techniques to investigate the effect of including RFDDGS on energy and N utilization in lactating cow diets when replacing corn grain and soybean meal. It was hypothesized that diets containing RFDDGS would contain less energy, and as a result, cows consuming RFDDGS would produce less milk.

MATERIALS AND METHODS

Experimental Design

Sixteen multiparous Holstein ($n = 8$) and Jersey ($n = 8$) cows averaging 93 ± 20 DIM at the beginning of the

Received December 10, 2014.

Accepted June 9, 2015.

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experiment were used, with average BW of 693.8 ± 12.9 and 429.2 ± 13.0 kg, respectively. The experimental design and methodology were similar to that of Birkelo et al. (2004). Two treatments were compared in a 4-period repeated switchback (Cochran and Cox, 1957) within a split-plot design. Cows were randomly assigned 1 of the 2 dietary treatments (control or **Co-P**; Table 1), which alternated over 4 periods; thus, measurements were collected on each animal consuming each treatment during 2 nonconsecutive experimental periods. Animals were blocked by date of calving, and the subplot of this study was breed, which was duplicated. The objectives of the current study were not to examine and report breed effects, but those results were reported elsewhere (G. Garcia Gomez, A. J. Foth, T. Brown-Brandl, H. C. Freetly, and P. J. Kononoff; unpublished data). Two diets were formulated that differed in the proportion of RFDDGS (Poet Nutrition, Sioux Falls, SD) included in the formulation. Diets included the control, which did not contain any RFDDGS, and Co-P, in which the coproduct RFDDGS was included at 30% of the diet DM while partially replacing the corn and soybean meal in a similar strategy as Birkelo et al. (2004). Specifically, the proportion of forage was held constant between treatments, but they differed in concentrate formulation. In the Co-P diet, RFDDGS replaced all the soybean meal and approximately half of the ground corn of the control diet. Diets were balanced using the Cornell-Penn-Miner Dairy model (Boston et al., 2000) to contain similar concentrations of CP but they differed in predicted energy, as this is what was tested. The study was conducted over 16 mo because the blocks were not tested simultaneously, consequently forages varied by year. Diet compositions and nutrient analysis are presented in Table 1. Each experimental period was 35 d in duration with 28 d for ad libitum diet adaptation, followed by 7 d of collection and 95% ad libitum feeding to minimize refusals, similar to the methodology in Birkelo et al. (2004). During the 28-d diet adaptation, cows were fed for ad libitum consumption to allow for approximately 5% refusals. All cows were less than 90 d pregnant at the conclusion of the final experimental period. Cows were housed in a temperature-controlled barn at the Dairy Metabolism Facility in the Animal Science Complex of the University of Nebraska-Lincoln in individual tie-stalls equipped with rubber mats and milked at 0700 and 1800 h. After milking all cows were moved to an indoor drylot sand surfaced pen for exercise where they were held for approximately 1 h. All animal care and experimental procedures were approved by the University of Nebraska-Lincoln Animal Care and Use Committee. Control and Co-P diets contained corn silage, alfalfa hay, grass hay, and concentrate mixed

Table 1. Ingredient composition of control and coproduct (Co-P) diets and analyzed chemical composition (mean $\pm$ SD) used to determine apparent digestibilities

Item	Diet	
	Control	Co-P
Ingredient, % of DM		
Corn silage	24.5	24.5
Alfalfa hay	18.4	18.4
Brome hay	6.94	6.94
Ground corn	22.9	8.95
RFDDGS ¹	—	28.8
Soybean meal	14.8	—
Ground soybean hulls	7.93	7.93
Soypass ²	2.01	2.01
Calcium carbonate	0.89	0.89
Sodium bicarbonate	0.65	0.65
Calcium diphosphate	0.30	0.30
Salt	0.22	0.22
Magnesium oxide	0.18	0.18
Trace-mineral premix ³	0.12	0.12
Vitamin premix ⁴	0.12	0.12
Chemical composition, ⁵ % DM	1.71	1.67
CP	18.6 $\pm$ 0.77	19.0 $\pm$ 1.00
Ether extract ⁶	2.26 $\pm$ 0.11	3.22 $\pm$ 0.18
NDF	36.7 $\pm$ 1.91	43.4 $\pm$ 1.37
Ash	7.66 $\pm$ 0.57	8.38 $\pm$ 0.62
Starch	26.4 $\pm$ 1.47	17.9 $\pm$ 1.31
NFC ⁷	34.9 $\pm$ 2.00	26.1 $\pm$ 2.41
ME, ⁸ Mcal/kg	2.67	2.46
Gross energy, cal/g	3,970.8 $\pm$ 77.9	4,114.8 $\pm$ 92.4

¹Reduced-fat dried distillers grains with solubles.

²LignoTech, Overland Park, Kansas.

³Contained 13.9% Ca, 0.03% P, 0.42% Mg, 0.20% K, 4.20% S, 0.08% Na, 0.03% Cl, 445 mg/kg of Fe, 60,021 mg/kg of Zn, 17,375 mg/kg of Cu, 43,470 mg/kg of Mn, 287 mg/kg of Se, 527 mg/kg of Co, and 870 mg/kg of I.

⁴Formulated to supply approximately 120,000 IU/d of vitamin A, 24,000 IU/d of vitamin D, and 800 IU/d of vitamin E in total ration.

⁵Determined from composite samples collected throughout the experiment and analyzed at the University of Nebraska-Lincoln, mean $\pm$ SD.

⁶Analyzed by Cumberland Valley Analytical Services, Hagerstown, Maryland.

⁷NFC = 100 - (% NDF + % CP + % fat + % ash) (NRC, 2001).

⁸Calculated using the Cornell-Penn-Miner Dairy model (Boston et al., 2000).

as a TMR, which was mixed in a Calan Data Ranger (American Calan Inc., Northwood, NH). Cows were fed once daily at 0900 h.

Individual feed ingredients were sampled (500 g) each day during the collection period and frozen at -20°C . They were later composited by period and a subsample sent to Cumberland Valley Analytical Services Inc. (Hagerstown, MD) for complete nutrient analysis of DM (AOAC International, 2000), N (Leco FP-528 N Combustion Analyzer; Leco Corp., St. Joseph, MI), NDF (Van Soest et al., 1991), ADF (method 973.18; AOAC International, 2000), sugar (DuBois et al., 1956), ether extract (2003.05; AOAC International, 2006), ash (942.05; AOAC International, 2000), and

minerals (985.01; AOAC International, 2000). Ingredient and RFDDGS analysis are presented in Table 2. Total mixed rations were sampled on each day of collection and used to determine particle size according to Kononoff et al. (2003) using the Penn State Particle Separator. Total fecal and urine outputs were collected from each individual cow during the collection period for 2 consecutive days. Feces were collected using aluminum pans placed in the gutter behind the stall, and urine was collected using a noninvasive urine cup collector (Lascano et al., 2010) and accumulated into a Surge (Hinsdale, IL) bucket milker. Urine was deposited 4 times a day into 55-L plastic containers and acidified with 50 mL of concentrated HCl, before subsampling and freezing (-20°C). Subsamples of milk (100 mL), feces (4% wet basis), urine (2% wet basis), and gas (10 to 15 L) were collected. Samples were later thawed and composited for each cow during each period. Likewise, fecal samples were deposited into large containers (Rubbermaid, Wooster, OH), subsampled, and frozen (-20°C). Subsamples of gas (10 to 15 L) were also collected. Samples of feces, orts, and each feed ingredient and the TMR were composited according to cow and period, dried at 55°C in a forced-air oven, and ground to pass through a 1-mm screen (Wiley mill, Arthur H. Thomas Co., Philadelphia, PA). Ground samples were analyzed for DM (100°C oven for 24 h). Milk production was measured daily and milk samples (40 mL) were collected during the a.m. and p.m. milkings for the 2 d of collection for each animal and preserved using 2-bromo-2-nitropropane-1,3 diol. Milk samples were analyzed for fat, true protein, lactose, SCC, and MUN (AOAC International, 2000) using a B2000 Infrared Analyzer (Bentley Instruments, Chaska, MN) by Heart of America DHIA (Manhattan, KS).

Feed samples, orts, and fecal samples were analyzed at the University of Nebraska–Lincoln for N (Leco FP-528, Leco Corp.), NDF (Van Soest et al., 1991), starch (Megazyme, AOAC method 996.11 and AACC method 76.13), and ash (AOAC International, 2000). Heat-stable α -amylase (0.5 mL per sample; number A3306; Sigma Chemical Co., St. Louis, MO) and sodium sulfite were included in the NDF procedure. Samples were analyzed for ether extract (AOAC International, 2000) by Cumberland Valley Analytical Services Inc. (Hagerstown, MD). Urine and milk samples were analyzed for N as previously described. All samples including feed, orts, feces, urine, and milk were analyzed for gross energy (Parr 1241 Adiabatic Calorimeter, Moline, IL). Prior to analysis, milk and urine samples were lyophilized (VirTis Freezemobile 25ES, SP Scientific, Gardiner, NY).

Heat production (**HP**) was determined through the use of headbox-type indirect calorimeters, which were

constructed at the University of Nebraska–Lincoln. Prior to collections, 3 headboxes were used to test the rate of recovery of gas by burning 100% ethyl alcohol in the sealed headbox and comparing this measure to calculated gas concentrations. These calculations were based on weight of alcohol burned and a measured volume of gas sample. Three lamp runs were conducted. Recovery rates of O_2 and CO_2 averaged 101.8 ± 3.21 and $100.8 \pm 3.51\%$, respectively.

Collection for each cow consisted of 2 consecutive 23-h intervals where gas concentrations were averaged for each interval. Feed was placed in the headbox, and ad libitum access to water was available from a water bowl inside the box. Doors were closed and the vacuum motor turned on 15 min before the start of collecting to allow for air equilibrium. Temperature and dew point within the box were recorded every minute using a probe (Model TRH-100, Pace Scientific Inc., Mooresville, NC) connected to a data logger (Model XR440, Pace Scientific Inc.). Line pressure was measured from a manometer (Item # 1221-8, United Instruments, Westbury, NY), and barometric pressure of the room was recorded using a barometer (Chaney Instruments Co., Lake Geneva, WI). Total volume of gas was measured using a gas meter (Model AL425, American Meter, Horsham, PA), and continuous proportional samples of outgoing and incoming air were diverted to collection bags (61×61 cm, 44 L; PMC, Oak Park, IL) using glass tube rotameters (Model 1350E Sho-Rate “50,” Brooks Instruments, Hatfield, PA). Gas samples were analyzed (Emerson X-stream 3-channel analyzer, Solon, OH) according to Nienaber and Maddy (1985) and corrected for pressure and temperature within the box. Heat production was estimated by calculation from oxygen consumption, and carbon dioxide and methane production with correction for urinary N loss according to Brouwer (1965) with gases values reported in liters and mass of urinary N reported in grams (Equation 1). The respiratory quotient was calculated as the ratio of CO_2 produced to O_2 consumed. Volume of CH_4 formed was multiplied by a constant (9.45 kcal/L) to estimate the amount of energy represented in the formation of gaseous products. Energy balance was adjusted for excess N intake according to Moe et al. (1970) using the following equations:

$$\text{HP} = 3.866 \times \text{O}_2 + 1.200 \times \text{CO}_2 - 0.518 \times \text{CH}_4 - 1.431 \times \text{N}, \quad [1]$$

$$\text{ME} = \text{intake energy} - \text{fecal energy} - \text{urinary energy} - \text{CH}_4 \text{ energy}, \quad [2]$$

$$\text{recovered energy (RE)} = \text{ME} - \text{HP}, \quad [3]$$

Table 2. Analyzed chemical composition (% DM) of feedstuffs from samples collected each experimental period and determined by a commercial feed testing laboratory of a control diet and coproduct (Co-P) diet containing 28.8% reduced-fat dried distillers grains with solubles (RFDDGS; Poet Nutrition, Sioux Falls, SD)¹

Chemical, % DM	Corn silage		Alfalfa hay		Brome hay		Control concentrate		Co-P concentrate		RFDDGS	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
DM	38.5	2.15	86.6	1.85	86.7	2.45	88.8	0.50	89.6	0.76	89.1	0.94
CP	7.76	0.49	20.5	1.48	14.7	1.68	24.1	0.86	24.1	0.64	32.3	1.13
Soluble protein	4.02	0.53	4.59	0.65	3.53	0.79	4.48	0.82	3.60	0.47	14.3	1.51
ADICP	0.86	0.11	2.74	1.39	1.42	0.71	0.77	0.26	1.56	0.49	5.80	0.71
NDICP	1.17	0.32	6.09	2.42	5.35	1.42	1.76	0.34	3.28	1.47	8.28	1.49
ADF	25.3	0.97	32.5	4.01	38.3	2.46	9.24	0.91	13.3	1.41	9.35	0.60
NDF	39.5	3.29	43.9	5.13	66.3	2.67	16.8	1.68	29.4	3.27	31.4	1.45
Lignin	3.11	0.47	7.62	1.18	4.2	0.29	1.35	0.49	2.70	0.74	3.27	0.47
NFC ²	45.6	2.89	28.5	2.78	11.2	2.53	50.0	1.55	36.5	2.42	27.0	1.02
Starch	35.7	2.62	2.59	0.67	0.88	0.57	34.7	1.83	19.2	1.44	7.45	0.73
Sugar	0.92	0.27	2.70	1.22	4.75	1.21	7.07	0.69	4.15	0.86	4.68	0.93
Ether extract	3.24	0.34	1.99	0.36	2.33	0.26	2.55	0.50	4.54	0.69	6.16	0.13
Ash	5.08	0.65	11.2	0.39	10.3	0.61	8.37	0.74	8.78	0.43	5.83	0.48
Ca	0.30	0.13	1.32	0.13	0.38	0.04	1.54	0.21	1.35	0.33	0.04	0.01
P	0.23	0.03	0.33	0.04	0.35	0.03	0.59	0.05	0.90	0.23	1.00	0.07
Mg	0.14	0.03	0.23	0.02	0.14	0.01	0.39	0.01	0.49	0.03	0.41	0.01
K	1.05	0.09	3.54	0.34	3.35	0.52	1.37	0.06	1.21	0.04	1.36	0.10
S	0.13	0.02	0.27	0.03	0.19	0.02	0.25	0.01	0.61	0.05	0.97	0.05
Na	0.01	0.00	0.02	0.01	0.01	0.00	0.61	0.09	0.78	0.08	0.33	0.03
Cl	0.17	0.03	0.33	0.06	1.32	0.08	0.40	0.15	0.43	0.05	0.22	0.04
Fe, mg/kg	209.5	83.0	212.5	42.9	187.8	30.9	275.3	31.1	413.7	163.0	104.5	13.9
Zn, mg/kg	25.0	3.80	29.1	4.56	28.5	3.18	199.1	30.5	212.3	26.3	80.5	5.74
Cu, mg/kg	8.27	4.28	8.13	1.30	10.3	3.99	62.3	6.98	56.1	7.39	1.75	0.50
Mn, mg/kg	27.0	8.49	33.8	5.00	31.3	7.86	135.9	18.3	162.3	22.7	20.8	1.50

¹Values determined by Cumberland Valley Analytical Services, Hagerstown, Maryland. ADICP = acid-detergent-insoluble CP; NDICP = neutral-detergent-insoluble CP.²NFC = 100 - (% NDF + % CP + % fat + % ash) (NRC, 2001).

tissue energy (**TE**) = RE – milk energy, and [4]

ME for RE (**ME_{RE}**) =

ME – ME for maintenance. [5]

Metabolizable energy for maintenance was determined by regression of RE on ME and is the ME at zero RE (Figure 1). Lactation energy received from ME of feed (**LE_{ME}**) was defined as milk energy for cows in negative energy balance and was equal to milk energy plus TE multiplied by a constant estimated by Moe et al. (1970) for the efficiency of ME use for milk production from TE for lactating animals in positive energy balance (Equation 6).

LE_{ME} (positive energy balance) =
milk energy + TE × 0.84. [6]

Metabolizable energy available for lactation (**ME_{LE}**) was defined as **ME_{RE}** for cows in positive energy balance and was equal to **ME_{RE}** minus TE divided by a constant for the efficiency of body gain from ME (Equation 7; Moe et al., 1970). Tissue energy in protein was calculated using Equation 8 and was defined as energy used for tissue protein synthesis (Freetly et al., 2006).

ME_{LE} (negative energy balance) =

ME_{RE} – TE/0.726. [7]

TE in protein = N balance × (5.88 kg of protein/

kg of N) × (5.7 Mcal/kg of protein). [8]

Statistical Analysis

Data were analyzed using the GLIMMIX procedure of SAS (SAS Institute Inc., Cary, NC). Treatment, breed, breed within block, and period within block and breed were modeled as fixed effects, and cow within block, based on calving date, was modeled as a random effect. The LSMEANS option was used to generate least squares means of treatments listed in this study. In cases where data were missing, the highest standard error of treatment means is reported. Significance was declared at $P \leq 0.05$ and trends at $0.05 < P \leq 0.10$.

RESULTS AND DISCUSSION

Fifty-six of a possible 64 energy balances were completed. Gas meter calibration was not completed in time and diet composition was altered after the first collection period of the first block, so the data from those 4 cows were not used for that period. One Jersey

Figure 1. Regression of recovered energy (milk + tissue energy) on ME (intake energy – fecal energy – urinary energy – methane energy) in kilocalories per metabolic BW (MBW; $y = 0.7614x - 158$; $R^2 = 0.86$). Recovered energy = 0 at 158 kcal/MBW, and efficiency of converting ME to lactation energy (k_l) is 76%.

cow in block 4 died from a nonrelated source (intestinal intussusception) after the first collection period of that block. During the third collection of block 2, one Jersey cow became ill and was removed from collections for that period. For a period in block 3, collection was reduced to 1 d instead of 2 consecutive days to avoid switching corn-silage sources during collections.

Diet Composition

Chemical composition of individual ingredients and diet composition as estimated by a commercial feed testing laboratory are listed in Tables 2 and 3, and analysis of RFDDGS is found in Table 2. Diets were formulated to have similar concentrations of CP and were observed to be $18.8 \pm 0.23\%$ CP (DM basis). Ether extract was 1% greater (DM basis) in the Co-P diet than the control diet. This was expected because of the greater fat content in RFDDGS compared with corn and soybean meal. The NDF content of the control diet was $30.8 \pm 0.69\%$ (DM basis), which was lower than the Co-P diet at $37.1 \pm 0.89\%$ (DM basis). This is typical of RFDDGS; in a study by Castillo-Lopez et al. (2014), NDF content increased by 2.9% in a diet with 30% RFDDGS compared with a control diet without RFDDGS. However, Mjoun et al. (2010) observed little difference in NDF content of diets with increasing levels of RFDDGS from 0 to 30%, but this was a function of removing soybean hulls as a source of NDF.

Diet particle size was similar between treatments with 2.85, 20.7, 45.3, and 31.1% remaining on the >19.0 mm, 19.0 to 8.0 mm, 8.0 to 1.18 mm, and <1.18 mm pans, respectively, for the control TMR and 2.87, 19.9, 41.4, and 36.1% for the RFDDGS TMR (Table 3). According to Kononoff et al. (2003), it is recommended that rations should include 30 to 50% of particles between 8.0 and 19.0 mm and 10 to 20% particles between 1.18 and 8.0 mm in diameter to maximize milk production and to avoid milk fat depression. The proportion of particles in diets between 8.0 and 19.0 mm in the current study is lower than recommended, and particles between 1.18 and 8.0 mm in diameter is greater.

Intake, Milk Production, and Composition

Dry matter intake did not differ ($P = 0.86$) between treatments and averaged 21.3 ± 0.53 kg/d (Table 4). During collection, animals were offered feed at 95% of their ad libitum intake, but refusals averaged 1.49 ± 1.39 kg/d (DM basis), or $7.0 \pm 6.5\%$. Hünerberg et al. (2013) also observed a reduction in DMI during gas collection. Similar to the current study, Mjoun et al. (2010) observed no change in DMI with increasing levels of RFDDGS compared with a control without RFDDGS.

Table 3. Computed mean and SD of the chemical composition of control and coproduct (Co-P) with 28.8% reduced-fat dried distillers with solubles diets and measured particle distribution diets¹

Item	Control		Co-P	
	Mean	SD	Mean	SD
Chemical, % DM				
DM	75.9	0.32	76.3	0.35
CP	18.8	0.23	18.8	0.21
Soluble protein	4.32	0.18	3.88	0.13
ADICP	1.20	0.12	1.60	0.14
NDICP	2.66	0.20	3.42	0.34
ADF	19.5	0.40	21.5	0.46
NDF	30.8	0.69	37.1	0.89
Lignin	3.14	0.15	3.81	0.18
NFC ²	42.3	0.54	35.5	0.65
Starch	26.7	0.43	18.9	0.38
Sugar	5.15	0.18	3.69	0.20
Ether extract	2.60	0.10	3.60	0.13
Ash	8.21	0.16	8.41	0.12
Particle size, ³ %				
>19.0 mm	2.85	0.66	2.87	0.74
19.0–8.0 mm	20.7	2.88	19.9	3.06
8.0–1.18 mm	45.3	4.86	41.4	6.15
<1.18 mm	31.1	5.58	36.1	4.91

¹Means and SD were calculated based on samples of each feedstuff collected during each period and estimated by a commercial feed testing laboratory (Cumberland Valley Analytical Services, Hagerstown, MD). ADICP = acid-detergent-insoluble CP; NDICP = neutral-detergent-insoluble CP.

²NFC = $100 - (\% \text{ NDF} + \% \text{ CP} + \% \text{ fat} + \% \text{ ash})$ (NRC, 2001).

³Determined using the Penn State Particle Separator on a wet basis (Heinrichs and Kononoff, 2002).

However, in a study increasing RFDDGS as a replacement of forage, Castillo-Lopez et al. (2014) observed an increase in DMI from 23.8 kg/d with RFDDGS at 10% of DM to 27.9 kg/d with 30% RFDDGS. In the next experiment, they observed no difference in DMI. In a comparison and in a study testing DDGS from 3 different ethanol plants, no difference in intake was observed across sources (Kleinschmit et al., 2006). Benchaar et al. (2013) observed a linear increase in DMI of lactating dairy cows with increasing DDGS from 0 to 30% of the diet. Overall, the lack of change in DMI in the current study was expected and is comparable to many studies with RFDDGS or DDGS.

Milk yield tended ($P = 0.10$) to increase with the addition of RFDDGS to the diet. Milk fat percentage or yield did not differ ($P = 0.81$ and 0.14), and no difference between treatments was observed ($P = 0.22$) for ECM, averaging 34.1 kg/d (Table 4). Benchaar et al. (2013) reported a linear increase in milk production but a decrease in milk fat percentage with increasing concentration of DDGS. This resulted in a quadratic effect tendency for FCM and ECM to increase with DDGS up to 20% of diet DM but then decrease at 30%. Abdelqader et al. (2009) also observed that the inclusion of DDGS reduced milk fat with 30% DDGS

in the diet when compared with corn grain at 14%, potentially due to a difference in fat and a reduction in effective fiber. It is also possible that the fat contained in corn grain may be less available in the rumen than that in DDGS and thus may have a lesser effect on rumen fermentation. Schingoethe et al. (2009) suggest the greater volume of milk produced is due to the greater energy content of DDGS compared with a control diet with soybean meal. In the current study, the greater energy with the Co-P treatment, or more available energy, may be an explanation for increased production and FCM, but we cannot rule out the possibility that some energy may have also originated from the of body reserves. However, given the short duration of experimental periods, this is difficult to test using our current experimental design.

In a review on the use of distillers grains in lactating cow diets, Schingoethe et al. (2009) suggested that milk protein is seldom affected unless dietary protein is limiting. Additionally, Paz et al. (2013) reported that diets with 20% DDGS delivered sufficient protein and amino acids to maintain or increase milk protein synthesis. Contrary to this, in the current study, milk protein was ($P < 0.01$) reduced from 3.56 to $3.41 \pm 0.08\%$ with the addition of RFDDGS, but yield of protein was not affected ($P = 0.51$) because of increased milk production (1.04 and 1.02 ± 0.03 kg/d for control and Co-P treatments, respectively). It is possible that protein in RFDDGS is less available for milk production than in DDGS, but a more likely explanation is that the diet containing RFDDGS is deficient in lysine, which is possible for diets that rely on corn-based ingredients (Paz et al., 2013). In a meta-analysis, Paz et al. (2013) reported a positive trend in milk protein concentration with increasing lysine as metabolizable protein com-

pared with diets deficient in lysine, such as diets with a high proportion of DDGS.

Gas Consumption and Production

Oxygen consumption was similar ($P = 0.88$) between treatments, but CO_2 production and CH_4 production were reduced ($P \leq 0.01$ for both CO_2 and CH_4) with RFDDGS in the diet (Table 5). The respiratory quotient value averaged 1.07 ± 0.03 suggesting energy was being stored (Blaxter, 1962). Methane production was reduced from 504.2 ± 11.9 L/d with the control diet to 472.1 ± 11.6 L/d with the Co-P diet, a 7% reduction. The volume of CH_4 produced per kilogram of milk yield was also reduced ($P < 0.01$) by Co-P from 15.6 ± 0.54 to 14.1 ± 0.53 L of CH_4 /kg of milk. Similarly, Benchaar et al. (2013) reported a linear decrease in CH_4 production per kilogram of milk produced with an increasing rate of DDGS in the diet. This suggests that a portion of energy retained from reduced CH_4 loss was used for milk production, implying it is possible to increase milk production directly by reducing energy loss as CH_4 . Others have reported a reduction in CH_4 production with DDGS in dairy and beef cattle (McGinn et al., 2009; Benchaar et al., 2013; Hünerberg et al., 2013). In these studies, the high concentration of fat affecting the rumen environment and altering fermentation by suppressing methanogens and using hydrogen is often cited as the cause of reduced CH_4 (Benchaar et al., 2013). In support of this, the effect of added fat to ruminant diets has been shown to reduce CH_4 energy losses (Andrew et al., 1991; Holter et al., 1992; Grainger et al., 2010; van Zijderveld et al., 2011). In the current study, total dietary fat of the Co-P treatment was 3.22% on a DM basis, and it likely was not high enough to be the sole reason suppression of CH_4 production was observed. We suggest one potential reason is that total carbohydrate fermentation in the rumen was reduced. Nonetheless, Knapp et al. (2014) has suggested a 2% increase in diet ether extract may reduce CH_4 emissions by 10% from reduced DMI, suppression of protozoa and methanogen populations, or alternative hydrogen sinks from biohydrogenation. In the current study, it is also possible that the increased proportion of RFDDGS increased the extent of hindgut fermentation, which may increase enteric CH_4 production and would not be captured by the headbox system used. It has been estimated that 6 to 14% of methane produced in cattle originated from the hindgut (Immig, 1996).

Energy Partitioning

Gross energy intake (GEI) was greater ($P = 0.04$) with the Co-P treatment, but digestible energy (DE)

Table 4. Dry matter intake, milk production and composition, BW, and BCS¹ of control and coproduct (Co-P) with 28.8% reduced-fat dried distillers grains with solubles treatments

Item	Diet		SEM	P-value
	Control	Co-P		
DMI, kg/d	21.3	21.4	0.53	0.86
Milk yield, kg/d	29.8	30.9	1.46	0.10
ECM, ² kg/d	33.7	34.5	1.22	0.22
Fat, %	4.32	4.34	0.14	0.81
Fat yield, kg/d	1.24	1.28	0.05	0.14
Protein, %	3.56	3.41	0.08	<0.01
Protein yield, kg/d	1.04	1.02	0.03	0.51
MUN, mg/dL	16.9	16.6	0.43	0.58
BW, kg	564.0	559.0	9.32	0.14
BCS	3.30	3.29	0.06	0.81
ECM/NE _L intake, kg/kg	1.10	1.10	0.03	0.81

¹BCS = 1-to-5 scale according to Wildman et al. (1982).

²ECM = $0.327 \times \text{milk yield (kg)} + 12.95 \times \text{fat (kg)} + 7.20 \times \text{protein (kg)}$ adjusted for 3.5% fat and 3.2% total protein (DRMS, 2014).

and ME did not differ ($P = 0.22$ and 0.24 , respectively) by treatment (Table 6). Energy lost as feces was significant ($P = 0.05$) and was 2.06 Mcal/d greater with RFDDGS. Energy lost in urine tended ($P = 0.08$) to be significant and was 0.31 Mcal/d greater with RFDDGS. Energy lost as CH_4 was reduced ($P < 0.01$) from 4.77 ± 0.11 to 4.46 ± 0.11 Mcal/d with Co-P treatment, but HP did not differ ($P = 0.49$) between animals consuming the control and Co-P diets. Total RE was determined by adding milk and tissue energy but did not differ ($P = 0.18$) by treatment. Milk energy was 1.39 Mcal/d greater with Co-P and was ($P = 0.01$) greater due to increased milk production. Tissue energy, or energy balance, did not differ ($P = 0.73$). In a similar study by Birkelo et al. (2004) comparing wet corn distillers grains with solubles replacing corn grain and soybean meal, a decrease in GEI was reported, along with no difference in milk energy, resulting in a lower energy balance. This observation is contrary to our results; however, they also reported a reduction in DMI with wet distillers grains. In the current study, DMI did not differ between treatments, but energy content increased in the Co-P diet, which resulted in greater GEI with RFDDGS inclusion.

When expressed as a percentage of total GEI, partitioning of DE and ME did not differ ($P \geq 0.26$) between treatments. Fecal and urinary energy as a percentage of GEI also did not differ ($P \geq 0.26$), suggesting the increased energy outputs were solely due to greater energy intakes. Methane energy was significantly lower with Co-P when expressed as a percentage of GEI and was reduced ($P < 0.01$) from 5.72 to $5.13 \pm 0.14\%$. Similar to the current study, Birkelo et al. (2004) reported energy lost as CH_4 , when expressed as a percentage of GEI, was reduced by 14% with the inclusion of wet distillers grains and solubles. However, they did observe an increase in urinary energy as a percentage of GEI, contrary to our findings, potentially due to greater protein metabolism.

Table 5. Daily consumption of oxygen and production of carbon dioxide and methane for control and coproduct (Co-P) with 28.8% reduced-fat dried distillers grains with solubles treatments

Item	Diet		SEM	P-value
	Control	Co-P		
O_2 consumption, L/d	5,917.2	5,906.1	110.5	0.88
CO_2 production, L/d	6,379.9	6,202.9	108.4	0.03
CH_4 production, L/d	504.2	472.1	11.9	<0.01
CH_4 /kg milk produced	15.6	14.1	0.54	<0.01
Heat production, ¹ Mcal/d	29.5	29.3	0.55	0.62

¹Heat production (HP) calculated with Brouwer's (1965) equation from oxygen consumption (L), carbon dioxide production (L), methane production (L), and urine N (g) ($\text{HP} = 3.866 \times \text{O}_2 + 1.200 \times \text{CO}_2 - 0.518 \times \text{CH}_4 - 1.431 \times \text{N}$).

Table 6. Energy partitioning of control and coproduct (Co-P) with 28.8% reduced-fat dried distillers grains with solubles treatments

Item ¹	Diet		SEM	P-value
	Control	Co-P		
GE intake, Mcal/d	84.3	88.1	2.26	0.04
DE, Mcal/d	56.7	58.3	1.48	0.24
ME, Mcal/d	48.9	50.4	1.40	0.22
Component, Mcal/d				
Feces	27.8	29.9	1.03	0.05
Methane	4.77	4.46	0.11	<0.01
Urine	3.05	3.36	0.13	0.08
Heat	30.0	29.7	0.55	0.49
Retained	19.1	20.7	1.11	0.18
Milk	22.1	23.5	0.96	0.01
Tissue	-3.20	-2.78	0.93	0.73
Feces, % of GE	33.1	34.1	0.65	0.26
Methane, % of GE	5.72	5.13	0.14	<0.01
Urine, % of GE	3.62	3.83	0.17	0.35
DE, % of GE	66.9	65.9	0.65	0.26
ME, % of GE	57.6	57.0	0.66	0.51
GE, Mcal/kg of DM	3.96	4.11	0.01	<0.01
DE, Mcal/kg of DM	2.65	2.71	0.03	0.16
ME, Mcal/kg of DM	2.28	2.34	0.03	0.14
NE_L , Mcal/kg of DM	1.43	1.47	0.02	0.10

¹GE = gross energy; DE = digestible energy.

Energy estimates of diets are listed in Table 6. Gross energy content of the diet was greater ($P < 0.01$) at 4.11 ± 0.01 Mcal/kg of DM for the Co-P treatment compared with the control diet at 3.96 ± 0.01 Mcal/kg of DM. This is a result of greater energy content of the diet and greater DMI with RFDDGS inclusion. Digestible energy or ME content of diets did not differ ($P \geq 0.14$). Net energy for lactation for control and Co-P treatments tended ($P = 0.10$) to be greater for cows consuming RFDDGS and were 1.43 and 1.47 Mcal/kg of DM for control and Co-P, respectively. These values are lower than those calculated by Birkelo et al. (2004), with 1.82 Mcal/kg of DM for a diet with wet distillers grains included at 30% . Lower energy values for RFDDGS are expected when compared with full-fat distillers grains because of the reduced fat, or increased Maillard reaction compared with wet distillers grains. It is interesting to note that with a lower inclusion rate of ground corn in the diet in the Co-P compared with the control treatment, similar DE, ME, and NE_L values were determined. This may indicate an economic benefit for greater use of energy while feeding a low-starch diet.

Based on the energy content of the diet, we were able to calculate the energy content of RFDDGS by assuming energy values from the NRC (2001) for DE, ME, and NE_L of 3.53 , 3.12 , and 2.01 Mcal/kg of DM, respectively, for corn and 3.0 , 3.29 , and 1.94 Mcal/kg of DM for soybean meal. Estimated values for RFDDGS were calculated by difference and were 3.82 Mcal/kg of DE at $1 \times$ maintenance, 3.41 Mcal/kg of ME at $1 \times$

maintenance, and 2.03 Mcal/kg of NE_L at $3 \times$ maintenance. These values are lower than values determined for wet distillers grains by Birkelo et al. (2004) but similar to NRC (2001) values for ground corn. Feed efficiency as estimated as ECM per unit of NE_L intake was not observed to be different ($P = 0.81$) and averaged 1.10 ± 0.03 kg/kg (Table 4).

Estimation of maintenance energy requirements were determined through regression of ME and RE scaled for metabolic BW (**MBW**) and solving for ME when RE equals zero (Figure 1). Maintenance was calculated to be 208 kcal/MBW with an efficiency of ME use for lactation (k_l) of 0.76. These values are greater than previous estimates of maintenance energy requirements and efficiencies of lactation for mature lactating dairy cows (121 kcal/MBW, Vermorel et al., 1982; 136.2 kcal/MBW, Birkelo et al., 2004). Yan et al. (1997) reported maintenance estimates ranged from 146 to 179 kcal/MBW, with a mean of 160 kcal/MBW in a meta-analysis of energy metabolism trials in Northern Ireland and determined the k_l to range from 0.61 to 0.68. This is lower than that observed in the current study, suggesting our animals had greater maintenance energy requirements and were more efficient at converting ME to milk. Maintenance requirements have been shown to be greater for first-lactation heifers (Freetly et al., 2006; Xue et al., 2011), which could explain the greater values calculated in by Yan et al. (1997) with an unknown distribution of primiparous and multiparous animals. Animals in the current study were all multiparous, suggesting the high maintenance energy was not due to young age. Nonetheless, it is reasonable to accept maintenance estimates of the current study (208 kcal/MBW) because of the high level milk production, which would result in increased organ function to support milk synthesis, and therefore increased maintenance.

Nitrogen Balance and Digestibilities

Nitrogen partitioning or nitrogen balances (intake nitrogen minus fecal, urinary, and milk nitrogen production) did not differ between treatments ($P \geq 0.63$; Table 7). Nitrogen intakes were 641.6 ± 17.6 g/d, and balances were 60.5 ± 11.4 g/d. Others have shown differences in nitrogen partitioning with diet changes. However, responses may differ between studies. Gehman and Kononoff (2010) evaluated the effects of wet distillers grains with solubles on nitrogen balance and showed an increase in urinary and milk nitrogen excretion with the inclusion of distillers grains but also greater nitrogen balances. Contrary to these findings, Birkelo et al. (2004) reported wet distillers grains with solubles reduced fecal and milk nitrogen, and increased

Table 7. Nitrogen partitioning of control and coproduct (Co-P) with 28.8% reduced-fat dried distillers grains with solubles treatments in grams per day and as a percentage of nitrogen intake

Item	Diet		SEM	P-value
	Control	Co-P		
Mass, g/d				
N intake	637.4	645.9	17.6	0.63
Fecal N	194.9	198.9	6.29	0.53
Urine N	200.3	215.8	7.24	0.13
Milk N	178.1	173.1	7.71	0.50
N balance ¹	63.4	57.7	11.4	0.71
N intake, % of intake N				
Fecal N	30.9	30.8	0.64	0.92
Urine N	31.7	33.6	1.14	0.21
Milk N	28.2	27.0	1.02	0.27
N balance	9.15	8.53	1.67	0.78

¹Nitrogen balance = intake N – fecal N – urine N – milk N.

urinary nitrogen, resulting in similar nitrogen balances. In a study with increasing levels of DDGS, Benchaar et al. (2013) observed intake, fecal, urinary, and milk nitrogen increased linearly, resulting in greater nitrogen balances. Feeding DDGS to growing steers has also resulted in linear increases of nitrogen intakes and urinary nitrogen but decreasing fecal nitrogen excretion (Walter et al., 2012). It has been suggested that when used as an energy source, the high proportion of CP in DDGS may result in greater nitrogen excretion, but greater fecal nitrogen may also be the result of a greater extent of hindgut fermentation. Consequently, this would result in an overestimation of fecal nitrogen excretion or a greater amount of microbial nitrogen exiting the rumen from a greater digestible feed (Tine et al., 2001; McGinn et al., 2009). However, sampling error may also be a factor in determining nitrogen partitioning from loss of feed, through the volatile loss of nitrogen from urine or drying fecal samples, or nitrogen gas production (Walter et al., 2012).

Dry matter (**DMD**) and organic matter digestibilities (**OMD**) were reduced ($P < 0.01$) by 2.68% with the inclusion of RFDDGS in the diet (Table 8). Crude protein digestibility did not differ ($P = 0.92$), averaging $69.2 \pm 0.64\%$. Digestibility of NDF tended ($P = 0.09$) to increase from 49.3 ± 1.22 to $52.3 \pm 1.18\%$ with RFDDGS inclusion, and ether-extract digestibility was significantly improved ($P < 0.01$) by 5.20%. Starch or NFC digestibilities did not differ between treatments ($P = 0.29$ and 0.59), and values were similar for those components. Castillo-Lopez et al. (2014) fed diets with increasing increments of RFDDGS from 0 to 30% to lactating dairy cows and reported no difference in DMD or NDF digestibilities. Nitrogen and NFC digestibilities also tended to increase linearly with RFDDGS. However, balance of forage, corn, cottonseed, and soy-based feeds were altered to maintain similar CP, potentially

Table 8. Apparent digestibilities of control and coproduct (Co-P) with 28.8% reduced-fat dried distillers grains with solubles treatments

Component, %	Diet		SEM	<i>P</i> -value
	Control	Co-P		
DM	69.5	66.8	0.49	<0.01
OM	71.7	69.0	0.47	<0.01
Ash	42.0	43.3	1.85	0.58
CP	69.1	69.2	0.64	0.92
NDF	49.3	52.3	1.22	0.09
Ether extract	73.3	78.5	0.83	<0.01
Starch	96.0	96.6	0.43	0.29
NFC ¹	95.8	95.2	0.94	0.59

¹NFC = 100 - (% NDF + % CP + % fat + % ash) (NRC, 2001).

resulting in different digestibility responses compared with the current study with only corn-grain and soybean-meal inclusion changing. Another potential reason for the different DMD responses is differences in processing or heating (Hünerberg et al., 2013). Also, Benchaar et al. (2013) reported a decrease in DMD and OMD with DDGS and suggested the cause was the high concentration of fat in DDGS but may have also been due to increasing concentrations of NDF. Responses of NDF digestibility tended to be quadratic, increasing from 0 to 20% DDGS and then decreasing at 30% DDGS. The increase in NDF digestibility was suggested to result from highly digestible fiber in DDGS, but small particle size increased rumen passage rate at 30% DDGS, which reduced digestibility. This is not the case for the current study; even with the fine particle size, NDF digestibility was improved. Fat content of RFDDGS was relatively low compared with DDGS, so the reduction in DMD is most likely not a result of high fat but of less available nutrients for fermentation. The reduction in DMD and OMD with an increase in NDF and ether-extract digestibilities could be explained by a reduction in digestibility of other nutrients. However, digestibility of all other nutrients tested did not decrease. A decrease in DMD and an increase in NDF digestibility may be expected with RFDDGS, but it is unknown why both occurred.

CONCLUSIONS

Replacement of corn and soybean meal with RFD-DGS appeared to result in more net energy per unit of DM consumed, likely because this change resulted in less energy lost as CH₄. A greater NE_L value for the Co-P diet was a function of increased DMI and greater energy content. Dry matter digestibility and OMD were reduced with RFDDGS inclusion by 4%, but NDF digestibility was increased by 6%. The reduction in DMD, OMD, and CH₄ production by Co-P indicate an

alteration of rumen fiber digestion, which is the most likely explanation for improved milk production. The addition of RFDDGS to the diet did not affect nitrogen partitioning, balance, or excretion. Milk production may be improved without negative effects on milk fat yield with RFDDGS, but the concentration of milk protein may be reduced. Future research should evaluate the relationship between RFDDGS intake and rumen microbial populations present that may be causing the reduction in CH₄ production.

ACKNOWLEDGMENTS

The authors thank the employees of the University of Nebraska–Lincoln Dairy Research Facility for care and management of all experimental animals. Additional gratitude is expressed to Poet LLC (Sioux Falls, SD) for partial financial support and K. Karges, formerly of Poet LLC, for his feedback and ideas.

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