Feeding a diet with corn distillers grain with solubles to dairy cows alters manure characteristics and ammonia and hydrogen sulfide emissions from manure

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ABSTRACT

The objective of the experiment was to examine effects of a diet containing a high concentration (28.8% dry matter basis) of corn distillers grain with solubles on manure characteristics and NH_3 and H_2S emissions from dairy cow manure. Eighteen cows were blocked by parity and days in milk, and cows in each block were assigned to the following treatments: the control diet (CON) or CON with distillers grains with solubles at 28.8% (dry matter basis) replacing mainly soybean meal (DG). The experiment was conducted for 11 wk, and feces and urine from individual cows were collected over 3 d in wk 11 (a total of 8 spot samples per cow). Fecal or urine samples were composited by cow, and the composite feces and urine were analyzed for indigestible neutral detergent fiber and creatinine concentration, respectively, for individual cows to estimate total fecal and urine outputs. Immediately before the manure incubation, composited feces and urine were sampled to determine manure characteristics. Manure was reconstituted according to daily fecal and urine excretion estimated for individual cows. Individual manures were incubated using a continuous air flux multichamber system over 10 d to measure NH_3 and H_2S emissions. All data from 18 manures were analyzed using the Mixed procedure of SAS (SAS Institute Inc., Cary, NC). The ratio of feces to urine and the contents of manure total and volatile solids were not different among treatments. Urine from DG had lower pH and DG manure had lower N content and greater S content compared with CON. During the 10-d incubation, NH_3 emission was considerably less for DG versus CON. The emission of H_2S over 10 d for DG was greater compared with that for CON. After the incubation, manure pH and N and S concentrations were greater for DG versus CON. In conclusion, manure from cows fed a high-DG diet decreased urinary N contribution to manure N and

lowered urine pH, which were the factors that caused the decrease in NH_3 emission from DG manure. However, the DG diet increased dietary S concentration and increased S excretion in urine and feces. This increased H_2S emission from DG manure during the 10-d manure incubation.

Key words: corn distillers grains with solubles, manure, ammonia, hydrogen sulfide

INTRODUCTION

An increase in ethanol production has resulted in an increase in production of dry corn distillers grains with solubles (DDGS). Because of its nutritional values (i.e., high protein, fat, and fiber) and its cheap price relative to other high-protein feeds (e.g., soybean meal; **SBM**), DDGS has been used as a feed for livestock animals. In dairy cows, studies with distillers grains have focused on its nutritional values and production responses (Schingoethe et al., 2009). In terms of production, it is clear that high inclusion of DDGS in a ration (e.g., 30% in dietary DM) increases risk of milk fat depression (Ramirez-Ramirez et al., 2016; Morris et al., 2018a). However, fewer studies have looked at environmental impacts of feeding diets high in DDGS to dairy cows.

Improving the sustainability of dairy operations is a current primary goal in the US dairy industry, and lessening environmental impacts from dairy production is one of the critical tasks to improve sustainability (<https://nifa.usda.gov/program/sustainable-agriculture-program>). Therefore, the environmental impact as well as production responses should be considered when feedstuffs are evaluated to determine their nutritional values and inclusion rates in dairy rations. In terms of environmental impact, DDGS has been studied for its effects on enteric methane production in dairy cows (Benchaar et al., 2013; Foth et al., 2015). However, limited studies have been conducted with DDGS to determine other environmental impacts such as nutrient excretion and nutrient losses from manure (e.g., gas emissions). Manure is not a waste but good fertilizer for

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crops. However, when managed at farms or applied to the field improperly, it can negatively affect air (emissions of ammonia and hydrogen sulfide), water (N and P leaching and runoff), and soil quality (acidification; Aillery et al., 2005).

Ammonia and hydrogen sulfide emitted from manure can be an environmental concern because ammonia volatilization is not only a loss of manure N but also negatively contributes to the environment such as air quality (formation of particulate matter $<2.5\ \mu\text{m}$), farm odor, soil acidification, and water quality (N leaching and runoff; NRC, 2003). Hydrogen sulfide emission from manure has been less studied compared with ammonia, especially in dairy cows, so the degree of H_2S emitted from dairy manure is not known. Hydrogen sulfide is a dangerous gas causing potential human and animal health concerns (Costigan, 2003): serious injury or death at $>500\ \text{ppm}$, injury with chronic exposure at $10\ \text{ppm}$. When DDGS replaces SBM in a dairy ration, this may alter manure characteristics and NH_3 and H_2S emissions from manure. The content of RDP and RUP is different between DDGS and SBM (Mjoun et al., 2010; Maxin et al., 2013). Therefore, when SBM is replaced fully with DDGS, dietary RDP and RUP will change, and this could be a factor that shifts N excretion from urine to feces (Hristov et al., 2011; Morris et al., 2018b) and may decrease NH_3 emission from manure (Lee et al., 2011). Hydrogen sulfide emission from manure may increase when a DDGS diet is fed compared with a SBM diet because high S content in DDGS increases dietary S content and increases fecal and urine S concentrations (i.e., manure S concentration; Morris et al., 2018b).

The objective of this study was to examine manure characteristics (N of feces, urine, and manure) altered by feeding a high-DDGS diet and its effects on NH_3 and H_2S emissions from manure. Our hypotheses were (1) NH_3 emissions from manure of cows fed a DDGS diet will be decreased compared with cows fed a SBM diet because of a shift in N excretion from urine to feces and (2) because manure of cows fed a DDGS diet versus a SBM diet contains greater S, H_2S emissions will be greater.

MATERIALS AND METHODS

Animals and Treatments

All procedures in this project that involved animals were approved by The Ohio State University Institutional Animal Care and Use Committee (IACUC 2016A00000042).

This experiment was part of a larger experiment, and details about the cows used and experimental design

and procedures were reported in our companion papers (Morris et al., 2018a,b). Briefly, an animal experiment was conducted in a randomized complete block design. A total of 36 Holstein cows (average $\pm$ SD; DIM, 80 ± 31 ; BW, $675 \pm 74\ \text{kg}$) were randomly assigned to 3 dietary treatments (i.e., total of 12 blocks and 3 cows per block): **CON**, a control diet containing SBM as a main protein feed ingredient; **DG**, a diet containing reduced-fat corn distillers grain with solubles at 28.8% (DM basis); and the DG diet supplemented with monensin (20 mg/kg of DM; Rumensin, Elanco Animal Health, Greenfield, IN). In the current study only cows fed the CON and DG diets were used to determine manure characteristics and gas emissions from manure. The CON diet consisted of corn silage (41.6% on a DM basis), alfalfa silage (9.7%), alfalfa hay (5.0%), corn grain (12.9%), SBM (15.1%), soyhulls (12.3%), supplemental fat (1.34%; Energy booster; Milk Specialties, Eden Prairie, MN), and minerals and vitamins. For the DG diet, DG replaced SBM, soybean hulls, fat source, and supplemental phosphorus (Morris et al., 2018a). The chemical composition of CON and DG was essentially the same (DM basis; CP, 17.6%; NDF, 30.5%, crude fat, 4.5%; NE_L , 1.57 Mcal/kg) except for P (0.36 and 0.48%, respectively) and S (0.21 and 0.41%, respectively). The experiment lasted for a total of 12 wk (1-wk covariate, 2-wk diet adaptation, and 9-wk experimental period). All diets were prepared once a day in the morning and fed to cows around 0500 h. The diets were prepared as TMR and fed for ad libitum intake (5 to 10% of refusal) with free access to water.

Sample Collection and Measurement

Spot urine and feces were collected in wk 12 (the last week of the experimental period). Spot feces and urine were collected every 9 h over 3 d to have a total of 8 spot fecal and urine samples, representing every 3 h in a 24-h cycle (i.e., 0900 and 1800 h on d 1, 0300, 1200, and 2100 h on d 2, and 0600, 1500, and 2400 h on d 3). Spot fecal samples (about 200 g per sample) were collected from the rectum, composited by cow, and frozen at -20°C . Spot urine samples (about 150 mL per sample) were collected by massaging the vulva. The samples were composited by cow (about a total of 1.2 kg) and frozen at -20°C . Before freezing the composite urine samples, a subsample was taken and acidified with 2 M HCl for chemical analyses.

The composite fecal samples were subsampled, and the subsample was dried at 55°C and ground (Wiley mill; Arthur A. Thomas Co., Philadelphia, PA) through a 1-mm sieve. The ground samples were analyzed for DM at 100°C for 24 h and OM (method 942.05, AOAC International, 2000). The samples were also analyzed

for total N using a combustion method (Elemental Analyzer; Flash 2000; Thermo Fisher Scientific, Waltham, MA) after pulverizing. Total S concentration in fecal DM was determined using inductively coupled plasma emission spectroscopy after microwave digestion with nitric acid (Isaac and Johnson, 1985). The composite fecal samples and feed and refusal samples collected during the sampling week were analyzed for indigestible NDF according to Huhtanen et al. (1994) except filter bags with a 25- μ m pore size (Ankom Technology Corp., Macedon, NY) were used for the ruminal incubation. The concentrations of indigestible NDF were used to estimate fecal outputs of individual cows.

Acidified composite urine samples were analyzed for total N (Morris et al., 2019) and S using inductively coupled plasma emission spectroscopy. Urinary creatinine concentrations were assayed for individual cows (kit 500701; Cayman Chemical, Ann Arbor, MI), and these were used to estimate urine outputs of individual cows according to Valadares et al. (1999) assuming a creatinine excretion rate of 29.0 mg/kg of BW (Tebbe and Weiss, 2018). The BW of individual cows were measured on 2 consecutive days during the sample collection.

Manure Characteristics and Incubation

For manure incubation, 9 blocks of cows out of 12 blocks were randomly selected (i.e., $n = 9$ per treatment). Feces and urine for individual cows were thawed right before the incubation, and feces (Van Baale et al., 2004) and urine were measured for pH. Manures were reconstituted for individual cows according to the ratio of feces to urine determined previously [i.e., the estimated outputs of feces (DM and indigestible NDF) and urine (creatinine)]. Total N and S were determined using N and S concentrations of feces and urine, respectively, and the reconstitution ratio of feces to urine. After reconstituting manure, a subsample (50 g per sample) was collected to determine the content of TS (i.e., DM) and volatile solids (VS; i.e., OM) as described for feces.

Manure incubation lasted for 10 d in a continuous air flux multichamber system. This system was developed initially by Wheeler et al. (2011) and used in this experiment with few exceptions. Briefly, the system included a total of 9 manure chambers (i.e., 1 blank chamber (no manure) and 8 chambers to incubate manures). In this experiment, 7 chambers (1 blank and 6 manure chambers) were used at each incubation (total of 3 incubations and 6 manures per incubation). Manures of cows in the same block were incubated together. For example, the first incubation was conducted with manures of cows in blocks 1 to 3 (6 manures) followed

by manures from blocks 4 to 6 and then blocks 7 to 9. The capacity of each chamber is 17.5 L, and the amount of manure incubated for each chamber was 2 kg with 5 L/min air flow. The air generated from the pumps (DDL 15-101, Gast Manufacturing Inc., Benton Harbor, MI) is controlled by flow meters (VFB-67; Dwyer Instruments Inc., Michigan City, IN) for individual chambers. When manure is placed in the chamber, the chamber is closed (sealed) with a lid. The lid of the chamber has an inlet for air in and an outlet for air out through which air from the pump flows in, flushes the chamber, and exits the chamber. The outlet of the chamber is connected to exhaust via tubing (Teflon-lined PVC with 0.32-mm inside diameter and 0.64-mm outside diameter; Tygon SE-200 Saint Gobain, Akron, OH). A sampling line (0.16-mm inside diameter and 0.32-mm outside diameter) from the exhaust tubing is connected to a diaphragm sampling pump (KPM27H; Koge Electronics Co. Ltd., Taipei, Taiwan). The sampling pump delivers the exhaust air containing gases from manure into the sampling bag (12-L multilayer, foil gas sampling bag, Restek, Bellefonte, PA) according to the sampling schedule controlled by an automatic sampling system (Arduino UNO; <https://www.arduino.cc>). The sampling schedule used in this experiment was that all the sampling pumps for chambers were turned on to sample gases for 30 s every 30 min over 24 h. The sampling bags were replaced every 24 h during the experiment (i.e., 10-d incubation).

Gas samples collected over 24 h were analyzed for NH_3 using a photoacoustic gas analyzer (detection range, 0.1 to 5,000 ppm; Gasera one, Gasera Ltd., Turku, Finland) and H_2S concentrations (detection range, 3 ppb to 10 ppm of H_2S ; Jerome J605; Arizona Instrument LLC, Chandler, AZ). Gas production from manure was determined using the following equations with an example of NH_3 :

$$\begin{aligned} &\text{g of NH}_3/\text{kg of manure per h (A)} = \\ &[\text{NH}_3 \text{ concentration (ppm)} - \text{blank NH}_3 \\ &\text{concentration (ppm)}] \times \text{air flow (L/min)} \\ &\times \text{density (0.769 g of NH}_3/\text{L at standard} \\ &\text{temperature and pressure)} \times 60 \text{ min} \div 2, \end{aligned} \quad [1]$$

$$\begin{aligned} &\text{g of NH}_3/\text{cow per d (B)} = A \times 24 \times \text{estimated} \\ &\text{daily manure output, and} \end{aligned} \quad [2]$$

$$\text{Cumulative g of NH}_3/\text{cow} = \sum_{i=1}^{10} B, \quad [3]$$

where the final value was divided by 2 to express the emission per kilogram of manure (2 kg of manure was

incubated) in Equation 1, daily manure output was estimated using estimated daily fecal and urine outputs for individual cows in Equation 2, and i indicates day (i.e., d 0 to 10) in Equation 3.

After the 10-d incubation, all the chambers were opened and mixed well to take 3 subsamples. The first sample was analyzed for pH. The second sample was assayed for the content of TS (DM) and VS (OM) as described for feces. The third samples were acidified with 2 M HCl and freeze dried to determine total N and S concentrations. The analytical methods used here were the same as described previously for feces and manure.

Statistical Analyses

All data were analyzed using the Mixed procedure of SAS (version 9.4, SAS Institute Inc., Cary, NC). Data of BW, fecal and urine composition, and initial manure characteristics were analyzed using a model with block and block by dietary treatment as random effects and dietary treatment as a fixed effect. For data of manure characteristics after incubations, a similar model was used except that block, block by dietary treatment, and chamber within block were used as random effects. This model was modified to analyze gas emission rate (g of NH₃ or mg of H₂S per cow hourly), where time and interaction of treatment by time were added as fixed effects and repeated measures for time were included. Cumulative gas emissions over 10 d were analyzed with the same model without day effect. Significant differences among treatments were declared at $P \leq 0.05$. Differences at $0.05 < P \leq 0.10$ were considered a trend toward significance.

RESULTS

Daily outputs of feces, urine, and manure are shown in Table 1. Fecal excretion was not different between treatments. However, urine excretion was lower (27.7 vs. 31.3 kg/d; $P = 0.02$) for DG compared with CON. Manure production was not different between treatments. Despite a significant decrease in urine output for DG versus CON, the ratio of feces to urine did not differ ($P = 0.11$) between treatments.

Feeding the DG diet tended to decrease (6.41 vs. 6.66; $P = 0.09$) fecal pH (Table 2). Dry matter content and N concentration on an as-is basis did not differ between CON and DG. However, N concentration in feces tended to be lower (24.5 vs. 25.5 g/kg of DM; $P = 0.05$) for DG versus CON manure when expressed on a DM basis. Cows fed the DG diet had greater ($P < 0.01$) S excretion in feces. Cows fed the DG diet had lower urine pH (7.48 vs. 8.54; $P < 0.01$) and greater (P

< 0.01) S concentrations in urine compared with CON. However, N concentration in urine was not affected by DG versus CON. According to the differences in fecal and urine composition and excretion rates between CON and DG, manure composition was also altered. Manure N and S concentrations were lower (either DM basis or as-is basis; $P < 0.01$) for DG versus CON. However, TS and VS of manure were not affected ($P \geq 0.12$) by dietary treatments.

During the 10-d manure incubation, the rate of NH₃ emission had an interaction of diet by day ($P < 0.01$; Table 3). Cumulative NH₃ production was lower (27.5 vs. 46.1 g/cow; $P < 0.01$) for DG versus CON (Figure 1). Cows fed the DG diet versus CON had greater ($P \leq 0.03$) H₂S emission rates and cumulative H₂S emissions from manure over 10 d (Figure 2).

Manure characteristics after the 10-d incubation were also different between treatments (Table 4). Manure pH was lower (7.24 vs. 7.47; $P = 0.02$) for DG versus CON. The content of VS was greater ($P < 0.01$) for DG versus CON, without a difference in TS. The concentrations of N and S in manure on a DM basis were greater ($P < 0.01$) for DG compared with CON.

DISCUSSION

Although manure is valuable fertilizer, it may affect the environment (odor and air, soil, and water quality; NRC, 2003) in dairy production. The environmental impacts of manure can be largely reduced via good manure management (e.g., manure application rates, manure application method; Montes et al., 2013). However, various gases (e.g., NH₃, greenhouse gases, and H₂S) are produced from manure via microbial fermentation or chemical changes that may cause farm odor and a decrease in air quality at stalls and during

Table 1. Effects of feeding a diet containing high corn distillers grain on fecal, urine, and manure outputs (n = 9 per treatment)

Item	Diet ¹		SEM	P-value
	CON	DG		
BW, kg	721	757	21.2	0.23
Feces, ² kg of DM/d	7.5	7.7	0.36	0.69
Feces, kg/d as is	50.8	51.4	2.51	0.86
Urine, ³ kg/d	31.3	27.7	1.59	0.02
Manure, kg/d as is	82.1	79.1	3.33	0.42
Feces:urine	1.64	1.90	0.106	0.11

¹CON = manure from cows fed a basal diet containing soybean meal; DG = manure from cows fed the control diet with 28.8% corn distillers grains with solubles (DM basis) replacing mainly soybean meal and soyhulls.

²Estimated for individual cows using indigestible NDF content in feeds and feces.

³Estimated for individual cows using urine creatinine concentration.

Table 2. Effects of feeding a diet containing high corn distillers grain on fecal, urine, and manure characteristics on d 0 (n = 9 per treatment)

Item	Diet ¹		SEM	P-value
	CON	DG		
Feces				
pH	6.66	6.41	0.092	0.09
DM, g/kg	14.8	15.0	0.19	0.26
N, g/kg as is	3.77	3.68	0.079	0.34
N, g/kg of DM	25.5	24.5	0.34	0.05
N, g/d	190.4	187.8	7.09	0.80
S, g/kg as is	0.37	0.45	0.013	<0.01
S, g/kg of DM	2.51	2.99	0.08	<0.01
S, g/d	18.7	22.9	0.98	<0.01
Urine				
pH	8.53	7.48	0.125	<0.01
N, g/kg	9.31	8.62	0.648	0.31
N, g/d	287.2	231.4	12.72	<0.01
S, g/kg	0.64	2.15	0.081	<0.01
S, g/d	20.0	58.3	1.90	<0.01
Manure				
TS, ² g/kg	110.3	116.3	2.59	0.12
VS, ³ g/kg	865.3	870.3	3.01	0.25
N, g/kg as is	5.86	5.34	0.212	<0.01
N, g/kg of DM	51.8	44.5	1.85	<0.01
N, g/d	477.6	419.2	15.8	0.02
Fecal N contribution, %	40.5	45.3	1.46	<0.01
Urine N contribution, %	60.5	55.5	1.172	<0.01
S, g/kg as is	0.47	1.03	0.022	<0.01
S, g/kg of DM	4.31	8.87	0.175	<0.01
S, g/d	38.7	81.3	2.45	<0.01
Fecal S contribution, %	48.5	28.3	1.20	<0.01
Urine S contribution, %	51.5	71.7	1.20	<0.01

¹CON = manure from cows fed a basal diet containing soybean meal; DG = manure from cows fed the control diet with 28.8% corn distillers grains with solubles (DM basis) replacing mainly soybean meal and soyhulls.

²Dry matter of manure.

³Volatile solids (OM of manure DM).

manure storage before manure application to the field (Fangueiro et al., 2015). The degree of gas emissions from manure is dependent on internal and external factors. The external factor could be temperature, humidity, wind, bedding, manure storage system, and so on. The internal factor is manure characteristics (i.e., manure volume, nutrient concentration, and chemical forms of nutrients). The manure characteristics can be

altered by dietary nutritional composition. In the current study, therefore, a high-DG diet (28.8% DDGS in dietary DM) was evaluated for changes in manure characteristics and potential gas emissions from manure. However, the data of gas emissions should be interpreted with caution because manure was incubated in a laboratory in vitro system, which is appropriate to identify relative differences between treatments but is

Table 3. Effects of feeding a diet containing high corn distillers grain on ammonia and hydrogen sulfide emissions from manure over 10 d¹ (n = 9 per treatment)

Item	Diet ²		SEM	P-value ³	
	CON	DG		Diet	Int
NH ₃ , g/cow per h	0.19	0.11	0.007	<0.01	<0.01
Cumulative NH ₃ , g/cow	46.1	27.5	1.58	<0.01	
H ₂ S, mg/cow per h	2.06	3.43	0.438	0.03	0.46
Cumulative H ₂ S, mg/cow	499.0	828.1	94.8	<0.01	

¹Daily gas emissions measured were multiplied by total daily excretion of manure for emission data on a head basis.

²CON = manure from cows fed a basal diet containing soybean meal; DG = manure from cows fed the control diet with 28.8% corn distillers grains with solubles (DM basis) replacing mainly soybean meal and soyhulls.

³Diet = diet effect (CON vs. DG); Int = interaction of diet by time (diet × time).

not indicative of actual emissions of gases from manure at farms.

Although feeding the DG diet to dairy cows did not alter the total amount of manure production compared with the CON diet, a significant decrease in urine output was observed. Changes in urine output by DG are not well studied in dairy cows. In beef cattle studies, the opposite results were found where increasing proportion of DG in a ration significantly increased urinary outputs (Salim et al., 2012; Li et al., 2014). However, in beef cattle studies, the DG diet usually increased dietary CP content compared with CON (i.e., treatment diets were not isonitrogenous). For example, Salim et al. (2012) increased proportion of corn distillers grains with solubles from 0 to 50% (DM basis),

which increased dietary CP from 13.3 to 20.6% (DM basis). Because dietary CP concentration affects urine outputs (Broderick, 2003), the elevation of dietary CP with inclusion of DDGS was probably the factor that increased urine output, not DG per se, in beef studies. In the current study, dietary CP concentration was not different between CON and DG, but the DG diet had lower RDP and greater RUP concentrations compared with CON, which decreased plasma urea N and MUN concentrations (Morris et al., 2018b). Because MUN is one of the factors that alters urine excretion (Nennich et al., 2006), the lower RDP for the DG diet versus CON likely reduced urine outputs. As a result, daily urinary N excretion was lower for DG versus CON, and

Figure 1. Ammonia emission rate (A; g/cow per hour; diet, $P < 0.01$; diet $\times$ hour, $P < 0.01$) and cumulative ammonia emission (B; g/cow; diet, $P < 0.01$) from manure of cows fed the control diet (CON) or the diet containing 28.8% corn distillers grain (DG). Error bars indicate SE.

Figure 2. Hydrogen sulfide emission rate (A; mg/cow per hour; diet, $P = 0.03$; diet $\times$ hour, $P = 0.46$) and cumulative hydrogen sulfide emission (B; mg/cow; diet, $P < 0.01$) from manure of cows fed the control diet (CON) or the diet containing 28.8% corn distillers grain (DG). Error bars indicate SE.

this could be a factor that potentially lowers NH_3 emissions from manure (see the discussion later) because manure N that originated from urine is the main chemical form of N (i.e., urea) contributing to NH_3 emitted from manure (Lee et al., 2011).

Manure N concentration was lower for DG versus CON, which occurred because urinary N concentration to manure was lower for DG versus CON. A potential shift of dietary N from urine to feces was reported when beef cattle were fed a DG diet compared with CON (Cole et al., 2008). In agreement, we observed about 5% units of N shift from urine to feces. This occurred due to decreased urinary N excretion for DG versus CON with no difference in fecal N excretion. Previously, Hristov et al. (2011) speculated that inclusion of DDGS in a ration can cause the shift of N and this shift can decrease NH_3 emission from manure.

Inclusion of DDGS generally increases dietary S concentration due to high S content of DDGS. Therefore, as expected, the DG diet considerably increased S excretion in feces and urine. When dietary S concentration was normal (about 0.2% in dietary DM, i.e., the CON diet in the current study), the rate of S excretion was similar between feces and urine, which is in agreement with a previous beef study (Li et al., 2014). This beef study also observed that an increase in dietary S concentration by increasing the proportion of DG in a diet increased S excretion via urine and an increase in S excretion via feces was minimal. We observed similar results about S excretion between feces and urine for the DG diet compared with CON. This resulted in significant contribution of urinary S to manure S. Although the major chemical form of S in urine and feces was not measured in the current study, it is expected to be sulfate (Hou et al., 2003) and organic S (microbes and feed residual) for urinary and fecal S,

respectively. If this was the case, the increase in urinary S contribution to manure S could increase H_2S emission from manure via microbial reduction of sulfate to H_2S (Yokoyama et al., 2016).

Fecal pH also tended to be lower for DG versus CON, but the difference was relatively small. However, the large reduction in urine pH was not expected in the current study (see more discussion later), and no information about the effects of feeding a DG diet on urine pH in dairy cows is available to our knowledge. Actually, a large decrease in fecal pH was initially speculated, resulting in low manure pH, when ruminants are fed a high-DG diet (Hristov et al., 2011). This speculation was made because of H_2SO_4 being added during DDGS production to control pH and cleanup, increasing acidity of DDGS. Therefore, high inclusion of DDGS in a ration can lower rumen pH (Castillo-Lopez et al., 2014; Ramirez-Ramirez et al., 2016), which could lower fecal pH. However, a tendency of the small reduction in fecal pH for DG in the current study may indicate that a change in fecal pH by the DG diet is minimal in spite of reduced rumen pH. The decrease in urine pH for DG versus CON was significant, and this was another factor that lowers NH_3 emissions from DG manure.

The ammonia emission rate (g/cow per h) and cumulative NH_3 emission (g/d) was about 40% lower for DG versus CON. The degree of the decrease in NH_3 emissions by DG in this study is similar to the decrease observed when feeding a low protein diet, which is probably one of the best nutritional strategies to lower NH_3 emission from manure in dairy cows (Hristov et al., 2011). Previously, a reduction of dietary CP from 16 to 14% of dietary DM decreased NH_3 emission from manure by about 40% (Lee et al., 2012a,b). Therefore, feeding the DG diet (about 30% DDGS in dietary DM) was as effective in lowering ammonia emission from manure as feeding a lower protein diet. The lower NH_3 emission from manure for DG can be explained by 2 factors. As discussed earlier, feeding the DG diet altered the route of N excretion from urine to feces compared with CON, which occurred when the CON and DG diets were iso-nitrogenous. Therefore, the contribution of urine N to manure N changed from 60.5 to 55.5%, which resulted in decreases in the contribution of urinary N to manure N by 19% and decreases in total manure N by 12% for DG compared with CON. In other words, N excretion was about 58.4 g/d less for DG versus CON manure and 96% of the lower N excretion (55.8 g/d) was due to decreased urinary N. In addition, proportion of urea-N in total urinary N was about 65 to 70% (data shown in Morris et al., 2018b), meaning that urea N in DG manure was about 36 g/d less than in CON manure. Because urea is the form of N that is rapidly converted to NH_3 in manure during a short-term manure incuba-

Table 4. Effects of feeding a diet containing high corn distillers grain on manure characteristics on d 10 (n = 9 per treatment)

Item	Diet ¹		SEM	P-value
	CON	DG		
pH	7.47	7.24	0.096	0.02
TS, ² g/kg	135.7	135.0	4.72	0.92
VS, ³ g/kg	843.0	855.1	2.57	<0.01
N, g/kg as is	3.99	4.27	0.096	0.06
N, g/kg of DM	29.6	31.8	0.71	<0.01
S, g/kg as is	0.62	1.29	0.051	<0.01
S, g/kg of DM	4.59	9.51	0.197	<0.01

¹CON = manure from cows fed a basal diet containing soybean meal; DG = manure from cows fed the control diet with 28.8% corn distillers grains with solubles (DM basis) replacing mainly soybean meal and soyhulls.

²Dry matter of manure.

³Volatile solids (OM of manure DM).

tion (Lee et al., 2011), the shift of dietary N excretion from urine to feces for DG versus CON likely explains the 40% decrease in NH_3 emission (i.e., DG manure vs. CON had 15 g less NH_3 -N volatilized over 10 d).

Another factor that decreased NH_3 emission from manure for DG was lower urine pH compared with CON. This may have caused manure pH to be lower for DG versus CON. Accidentally, we did not measure manure pH on d 0 of the incubation, but the lower manure pH likely occurred according to lower manure pH at d 10 and greater manure N concentration for DG versus CON (see more discussion later). Although the contribution cannot be quantified in the current study, it is possible that the reduced urine pH for DG decreased NH_3 emissions from manure. Acidification of manure is an effective management strategy to lower gas emissions, especially NH_3 emission (Sommer et al., 2017). However, acidification of manure in commercial farms is limited because highly concentrated acid (e.g., H_2SO_4) needs to be handled by a specialized worker and is dangerous, increasing the cost for manure management. Therefore, reducing manure pH by feeding a diet containing DDGS can be a potential strategy to lower NH_3 emissions from manure that can be immediately applied in practice. Why, then, did the DG diet decrease urine pH? The decrease in urine pH occurred because of high S concentration in the DG diet due to high inclusion of DDGS. Sulfur concentration in DDGS is high and varies from 0.7 to 1.2% (DM basis; DuFour, 2017). Therefore, inclusion of DG in a ration resulted in an increase in dietary S greater than the typical S level (0.2% in dietary DM). Because of the high S concentration, DCAD ($\text{Na} + \text{K} - \text{Cl} - \text{S}$) in the DG diet decreased from 192 to 65 mEq/kg of DM in the current study (data shown in Morris et al., 2018a). The urine pH of 7.48 with the reduced DCAD (65 mEq/kg of DM) of the DG diet is in agreement with a meta-analysis by Santos et al. (2019), where changes in urine pH by various DCAD in dry cows are shown. Feeding a negative DCAD diet reduces urine pH (acid-base homeostasis; Santos et al., 2019), and this is a well-established strategy for prepartum cows to improve health (e.g., hypocalcemia) and production of cows after parturition (Lean et al., 2019; Santos et al., 2019). After parturition, however, DCAD should be positive, and low DCAD can negatively affect DMI, milk yield and composition, and feed digestibility (Iwaniuk and Erdman, 2015). In our study, we observed that the DG diet decreased DMI, ECM, and milk fat and protein yields compared with CON (Morris et al., 2018a). Negative DMI and production responses (especially milk fat depression; Ramirez-Ramirez et al., 2016) to feeding a DG diet have been often explained by high PUFA supply. However, we suggest that the

negative DMI and production effects by a DG diet need to be reevaluated where reduced DCAD in addition to high PUFA supply may have been responsible.

The DG diet (about 30% DG on a DM basis) in the current study may not be an ideal nutritional strategy in practice, although it decreased NH_3 emission from manure by 40%. First, the DG diet decreased production of cows as mentioned previously. Therefore, to fix this issue, more studies are needed to examine lower DG inclusion rates in a ration but at an inclusion rate that is sufficient to decrease NH_3 emission from manure. Second, the DG diet increased H_2S emission from manure. In the current study, the H_2S emission rate and accumulated H_2S emission over 10 d of incubation increased by about 67% for the DG diet versus CON. It is likely that feeding the DG diet resulted in increases in H_2S emission from manure because of increased dietary S concentration and S intake of cows, which resulted in increases in S concentration in manure (Li et al., 2014; Morris et al., 2018b). In addition, as discussed earlier, the DG diet reduced urine pH (then probably manure pH), and the form of S in manure can become more volatile (pK_a of $\text{H}_2\text{S} = 7.0$) at relatively low pH (Fangueiro et al., 2015). However, when H_2S emissions from manure were measured after feeding a DG diet in previous studies, results were quite variable in pigs (no dairy and beef cattle data are available, to our knowledge). In agreement with our results, Powers et al. (2009) observed H_2S emissions from manure of pigs fed a 20% DG diet where H_2S emission was greater for the DG diet versus CON, although the difference in dietary S concentration between CON and the DG diet in this study was small (0.19 and 0.20% in dietary DM, respectively). Spiehs et al. (2012) observed no differences in H_2S concentration in air of the simulated manure deep pit when manure of pigs fed a 20% DG diet (DM basis) compared with CON was incubated. In the study by Spiehs et al. (2012), the calculated dietary S concentration was 0.18 and 0.24% for CON and the DG diet in this study. However, this study observed high variation for H_2S measurements (CV of 49.5%), which may have masked treatment effects. In addition, although manure outputs were greater for the DG diet versus CON in the study by Spiehs et al. (2012), this was not considered when H_2S emission from manure was compared between treatments and only H_2S concentration measured in air was reported. Trabue et al. (2016), in contrast to our results, observed a decrease in H_2S emitted from manure (0.87 vs. 1.80 g/d per animal) of pigs fed a DG diet (35% on a DM basis) versus CON. However, manure characteristics (e.g., low manure pH and high S concentration in manure) indicated that DG manure would have potential to have more H_2S production. In the study by Trabue et

al. (2016), this discrepancy was explained by crusting of manure (i.e., crusting of manure reduces H₂S emissions). In the current study, we also observed formation of crust on the surface of all manures, and we found no visual difference in crusting between CON and DG. This is supported by no difference in TS measured after the 10-d incubation. The content of VS was statistically different, but the difference was only 1.4% greater for DG versus CON. However, existence of the crust may have affected H₂S emissions to some degree in the current study because we observed greater variability of H₂S production within DG manures compared with CON manures (see Figure 2). Producing H₂S from manure was a relatively slower process compared with NH₃ emission according to gradual increases in H₂S emission over days of the incubation (the interaction of treatment by time). As described previously, if sulfate was the major source for H₂S emitted from manure, sulfate should be reduced to H₂S by manure microbes, which could be a time-requiring process.

Manure N and S concentrations after the 10-d incubation were greater for DG versus CON. This supports the considerable reduction in NH₃ emissions from DG manure compared with CON. Manure S concentration did not change much over 10 d because S lost from manure via H₂S production was only 1% of total S in manure for both CON and DG. This indicates that H₂S emission from manure occurs in the long term, and a long-term incubation of manure to measure H₂S production from manure is necessary.

CONCLUSIONS

Inclusion of DG in a ration at 28.8% on a DM basis compared with a SBM-based diet (CON) decreased urine output and urinary N excretion without affecting fecal output and fecal N excretion. This resulted in low manure N concentration and reduced urinary N contribution to manure N for the DG diet versus CON. In addition, inclusion of DDGS in a ration reduced DCAD compared with CON due to high S content, which reduced urine pH. Therefore, altering the route of N excretion and manure pH by feeding the DG diet considerably reduced NH₃ emission from manure during the 10-d manure incubation. Because of high S content in DG, the DG diet increased dietary S content compared with the CON diet and increased fecal and urinary S excretion. Therefore, greater S concentration and lower pH for DG manure versus CON manure resulted in greater H₂S emission. More studies are needed to determine the optimum level of DG in a ration that reduces NH₃ emissions from manure without negatively affecting DMI and production. A long-term incubation

of manure will be necessary to understand H₂S emissions during manure storage.

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