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# Gas Emissions from Dairy Cows Fed Typical Diets of Midwest, South, and West Regions of the United States

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Gas emissions were determined for dairy cows fed three diets formulated to represent feed ingredients typical of the Midwest, South, or West regions of the United States. Dairy cows were housed and monitored in 12 environmentally controlled rooms (4 cows diet$^{-1}$). Two experiments were performed, representing two lactation stages (initial days in milk were 115 ± 39 d in Stage 1 and 216 ± 48 d in Stage 2). The results demonstrated that the combination of different dietary ingredients resulted in different gas emissions while maintaining similar dry matter intake (DMI) and milk yield (MY). Diet effect on ammonia ($NH_3$) emissions was more prominent in Stage 1. During Stage 1, cows fed the Midwest diet had the highest daily $NH_3$ emission, corresponding to the highest crude protein (CP) concentration among the three regions. The differences in $NH_3$ emissions (39.0%) were much larger than the percent difference in CP concentrations between diets (6.8%). Differences in N intake, N excretion, or milk urea N alone may not serve as a strong indicator of the potential to reduce $NH_3$ emissions. Lower emissions of methane ($CH_4$) per unit DMI or per unit MY were observed for cows offered the South diet during Stage 1 as compared with that from cows offered the Midwest or West diets. No diet effect was observed for hydrogen sulfide ($H_2S$) emission per unit S intake, nor for nitrous oxide ($N_2O$) emission. The measured $NH_3$ and $CH_4$ emissions were comparable, but the $N_2O$ emissions were much higher than those reported for tie-stall dairy barns in the literature.

Gas emissions from animal feeding operations (AFOs) are receiving increasing attention because of concerns related to human and animal health, nuisance, and contributions to climate change. The gas emissions of interest include ammonia ($NH_3$), hydrogen sulfide ($H_2S$), methane ($CH_4$), nitrous oxide ($N_2O$), carbon dioxide ($CO_2$), volatile organic compounds, and odor. Ammonia gas is emitted from AFOs because of the relatively inefficient conversion of feed nitrogen (N) into animal product (meat, egg, and/or milk). Atmospheric $NH_3$ is an important pollutant due to its impact on ecosystems. Ammonia can react in the atmosphere with other gases to form fine particulates. Deposition of $NH_3$ can lead to overenrichment of nutrients and cause eutrophication of surface water. Hydrogen sulfide is produced by decomposition of animal manure whenever there are sulfur compounds, anaerobic conditions, and sufficient moisture. It is an extremely toxic and irritating gas at high levels, and has a generally objectionable odor of rotten eggs. The $CH_4$, $N_2O$, and $CO_2$ are greenhouse gases (GHG) and contribute to global climate change. The $NH_3$ emissions from AFOs are estimated to account for 71% of total human-induced $NH_3$ emissions in the United States based on estimations in the National Emission Inventory (USEPA, 2004). The Food and Agriculture Organization of the United Nations estimated that the global animal agriculture sector is responsible for 18% of global, human-induced GHG emissions (Steinfeld et al., 2006).

Dietary strategies have the potential to reduce gas emissions from AFOs (Powers et al., 2007; James et al., 1999). Reducing N inputs by reducing dietary crude protein (CP) or by providing an optimal balance between rumen degradable protein and rumen undegradable protein without negatively impacting performance was shown to reduce N excretion in dairy cows (Reynal and Broderick, 2005). Reducing dietary CP content not only reduced total N excretion but also resulted in a greater proportion of the N excretion in urine (Misselbrook et al., 2005). Frank et al. (2002) reported that $NH_3$ emissions from

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**Abbreviations**: AFO, animal feeding operation; AOAC, Association of Analytical Chemists; CP, crude protein; DIM, days in milk; DL, detection limit; DMI, dry matter intake; GHG, greenhouse gas; MUN, milk urea nitrogen; MY, milk yield; RH, relative humidity.

dairy cattle manure were reduced by two-thirds when lowering diet CP from 19 to 14%, with no negative effects on milk yield (MY) or composition. Johnson and Johnson (1995) found that highly digestible diets yielded lower $CH_4$ emissions than poor-quality lower digestible diets; thus, dietary modification may provide a mechanism for reducing $CH_4$ emissions from livestock operations. Increasing the feed efficiency of dairy cows producing milk can reduce $CH_4$ emissions (Boadi et al., 2004). Aguerre et al. (2011) observed that increasing forage-to-concentrate ratios increased $CH_4$ emission, but had little effect on performance, manure excretion, or the emissions of $NH_3$ and $CO_2$.

The objective of this study was to determine whether different feed ingredient combinations resulted in measurable differences in gas emissions from dairy cows when energy and nutrient composition were similar among diets. This study directly measured gas emissions from dairy cows fed diets of typical feed ingredients from the Midwest, South, or West regions of the United States.

## Materials and Methods

### Cows and Experimental Setup

All animal procedures were approved by the Michigan State University Institutional Animal Care and Use Committee. Holstein dairy cows were housed and monitored individually in 12 environmentally controlled rooms (height 2.60 m by width 2.37 m by length 4.11 m) at the Animal Air Quality Research Facility at Michigan State University. Cows were confined in a 107-cm-long by 183-cm-wide raised stall covered with a rubber mat surface. A feeder was placed at the front of the stall and a pan of the same width as the stall was placed at the rear to collect urine and feces. Each room accommodated one cow weighting approximately 600 kg. The system simulated a tie-stall dairy barn. Cows remained in their individual room during the diet adjustment period (2 wk) and measurement days associated with each experiment. Cows were fed and milked twice daily using a vacuum line and portable milking machine. Individual feed intake and MY were recorded daily. Two experiments were performed, representing two lactation stages (cows in Stage 1 were 115 ± 39 initial days in milk [DIM]; cows in Stage 2 were 216 ± 48 initial DIM, and were 123 ± 35 d in pregnancy). In each experiment, cows were randomly selected from a group of 18 cows and randomly assigned to the rooms. Only 50% of the cows were common to both experiments. The duration of the Stage 1 experiment was 20 d and the Stage 2 experiment was 23 d.

Conditions within the environmental rooms were managed to optimize cow health and productivity. Each room was individually heated and cooled using 100% ambient air with all of the air exhausted to the outside (no recycling, room ventilation rates ranged from 900 to 1100 $m^3 h^{-1}$). Ventilation rates of each room were continuously measured using a 15.24-cm-orifice plate in the incoming duct of each room and a differential pressure transducer (Setra Model 239). Orifice plates and pressure transducers specific to each room were calibrated at a University of Illinois laboratory during facility construction; no changes have taken place since construction. Throughout this study, ventilation rates were checked against room-specific mass flow meters that are calibrated annually. Air temperature and relative humidity (RH) in each room were measured using a Campbell Scientific CS500 temperature and RH probe and recorded every 2 s. The average air temperature and RH measured during the experiments were 12.5 ± 1.3°C and 46.5 ± 10.6%, respectively.

### Diet

Diets were formulated to meet protein, energy, and fiber requirements of cows at midlactation (from Day 100 to Day 200 after calving; NRC, 2001) and represent typical commercial diets based on ingredient availability of the respective regions. Stage 1 represented the beginning of midlactation when cows have reached peak MY, are achieving maximum dry matter intake (DMI), and not experiencing the weight losses associated with the early lactation period (the first 100 d of lactation). Stage 2 represented the end of midlactation when MY usually declines and cows start to gain weight to replenish the adipose tissue lost during early lactation, or due to the increased size of the growing fetus. The main target during midlactation period is to maintain peak MY as long as possible. Kalscheur et al. (1999) found that diet CP often is a limiting nutrient in early lactation, but in midlactation cows fed the low-CP diets (CP = 13.3%) can maintain equivalent MY compared with cows fed the higher-CP diets (CP = 15.3%). Recent studies have reported MY of 40 to 50 kg $d^{-1}$ when corn silage–based rations containing 14 to 14.5% CP were fed (Recktenwald and Van Amburgh, 2006; Hofheer et al., 2010).

The three dietary treatments represented feed ingredients commonly fed in three regions of the United States: Midwest, South, and West. Each diet was fed to four cows. Forages, corn, and soybean meal were from a single source while other ingredients were sourced from the region in which the diets represented. Ingredients were mixed and stored daily. Total mixed rations were mixed daily. Feed was weighed for each animal and placed in the feed bunk twice daily. Formulated and analyzed nutrient compositions of the three diets for each of the experiments are listed in Table 1. The diets in Stage 2 had lower acid detergent fiber and neutral detergent fiber but higher soluble protein than those in Stage 1.

### Sampling and Chemical Analyses of Feed, Milk, and Manure

Feed samples were collected twice weekly from each total mixed ration and frozen in sealed bags. The samples were later thawed, composited, and sent to the Dairy One Forage Testing Laboratory (Ithaca, NY) for chemical analyses of CP, available protein, acid detergent insoluble crude protein, soluble protein, acid detergent fiber, neutral detergent fiber, Ca, P, Mg, S, dry matter, and net energy of lactation. The total mixed ration CP content was confirmed at a Michigan State University laboratory using total Kjeldahl N (TKN) Association of Analytical Chemists (AOAC) Official Method 984.13 (AOAC, 2006). Morning and evening milk samples were collected daily in the last 5 d of each experiment and analyzed within a day for fat, protein, and milk urea nitrogen (MUN) with infrared spectroscopy by the Michigan Dairy Herd Improvement Association (East Lansing). Manure was mixed thoroughly every morning and removed partially to maintain an equal depth of 5 cm so as to provide an emissions surface while preventing overflow of the pan. Each time manure was removed a homogenous subsample was collected, frozen, then analyzed separately by day at the end of the study. Manure TKN and $NH_3$–N content was measured in a Michigan State University laboratory using AOAC Official Method 984.13 (AOAC, 2006) and AOAC Official Method 928.08 for distillation (AOAC, 2000), respectively.

## Gas Emission Measurements

Using software control system LabVIEW Version 8.2 (National Instruments Corporation), gas concentrations were measured in a sequential manner from Room 1 to 12. Measurement of incoming air was followed by measurements of each of the 12 rooms' exhaust air for 15 min continuously throughout the 20-d experiment in Stage 1 and the 23-d experiment in Stage 2. Each measurement cycle through all 12 rooms plus the background air requires 195 min to complete (13 × 15 min $room^{-1}$). Therefore, there were seven or eight daily observations per room as described by Liu et al. (2011). The incoming air line and rooms' exhaust

**Table 1. Feed components and analyzed nutrient composition of diets† fed to dairy cows.**

| Category | Feed components or composition | Stage 1 (initial DIM‡ = 115 ± 39 d) | | | Stage 2 (initial DIM = 216 ± 48 d) | | |
|---|---|---|---|---|---|---|---|
| | | Midwest | South | West | Midwest | South | West |
| Ingredients, %, dry basis | Corn silage | 46.7 | 63.0 | 62.6 | 44.6 | 62.6 | 65.9 |
| | Alfalfa haylage | 24.6 | – | – | 27.9 | – | – |
| | Concentrate mix | 22.1 | 21.3 | 22.4 | 21.7 | 20.9 | 21.7 |
| | Alfalfa hay | 2.8 | 9.2 | 9.7 | 2.9 | 10.6 | 8.3 |
| | Whole cottonseed | 3.7 | 6.5 | 5.3 | 2.9 | 5.8 | 4.1 |
| Ingredients of concentrate mix, %, dry basis | Corn—yellow ground | 45.85 | – | 16.35 | 40.45 | – | 32.5 |
| | Beet pulp—dried | – | – | 17.08 | – | – | 9.33 |
| | DDGS§ | 21.9 | 22.45 | 21.25 | 24.48 | 27.9 | 7.77 |
| | Calcium carbonate | 2.28 | 2.90 | 2.48 | 2.35 | 2.83 | 2.48 |
| | Soybean meal | 18.07 | 16.51 | 21.62 | 15.05 | 11.21 | 1.49 |
| | Sodium bicarbonate | 0.80 | 0.90 | 0.85 | 0.89 | 1.02 | 0.93 |
| | Salt (NaCl) | 0.83 | 0.92 | 0.89 | 0.84 | 1.02 | 0.92 |
| | Magnesium oxide | 0.21 | 0.21 | 0.08 | 0.36 | 0.27 | 0.15 |
| | Dairy V PMX | 0.05 | 0.05 | 0.05 | 0.06 | 0.07 | 0.07 |
| | Selenium 1600 | 0.04 | 0.05 | 0.04 | 0.04 | 0.04 | 0.04 |
| | Soybean hulls | 9.81 | 17.2 | – | 15.25 | 7.50 | – |
| | Dical 21% Phosphate | – | – | 0.15 | 0.16 | 0.05 | – |
| | Hominy | – | 38.65 | – | – | 48.00 | – |
| | Dairy trace mineral | 0.10 | 0.12 | 0.11 | 0.07 | 0.08 | 0.07 |
| | Almond hulls | – | – | 18.95 | – | – | – |
| | Vitamin E-50 | 0.02 | 0.02 | 0.02 | – | – | – |
| | Ferrous sulfate | 0.03 | 0.02 | 0.06 | – | – | – |
| | Sunflower meal | – | – | – | – | – | 18.66 |
| | Amino plus | – | – | – | – | – | 14.90 |
| | Wheat middlings | – | – | – | – | – | 7.50 |
| | Choice white grease | – | – | – | – | – | 3.18 |
| | Fat | 5.06 | 5.96 | 5.91 | 5.06 | 5.96 | 5.91 |
| Analyzed composition, %, dry basis | CP¶ | 16.2 | 15.5 | 15.1 | 16.5 | 15.3 | 15.3 |
| | Available protein | 15.1 | 14.4 | 14.1 | 15.4 | 14.4 | 14.6 |
| | ADICP# | 1.1 | 1.0 | 1.0 | 1.1 | 0.8 | 0.7 |
| | Soluble protein | 5.7 | 4.5 | 4.3 | 6.8 | 6.2 | 5.8 |
| | Acid detergent fiber | 28.9 | 28.8 | 27.1 | 25.9 | 25.9 | 25.6 |
| | Neutral detergent fiber | 41.7 | 44.5 | 41.0 | 37.9 | 40.4 | 38.5 |
| | Calcium | 0.81 | 0.70 | 0.80 | 0.89 | 0.67 | 0.71 |
| | Phosphorus | 0.42 | 0.41 | 0.40 | 0.42 | 0.45 | 0.42 |
| | Magnesium | 0.33 | 0.32 | 0.31 | 0.36 | 0.37 | 0.37 |
| | Sulfur | 0.28 | 0.32 | 0.24 | 0.23 | 0.22 | 0.20 |
| | DM††, % | 42.3 | 49.0 | 48.0 | 49.9 | 55.7 | 53.4 |
| | NEL‡‡, MJ $kg^{-1}$ | 6.6 | 6.6 | 6.7 | 6.8 | 6.7 | 6.8 |

† Diets that represented feed ingredients commonly fed in the midwestern (Midwest), southern (South), or western (West) United States.

‡ DIM, days in milk.

§ Dried distillers grain with solubles.

¶ Crude protein.

# Acid detergent insoluble crude protein.

†† Dry matter.

‡‡ Net energy lactation.

sampling lines were purged for 9.5 min before the start of each room sampling. Following purging, data were collected for 5.5 min. All gases were measured simultaneously within a sample air stream. The air sample was pulled to a sampling manifold using a Cole-Parmer vacuum pump at a rate of 30 L $min^{-1}$ and then diverted into three gas analyzers: a chemiluminescence analyzer (Thermo Fisher TEI Model 17C; detection limit [DL] = 0.001 $\mu mol\ mol^{-1}$) that determined $NH_3$, NO, and $NO_2$ concentrations; a pulsed fluorescence $SO_2$–$H_2S$ analyzer (TEI Model 450i; DL = 0.003 $\mu mol\ mol^{-1}$; error = 1% of full scale at 1 $\mu mol\ mol^{-1}$); and a Lumasense Technologies INNOVA 1412 photoacoustic analyzer that measured concentrations of $CO_2$ (DL = 5.1 $\mu mol\ mol^{-1}$), $CH_4$ (DL = 0.1 $\mu mol\ mol^{-1}$), $NH_3$ (DL = 0.2 $\mu mol\ mol^{-1}$), and $N_2O$ (DL = 0.03 $\mu mol\ mol^{-1}$). Weekly zero and span calibration were performed on chemiluminescence and pulsed fluorescence analyzers. The INNOVA analyzer was calibrated at the beginning and end of each experiment, and weekly span checks were performed. Zero and span calibrations of <1 $\mu mol\ mol^{-1}$ drift were maintained throughout each experiment. For statistical analyses, the observations that were below the DL of the analyzers were recorded as half of the DL as a common practice (Woodside and Kocurek, 1997). Gas emission rates were calculated as the product of ventilation rates and concentration differences between exhaust and incoming air using the following equation:

$$ER = Q \times \frac{273}{T} \times \left(C_o - C_i\right) \times 10^{-6} \times \frac{MW}{V_m} \qquad [1]$$

where ER is emission rate, g $min^{-1}$; $Q$ is ventilation rate at room temperature and pressure, L $min^{-1}$; $T$ is air temperature in room exhaust, °K; $C_o$ is gas concentration in room exhaust, $\mu mol\ mol^{-1}$; $C_i$ is gas concentration in the incoming air, $\mu mol\ mol^{-1}$; MW is molecular weight of the gas, g $mol^{-1}$; and $V_m$ is molar volume of gas at standard conditions (22.414 L $mol^{-1}$). Daily emission rates (g $d^{-1}$) of each room were calculated as sum of the emitted mass during the seven or eight measurement cycles of the day.

### Data Analyses

Data were analyzed statistically by ANOVA using the MIXED model procedure of SAS (SAS for Windows, Version 9.1.3, SAS Institute, Cary, NC). Emission data were expressed as daily emission rates (g $d^{-1}$ $cow^{-1}$), emission per MY, emission per DMI, and emission per N or S intake. The model tested the fixed effect of diet. Date was a random variable, and cow was treated as a nested term within diet. Effect of stage was evaluated by pooling together the data from the two stages. Tukey's test (Games and Howell 1976) was used to compare effects of diet or stage. Probability of statistical significance was $P \leq 0.05$.

## Results

### Effects of Diet and Lactation Stage on Feed Intake, Milk Yield, and Excretion

Effects of diet on DMI, MY, and manure excretion at the two lactation stages are presented in Table 2. The daily DMI and N intake were not significantly different among the three diets in either stage. Regardless of stage, the daily S intake was lowest for the West diet, corresponding to the lowest dietary S concentration. Average N and average S intakes were 3% higher and 21% lower during Stage 2 compared with that in Stage 1, corresponding to higher CP and lower S concentrations of the diets in Stage 2.

In both Stages 1 and 2, no diet effects were observed for daily MY, MY per DMI, MY per N intake, N in daily MY, or milk N efficiency, expressed as N from milk protein as a percentage of total N intake. Diet effects on milk protein concentrations and MUN were observed in Stage 2, and cows fed the Midwest diet had the highest milk protein concentrations and MUN, corresponding to the highest CP concentration in the Midwest diet as compared with other diets. Both milk protein and milk fat concentrations increased in Stage 2 as compared with that in Stage 1, not uncommon for cows in later lactation. Also consistent with cows in later stages of lactation, the daily MY, MY per DMI, MY per N intake, and N in daily MY all decreased in Stage 2.

No diet effects were observed for daily manure excretion (dry matter) or N excretion rate. A diet effect on N concentration in manure was observed in Stage 1, and cows offered the South diet had lowest N concentration in manure. Weight of daily excretion, manure N concentration, N in excretion, and manure weight per MY all decreased in Stage 2 as compared with that in Stage 1.

### Effects of Diet and Lactation Stage on Gas Emissions

Effects of diet on gas emissions during the two lactation stages are presented in Table 3. Both the TEI Model 17C and the INNOVA instruments measured $NH_3$ concentrations, and the results were in agreement with each other. For simplicity, the $NH_3$ measurements from the TEI Model 17C analyzer were used for the analyses.

A diet effect on $NH_3$ emission was observed in Stage 1 but not in Stage 2. During Stage 1, cows fed the Midwest diet had the highest daily $NH_3$ emission rate, corresponding to the highest CP and soluble protein concentrations in the Midwest diet as compared with other diets. The $NH_3$ emission per unit N intake was not influenced by diet. Diet effects on daily emissions of $N_2O$ and $CH_4$ were not significant in Stages 1 or 2. However, the emissions of $CH_4$ relative to DMI were significantly lower when cows were offered the South diet as compared with that when cows were offered the other two diets in both Stages 1 and 2 ($P \leq 0.05$, Table 3). During Stage 1, the daily emission of $H_2S$ was lowest when the West diet was offered, corresponding to the lowest S intake. The $H_2S$ emission per unit S intake was not influenced by diet in both Stages 1 and 2.

The average daily $NH_3$ emission in Stage 2 was not significantly different from that in Stage 1, while the average daily emissions of $CH_4$, $CO_2$, $N_2O$, and $H_2S$ increased by 12.5, 16.9, 13.3, and 76.2%, respectively, in Stage 2 as compared with that in Stage 1. The average $NH_3$ emission relative to MY was not significantly different during Stage 2 compared to Stage 1, while the average emissions of $CH_4$, $CO_2$, $N_2O$, and $H_2S$ per unit MY increased by 22.5, 47.9, 22.8, and 91.7%, respectively, in Stage 2 as compared with that in Stage 1. The differences in $CH_4$ and $CO_2$ emissions between stages could be related to increased respiratory activity in Stage 2. The differences in $N_2O$ and $H_2S$ emissions are difficult to explain, and they could be related to different climate conditions or measurement error considering both measured $N_2O$ and $H_2S$ concentrations were very low and associated with high uncertainties.

# Discussion

## Nitrogen Emissions

Summary of measured emission rates of $NH_3$, $N_2O$, and $CH_4$ from dairy cow operations in literature are presented in Table 4. Monteny and Erisman (1998) reported that $NH_3$ emission rates were lower from tie-stalls than that from cubicle (loose) dairy cow houses, mainly because of a reduction of surface area of the pit and the urine-and-feces-fouled floor in tie-stalls, and the ranges in reported $NH_3$ emission from tie-stalls appear to be large due to systematic and natural variation in floor type, manure handling, and indoor manure storage facilities. In the present study, the emission rates across all diets averaged 30.0, 12.8, 0.14, and 0.03 g d$^{-1}$ cow$^{-1}$ for $NH_3$, $N_2O$, $NO_2$, and NO, respectively. The $NH_3$ emission rates measured in the present study were on the high end of the ranges that have been reported for tie-stalls. The measured $N_2O$ emission rates were more than 20 times higher than the limited data for tie-stall barns in the literature (Table 4). As seen in Table 4, large variations have been observed for measured $N_2O$ emission rates. High $N_2O$ emission rates have been reported for free-stall barns (Zhang et al., 2005) and open lots (Leytem et al., 2011; Denmead et al., 2008). Although high uncertainties have been observed for $N_2O$ measurement using the INNOVA analyzer, the analyzer functioned properly in the present study and in other studies conducted in the same time period. There was no reason to invalidate the $N_2O$ data. The high $N_2O$ emission rates observed in the present study were difficult to explain, and they could be results of the specific management practices used.

Monteny et al. (1998) related $NH_3$ emissions to MUN concentration as well as diet CP concentration. The present study showed that diet effects of different feedstuffs on MUN concentration and $NH_3$ emissions were significantly influenced by stage of lactation (Fig. 1). During Stage 1, the CP concentration in the West diet was 6.8% lower than that in the Midwest diet (15.1 vs. 16.2% CP), and the $NH_3$ emission rates were 39.0% lower (from 36.2 to 22.1 g d$^{-1}$ cow$^{-1}$) when cows were offered the West diet as compared to that when cows were offered the Midwest diet, although no diet effect was observed for MUN concentrations. During Stage 2, the CP concentration in the West diet was 7.3% lower than that in the Midwest diet (15.3 vs. 16.5% CP), but no significant differences in the $NH_3$ emission rates were observed between diets, although 15% lower MUN concentration was observed when cows were offered the West diet as compared with the Midwest diet.

**Table 2. Effects of diet† on feed intake, milk yield, and excretion at the two lactation stages in dairy cows.**

| Cow condition and performance | Stage 1 (initial DIM‡ = 115 ± 39 d) | | | | | Stage 2 (initial DIM = 216 ± 48 d) | | | | | *P* value for stage effect |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Least squares means | | | SEM | *P* value | Least squares means | | | SEM | *P* value | |
| | Midwest | South | West | | | Midwest | South | West | | | |
| Feed intake | | | | | | | | | | | |
| DMI§, kg d$^{-1}$ cow$^{-1}$ | 20.8 | 21.8 | 20.5 | 1.0 | 0.61 | 22.2 | 21.1 | 20.2 | 1.1 | 0.45 | 0.69 |
| N intake, g d$^{-1}$ cow$^{-1}$ | 539 | 541 | 496 | 25 | 0.36 | 591 | 518 | 520 | 30 | 0.19 | 0.05 |
| S intake, g d$^{-1}$ cow$^{-1}$ | 58.3¶ | 69.9$^{b}$ | 49.3$^{a}$ | 2.8 | <0.01 | 52.0$^{b}$ | 45.9$^{ab}$ | 41.0$^{a}$ | 2.6 | 0.04 | <0.01 |
| Milk yield | | | | | | | | | | | |
| Milk protein concentration, % | 3.06 | 3.09 | 3.06 | 0.03 | 0.96 | 3.34$^{b}$ | 3.16$^{a}$ | 3.33$^{b}$ | 0.05 | <0.01 | <0.01 |
| Milk fat concentration, % | 4.18 | 4.26 | 3.94 | 0.16 | 0.37 | 4.33 | 4.27 | 4.46 | 0.13 | 0.48 | 0.29 |
| MUN#, mg dL$^{-1}$ | 13.0 | 12.6 | 12.0 | 0.6 | 0.53 | 10.9$^{b}$ | 10.4$^{b}$ | 9.3$^{a}$ | 0.2 | <0.01 | <0.01 |
| Daily MY, kg d$^{-1}$ cow$^{-1}$ | 37.0 | 37.5 | 33.2 | 1.7 | 0.19 | 33.3 | 32.2 | 30.4 | 2.6 | 0.74 | <0.01 |
| MY†† per DMI, kg kg$^{-1}$ | 1.79 | 1.73 | 1.63 | 0.08 | 0.39 | 1.49 | 1.53 | 1.52 | 0.07 | 0.92 | <0.01 |
| MY per N intake, kg kg$^{-1}$ | 69.2 | 70.0 | 67.4 | 3.2 | 0.85 | 56.2 | 62.2 | 59.3 | 2.7 | 0.32 | <0.01 |
| N in daily MY, g d$^{-1}$ cow$^{-1}$ | 180 | 185 | 164 | 8 | 0.19 | 176 | 158 | 162 | 11 | 0.50 | <0.01 |
| Milk N efficiency‡‡, % | 33.6 | 34.4 | 33.2 | 1.0 | 0.67 | 29.9 | 30.6 | 31.4 | 0.6 | 0.21 | <0.01 |
| Excretion (feces and urine) | | | | | | | | | | | |
| Dry matter daily excretion§§, kg d$^{-1}$ cow$^{-1}$ | 10.3 | 10.5 | 9.3 | 0.7 | 0.46 | 9.3 | 8.4 | 8.4 | 0.5 | 0.38 | <0.01 |
| Manure N concentration, % dry basis | 2.38$^{b}$ | 2.16$^{a}$ | 2.32$^{b}$ | 0.04 | <0.01 | 2.28 | 2.01 | 2.22 | 0.08 | 0.08 | <0.01 |
| N in excretion, g d$^{-1}$ cow$^{-1}$ | 244 | 228 | 216 | 17 | 0.51 | 212 | 168 | 188 | 13 | 0.10 | <0.01 |
| Manure per MY, g kg$^{-1}$ | 6.7 | 6.1 | 6.5 | 0.5 | 0.66 | 6.5 | 5.3 | 6.2 | 0.4 | 0.16 | <0.01 |
| N excretion rate¶¶, % | 45.5 | 42.2 | 43.9 | 2.0 | 0.44 | 36.2 | 32.7 | 36.4 | 0.01 | 0.11 | <0.01 |

† Diets that represented feed ingredients commonly fed in the midwestern (Midwest), southern (South), or western (West) United States.

‡ Days in milk.

§ Dry matter intake.

¶ Values within a row in the same stage differ significantly if without common letter ($P \le 0.05$).

\# Milk urea N.

†† Milk yield.

‡‡ Milk N efficiency was expressed as N from milk protein in percent of total N intake.

§§ Daily excretion was estimated by dividing the total manure (feces and urine) in the entire experimental period by number of days in the experiment.

¶¶ The N excretion rate was expressed as N in excretion in percent of total N intake.

Powell et al. (2011) reported that when MUN declined from 14 to 10 mg dL$^{-1}$, $NH_3$ emissions from dairy barns were reduced by 10.3 to 28.2%. van Duinkerken et al. (2011) observed that $NH_3$ emission increased by 2.5 and 3.5%, respectively, when MUN increased by 1 mg per 100 g milk. Burgos et al. (2010) reported that there was a strong relationship between $NH_3$ emissions and MUN but also admitted that stage of lactation is significant when using MUN to predict total $NH_3$ emissions. In the present study, low MUN also coincided with low $NH_3$ emissions, but there were more uncertainties associated with the actual reductions of $NH_3$ emissions that occur in different stages. Part of the uncertainties could be attributed to the narrow differences in diet CP and MUN. Results indicate that the small differences in N intake, N excretion, or MUN alone may not serve as a strong indicator of the potential to reduce $NH_3$ emissions. The stage of lactation could be an important factor that influences the diet effects on $NH_3$ emissions. The diet effect on $NH_3$ emission was more prominent in Stage 1 when DIM of cows were lower as compared with that in Stage 2.

## Nitrogen Mass Balance

During Stage 1, the daily N input from feed across diets averaged 527 g N d$^{-1}$ cow$^{-1}$. The measured total daily N output averaged 440 g N d$^{-1}$ cow$^{-1}$ (83.6% of N input), including 177 g N d$^{-1}$ cow$^{-1}$ in MY, 230 g N d$^{-1}$ cow$^{-1}$ remaining in manure, 25.4 g N d$^{-1}$ cow$^{-1}$ in $NH_3$ emission, 7.6 g N d$^{-1}$ cow$^{-1}$ in $N_2O$ emission, 0.04 g N d$^{-1}$ cow$^{-1}$ in $NO_2$ emission, and 0.01 g N d$^{-1}$ cow$^{-1}$ in NO emission. During Stage 2, the daily N input from feed across diets averaged 544 g N d$^{-1}$ cow$^{-1}$. The measured total daily N output averaged 388 g N d$^{-1}$ cow$^{-1}$ (71.3% of N input), including 166 g N d$^{-1}$ cow$^{-1}$ in MY, 189 g N d$^{-1}$ cow$^{-1}$ remaining in manure, 24.2 g N d$^{-1}$ cow$^{-1}$ in $NH_3$ emission, 8.7 g N d$^{-1}$ cow$^{-1}$ in $N_2O$ emission, 0.04 g N d$^{-1}$ cow$^{-1}$ in $NO_2$ emission, and 0.02 g N d$^{-1}$ cow$^{-1}$ in NO emission.

Comparing the N input and the total N output resulted in 16.4% unaccounted N loss during Stage 1 and 28.7% unaccounted N loss during Stage 2, which could be due in part to the change in body N, fetal growth, and other forms of N loss (sweat, hair, skin, etc.). Uncertainties and error during collection, storage, and analysis of manure and feed N contributed to the unaccounted N. During Stage 2, N intake increased, while N in MY, N in manure excretion, and N in gas emissions all decreased, thus resulting in a larger unaccounted N loss in the advanced lactation stage when fetal growth was considerable. Based on the model estimation provided in NRC (2001), the metabolizable protein required for pregnancy is not negligible after 100 d of pregnancy, and it can reach 385 g d$^{-1}$ at 285 d of pregnancy, which indicates consumption of N for pregnancy could be as high as 62 g N d$^{-1}$ (around 11% of the total N input in this study).

**Table 3. Effects of diet† on gas emissions at the two lactation stages in dairy cows.**

| Gas emissions | Stage 1 (initial DIM‡ = 115 ± 39 d) | | | | | Stage 2 (initial DIM = 216 ± 48 d) | | | | | *P* value for stage effect |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Least squares means | | | SEM | *P* value | Least squares means | | | SEM | *P* value | |
| | Midwest | South | West | | | Midwest | South | West | | | |
| $NH_3$ | | | | | | | | | | | |
| Daily emissions, g d$^{-1}$ cow$^{-1}$ | 36.2$^{b}$§ | 32.1$^{ab}$ | 22.1$^{a}$ | 3.9 | 0.05 | 31.1 | 31.3 | 24.2 | 4.8 | 0.44 | 0.20 |
| Emissions per MY¶, g kg$^{-1}$ | 1.01 | 0.85 | 0.67 | 0.12 | 0.16 | 0.94 | 1.02 | 0.80 | 0.17 | 0.60 | 0.81 |
| Emissions per DMI#, g kg$^{-1}$ | 1.81 | 1.50 | 1.09 | 0.24 | 0.13 | 1.42 | 1.51 | 1.20 | 0.23 | 0.58 | 0.13 |
| Emissions per N intake, g kg$^{-1}$ | 69.8 | 60.6 | 45.2 | 9.7 | 0.21 | 53.5 | 61.5 | 48.1 | 9.2 | 0.53 | 0.13 |
| $N_2O$ | | | | | | | | | | | |
| Daily emissions, g d$^{-1}$ cow$^{-1}$ | 12.3 | 12.1 | 11.4 | 0.8 | 0.34 | 14.1 | 13.7 | 13.1 | 0.8 | 0.64 | 0.03 |
| Emissions per MY, g kg$^{-1}$ | 0.34 | 0.32 | 0.35 | 0.02 | 0.53 | 0.43 | 0.43 | 0.43 | 0.02 | 0.99 | < 0.01 |
| Emissions per DMI, g kg$^{-1}$ | 0.59 | 0.55 | 0.56 | 0.04 | 0.13 | 0.64 | 0.65 | 0.65 | 0.02 | 0.76 | 0.04 |
| Emissions per N intake, g kg$^{-1}$ | 22.9 | 22.4 | 23.2 | 1.4 | 0.55 | 24.1$^{a}$ | 26.6$^{b}$ | 25.7$^{ab}$ | 1.0 | 0.05 | 0.09 |
| $CH_4$ | | | | | | | | | | | |
| Daily emissions, g d$^{-1}$ cow$^{-1}$ | 405 | 354 | 386 | 17 | 0.08 | 478 | 413 | 404 | 22 | 0.07 | < 0.01 |
| Emissions per MY, g kg$^{-1}$ | 11.1$^{b}$ | 9.5$^{a}$ | 11.6$^{b}$ | 13.9 | 0.04 | 14.7 | 13.1 | 13.3 | 0.7 | 0.28 | < 0.01 |
| Emissions per DMI, g kg$^{-1}$ | 19.7$^{b}$ | 16.2$^{a}$ | 18.8$^{b}$ | 0.8 | 0.01 | 21.7$^{b}$ | 19.7$^{a}$ | 20.1$^{a}$ | 0.5 | < 0.01 | < 0.01 |
| $CO_2$ | | | | | | | | | | | |
| Daily emissions, g d$^{-1}$cow$^{-1}$ | 13816 | 13434 | 12827 | 785 | 0.26 | 16352 | 15729 | 15032 | 796 | 0.48 | < 0.01 |
| Emissions per MY, g kg$^{-1}$ | 378 | 259 | 387 | 25 | 0.45 | 501 | 497 | 497 | 25 | 0.99 | < 0.01 |
| Emissions per DMI, g kg$^{-1}$ | 669 | 619 | 629 | 36 | 0.08 | 742 | 751 | 749 | 23 | 0.92 | < 0.01 |
| $H_2S$ | | | | | | | | | | | |
| Daily emissions, g d$^{-1}$ cow$^{-1}$ | 0.044$^{b}$ | 0.045$^{b}$ | 0.027$^{a}$ | 0.004 | 0.02 | 0.085 | 0.084 | 0.054 | 0.012 | 0.08 | < 0.01 |
| Emissions per MY, g kg$^{-1}$ | 0.0012$^{b}$ | 0.0012$^{b}$ | 0.0009$^{a}$ | 0.0001 | 0.05 | 0.0026 | 0.0026 | 0.0018 | 0.0003 | 0.15 | < 0.01 |
| Emissions per DMI, g kg$^{-1}$ | 0.0021$^{b}$ | 0.0021$^{b}$ | 0.0013$^{a}$ | 0.0002 | 0.01 | 0.0039 | 0.0040 | 0.0027 | 0.0006 | 0.08 | < 0.01 |
| Emissions per S intake, g kg$^{-1}$ | 0.75 | 0.65 | 0.55 | 0.07 | 0.12 | 1.70 | 1.82 | 1.39 | 0.24 | 0.26 | < 0.01 |

† Diets that represented feed ingredients commonly fed in the midwestern (Midwest), southern (South), or western (West) United States.

‡ Days in milk.

§ Values within a row in the same stage differ significantly if without common letter ($P \leq 0.05$).

¶ Milk yield.

# Dry matter intake.

Comparison of N balance in this study (Stages 1 and 2) and in other studies is shown in Fig. 2. Due to the relatively low CP of the diets (15.1–16.5%), the milk N efficiency in the present study (29.9–34.4%) was relatively high compared with the 15 to 25% milk N efficiency estimated by Aarst et al. (1992). Frank and Swensson (2002) have reported milk N efficiency as high as 42% with low-protein diets of 13.1 to 13.5% CP, and feeding to standard recommendations of 19% diet CP achieved milk N efficiency of 25 to 32%. Typical N excretion (urine and feces combined, before any changes due to dilution water addition, drying, volatilization, or other physical, chemical, or biological processes) from lactating dairy cows has been estimated to be 491 g N $d^{-1}$ $cow^{-1}$ when daily N intake is 700 g N $d^{-1}$ $cow^{-1}$ (Nennich et al., 2005), a 70% N excretion rate. van Dorland et al. (2007) reported N intake in manure and in milk as 63 and 25%, respectively, which resulted in 12% unaccounted N loss (diet CP = 21.7%). Castillo et al. (2001) reported an N balance with N intake in feces, urine, and milk being 31.8, 36.1, and 26.1%,

**Table 4. Measured emission rates of $NH_3$, $N_2O$, and $CH_4$ from dairy cow operations in the literature.**

| Reference and location | CP† | MY‡ | Measurement method | Emission rates§ | | | Conditions |
|---|---|---|---|---|---|---|---|
| | | | | $NH_3$ | $N_2O$ | $CH_4$ | |
| | % | kg $d^{-1}$ $cow^{-1}$ | | —— g $d^{-1}$ $cow^{-1}$ —— | | | |
| Liu et al. (the present study), Michigan | 15.1–16.5 | 30.4–37.5 | Environmental chamber/room | 30.0 ± 5.2 | 12.8 ± 1.0 | 409 ± 41 | Simulated tie-stall, Holstein cows |
| Moate et al. (2011), Australia | 18.1–20.2 | 21.4–23.4 | Environmental chamber/room | – | – | 460–500 | Simulated tie-stall, Holstein cows |
| Stackhouse et al. (2011), California | 13.1–22.5 | – | Environmental chamber/room | – | 0.02–0.48 | 48–100 | Simulated tie-stall, Holstein and Black-Angus steers |
| Hamilton et al. (2010), California | 20.2–21.0 | 39.8–40.8 | Environmental chamber/room | – | 0.48 | 272 | Simulated tie-stall, Holstein cows |
| Powell et al. (2008), Wisconsin | 15.7–21.5 | 36.0–39.0 | Environmental chamber/room | 5.6–20.5 | – | – | Tie-stall |
| Amon et al. (2001), Austria | – | 16.1–22.4 | FTIR¶ | 5.8 | 0.62 | 194 | Tie-stall |
| Adviento-Borbe et al. (2010), Pennsylvania | 16.4–17.3 | 38.3–43.9 | Static flux chamber (0.018 $m^3$) | 22.1–36.4 | 0.004–0.005 | 5.2–12.2 | Free-stall floor, scraped 2× daily |
| Harper et al. (2009), Wisconsin | – | – | Inverse dispersion technique | 7.9–38.4 | – | – | Free-stall barns, sand bedding, routinely scraped |
| Li et al. (2009), Virginia | 15.9–17.8 | 40.8–41.6 | Dynamic flux chamber (0.049 $m^3$) | 3.3–3.5 | – | – | Free-stall, Holstein cows, flushed 4× daily |
| van Dorland et al. (2007), Switzerland | 20.6–23.8 | 24.5–26.4 | Environmental chamber/room | – | – | 414–463 | Free-stall, Holstein and Brown Swiss cows |
| Moreira and Satter (2006), Wisconsin | 18.5–19.3 | 27.7–36.6 | N:P indirect method | 91–214 | – | – | Free-stall, scraped 2× or 6× daily |
| Cassel et al. (2005), California | 13.7–17.4 | – | Micrometeorological mass balance method | 61–125 | – | – | Free-stall, elevated area for animals, cement alley lane for manure |
| Zhang et al. (2005), Denmark | – | 23.0–34.9 | Direct measurement of concentrations in barns | 10–122 | 0.1–8.2 | 369–594 | Free-stall with different floors |
| Johnson et al. (2002), Washington | 16.4–17.2 | 32.3–39.3 | Tracer gas method (sulfur hexafluoride) | – | – | 389–456 | Free-stall, Holstein cows |
| Ngwabie et al. (2009), Sweden | – | 31–33 | Direct measurement of concentrations in barns | 21–27 | – | 26–312 | Cubicles with liquid manure system, Holstein cows |
| Luo and Saggar (2008), New Zealand | – | – | Static flux chamber | – | 0.45 | 22.9 | Stand-off pad (300 $m^2$), nonlactating Friesian cows |
| Leytem et al. (2011), Idaho | 17.6 | 34 | Inverse dispersion technique | 130 | 10 | 490 | Open lots (55 $m^2$ $cow^{-1}$), Holstein cows |
| Bjorneberg et al. (2009), Idaho | – | – | Open-path FTIR | 40–250 | – | 200–550 | Open lots (60 $m^2$ $cow^{-1}$) |
| Denmead et al. (2008), Australia | – | – | Backward Lagrangian stochastic model | 29–84 | 3.5–5.7 | – | Open lots (12–23 $m^2$ $cow^{-1}$) |
| Mukhtar et al. (2008), Texas | – | – | USEPA-approved flux chamber protocol | 17–32 | – | – | Open lots (50 $m^2$ $cow^{-1}$) |
| Flesch et al. (2007), Texas | – | – | Backward Lagrangian stochastic model | 150 | – | – | Open lots (14 $m^2$ $cow^{-1}$) |
| Laubach and Kelliher (2005), New Zealand | – | – | Backward Lagrangian stochastic model | – | – | 402 | Fenced grazing (61 $m^2$ $cow^{-1}$) |
| Jungbluth et al. (2001), Germany | – | – | – | – | 0.14 –2.01 | 194–390 | Data from dairy farms including tie-stall and loose housing systems |

† Crude protein.

‡ Milk yield.

§ When necessary, data were converted from original units for comparison across studies.

¶ Fourier transform infrared.

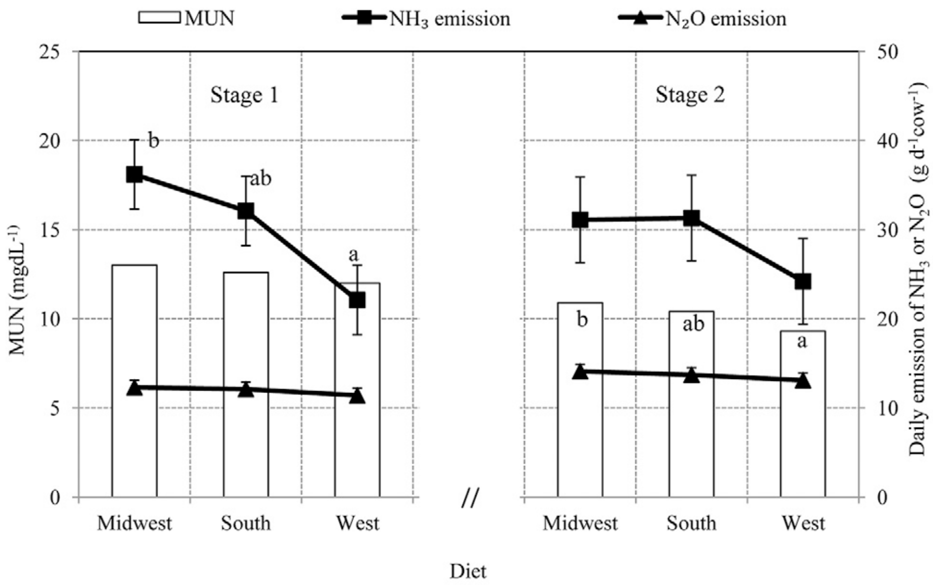

**Fig. 1. Diet effects on milk urea nitrogen (MUN) concentrations and daily emissions of $NH_3$ and $N_2O$. Diets represented feed ingredients commonly fed to dairy cows in the midwestern (Midwest), southern (South), or western (West) United States.**

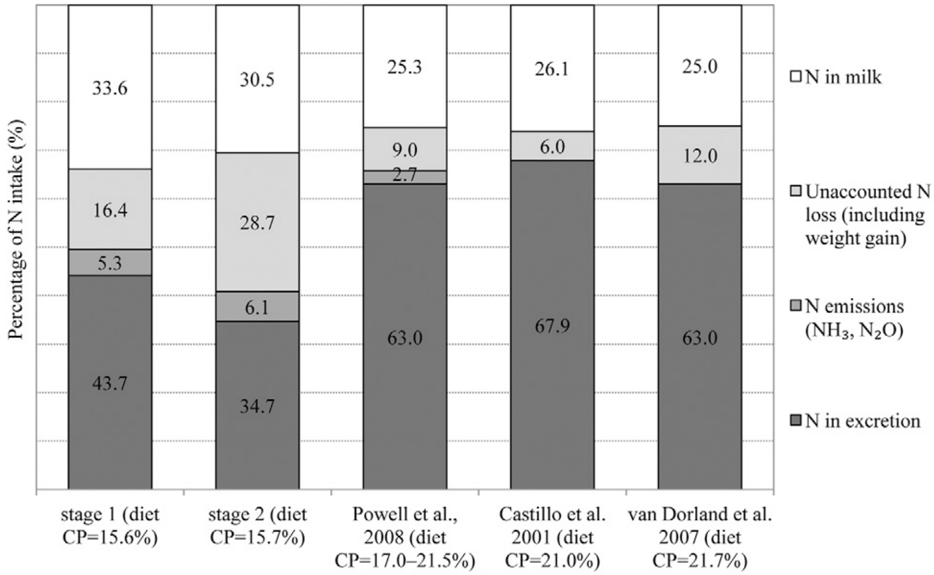

**Fig. 2. Comparison of N balance in dairy cows in this study (Stages 1 and 2 of lactation) and in other studies. CP, crude protein.**

respectively, resulting in 6.0% of unaccounted N loss (diet CP = 21.0%). Manure samples were collected immediately after excretion and thus N lost to gas emissions was not included and was likely minimal (Castillo et al., 2001). Powell et al. (2008) reported an N balance for Holstein cows with N intake in manure, milk, gas emissions, and live weight gain as 63.0, 25.3, 2.7, and 1.3%, respectively, which resulted in 7.7% unaccounted N loss (diet CP = 17.0–21.5%). The percentage of total N excreted (remaining in manure + gas emissions) in the present study (49.0% in Stage 1 and 40.8% in Stage 2) was relatively low compared with that reported by van Dorland et al. (2007), Castillo et al. (2001), Nennich et al. (2005), and Powell et al. (2008), of 63.0, 67.9, 70.0, and 63.0%, respectively. This could be due in part to the relatively low diet CP concentration and fetal growth needs (not accounted for in this study). This result indicates that measurement errors could result in large uncertainties if a mass balance approach is to be used to estimate N emissions from dairy cows.

## Emissions of Methane and Carbon Dioxide

Fats in diets have been previously shown to depress $CH_4$ production from ruminants (Dong et al., 1997; Dohme et al., 2000; Grainger et al., 2010; Moate et al., 2011). Also, manure from cows fed diets containing higher CP levels can yield more $CH_4$ (Amon et al., 2006). Greater emissions of $CH_4$ per unit DMI from cows offered the Midwest diet could be related to the lower fat concentration and higher CP in the Midwest diet. Lower emissions of $CH_4$ per unit DMI or per unit MY were observed from cows offered the South diet in Stage 1 as compared with that from cows offered the West diet, although the South diet had similar fat concentration and higher CP concentration as compared with the West diet. The lower $CH_4$ emissions from cows offered the South diet could be attributed to the hominy and cottonseed in the South diet. Both hominy and cottonseed have been shown to reduce $CH_4$ emissions (Moate et al., 2011; Grainger et al., 2010).

The emission rates across all diets averaged 409 and 14,620 g $d^{-1}$ $cow^{-1}$ for $CH_4$ and $CO_2$, respectively. The measured $CH_4$ emission rates in the present study agreed well with those reported by other researchers (Table 4). The measured $CO_2$ emission rates were comparable with 9884 to 14,589 g $d^{-1}$ $cow^{-1}$ reported by Kinsman et al. (1995) and 12,326 to 13,827 g $d^{-1}$ $cow^{-1}$ reported by van Dorland et al. (2007). In the present study, a correlation was observed between daily emissions of $CH_4$ and $CO_2$ ($r^2$ = 0.48). A correlation has also been reported by Ngwabie et al. (2009), Amon et al. (2001), and Kinsman et al. (1995).

## Greenhouse Gas Emissions in Carbon Dioxide Equivalent Units

Both $CH_4$ and $N_2O$ have been identified as important GHG along with $CO_2$. The 100-yr global warming potential (GWP) of $CH_4$ is 25 times that of $CO_2$, and GWP of $N_2O$ is 298 times that of $CO_2$ (IPCC, 2007). The average GHG emissions from cows in the present study were estimated to be 28.8 kg $d^{-1}$ $cow^{-1}$ in $CO_2$ equivalent units, or 0.87 kg $CO_2$ equivalent kg $milk^{-1}$; the

$CO_2$ equivalent unit distribution between $CO_2$, $CH_4$, and $N_2O$ was 50.8, 35.5, and 13.8%, respectively. The $CO_2$ generated by agriculture is often considered to be biogenic in nature or "carbon neutral" (in contrast to $CO_2$ from fossil-fuel combustion, which adds new carbon to the atmospheric–biospheric circulation system), and therefore sometimes is excluded or deferred in accounting of total GHG emissions.

## Conclusions

The three diets formulated to represent feed ingredients typical of the Midwest, South, or West regions of the United States were investigated for their effects on feed intake, MY, excretion, and gas emissions for lactating dairy cows. One limitation of the present study is that we evaluated the regional ingredients while maintaining all animals in the environmental rooms in the Midwest, though this also allowed us to not have temperature and ingredient effects confounded. The results showed that the three diets resulted in different gas emissions while maintaining similar DMI and MY, which indicated new thoughts on alternative dietary strategies for mitigating gas emissions. Ingredient selection will have some influence on gas emissions because of compositional parameters beyond those that serve as the primary formulation criteria. The study also provided valuable data on N balance, GHG emissions, and the effects of lactation stage for dairy operations.

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