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## Short communication: Comparison of the GreenFeed system with the sulfur hexafluoride tracer technique for measuring enteric methane emissions from dairy cows

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### ABSTRACT

The objective of this study was to compare 2 commonly used techniques for measuring methane emissions from ruminant animals: the GreenFeed (GF) system and the sulfur hexafluoride (SF<sub>6</sub>) technique. The study was part of a larger experiment in which a methane inhibitor, 3-nitrooxypropanol, fed at 4 application rates (0, 40, 60, and 80 mg/kg of feed dry matter) decreased enteric methane emission by an average of 30% (measured by both GF and SF<sub>6</sub>) in a 12-wk experiment with 48 lactating Holstein cows fed a total mixed ration. The larger experiment used a randomized block design and was conducted in 2 phases (February to May, phase 1, and June to August, phase 2), with 2 sets of 24 cows in each phase. Using both GF and SF<sub>6</sub> techniques, methane emission data were collected simultaneously during experimental wk 2, 6, and 12 (phase 1) and 2, 9, and 12 (phase 2), which corresponded to a total of 6 sampling periods. During each sampling period, 8 spot samples of gas emissions (staggered over a 3-d period) were collected from each cow using GF, as well as 3 × 24-h collections using the SF<sub>6</sub> technique. Methane emission data were averaged per cow for the statistical analysis. The mean methane emission was 373 (standard deviation = 96.3) and 405 (standard deviation = 156) g/cow per day for GF and SF<sub>6</sub>, respectively. Coefficients of variation for the 2 methods were 25.8 and 38.6%, respectively; correlation and concordance between the 2 methods were 0.40 and 0.34, respectively. The difference in methane emission between the 2 methods (SF<sub>6</sub> – GF) within treatment was from 46 to 144 and 24 to 27

g/d for phases 1 and 2, respectively. In the conditions of this experiment, the SF<sub>6</sub> technique produced larger variability in methane emissions than the GF method. The overall difference between the 2 methods was on average about 8%, but was not consistent over time, likely influenced by barn ventilation and background methane and SF<sub>6</sub> concentrations.

**Key words:** methane, GreenFeed, sulfur hexafluoride, dairy cow

### Short Communication

An important component of agricultural greenhouse gas (GHG) mitigation efforts is an accurate measurement of GHG emissions. Several procedures for measuring enteric methane emissions, 1 of the 2 most important GHG from animal agriculture, have been developed and used with variable success (Hammond et al., 2016). Among the most widely used procedures for measuring enteric methane are the sulfur hexafluoride (SF<sub>6</sub>) tracer technique (Johnson et al., 1994) and, more recently, the GreenFeed system (GF; Zimmerman et al., 2011). Comparative studies reported relatively good agreement between SF<sub>6</sub> or GF with respiration chamber data (Pinares-Patiño et al., 2011; Muñoz et al., 2012; Hammond et al., 2015). With respect to SF<sub>6</sub>, recent modifications to the technique decreased between-animal coefficients of variation (CV) for methane yield (i.e., g/kg of DMI) to levels comparable to CV obtained using chambers (Deighton et al., 2014). Similarly, a study with growing dairy heifers concluded that estimates of methane emission generated by GF were comparable to values obtained by respiration chambers (Hammond et al., 2015). Those authors pointed out, however, that deployment of the GF units and replication must be carefully considered to ensure

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sufficient numbers of measurements are obtained. In a study with lactating dairy cows, methane emissions measured by GF were similar to values derived from respiration chambers and between-animal variability was also within the range observed in respiration chambers (Huhtanen et al., 2013). A direct comparison between SF<sub>6</sub> and GF yielded larger CV for methane emissions and a poor relationship between methane emissions and DMI for the SF<sub>6</sub> method in a naturally ventilated tiestall barn (Dorich et al., 2015). The objective of the current study was to compare enteric methane emission data from dairy cows derived simultaneously using GF and the SF<sub>6</sub> technique, as modified by Deighton et al. (2014).

Animals involved in this experiment were cared for according to the guidelines of the Pennsylvania State University Animal Care and Use Committee. The committee reviewed and approved all procedures carried out in the study. The experiment was part of a larger randomized block design production experiment with 48 Holstein cows (details in Hristov et al., 2015a) and was conducted at the tiestall barn of the Pennsylvania State University's Dairy Research and Teaching Center. The experiment was conducted in 2 phases; the duration of each phase was 14 wk, including 2-wk covariate and 12-wk experimental periods. The objective of the production experiment was to evaluate the effect of an inhibitor, 3-nitrooxypropanol (**3NOP**), on enteric methane emissions in lactating dairy cows. Cows were subjected to the following treatments: control (no additive) and 3NOP applied at 40 (low 3NOP), 60 (medium 3NOP), and 80 (high 3NOP) mg/kg of feed DM. The diet was based on corn silage and alfalfa haylage, corn grain, whole roasted soybeans, a bakery by-product meal, and canola meal and was fed as a TMR (Hristov et al., 2015a) once daily with 3NOP or placebo premixes mixed in the TMR. The experiment was conducted in 2 phases (with 24 cows in each phase), with phase 1 from February to May 2014 and phase 2 from June to August 2014. Phase 2 began immediately following phase 1.

Enteric methane emissions were measured using GF (C-Lock Technology Inc., Rapid City, SD) and the SF<sub>6</sub> technique (Deighton et al., 2014). Methane emission data were collected simultaneously from individual cows using both techniques during experimental wk 2, 6, and 12 (phase 1) and 2, 9, and 12 (phase 2), which corresponded to a total of 6 sampling periods. With GF, during each sampling period gas emission data were collected in 3 consecutive days starting at 0900, 1500, and 2100 h (sampling d 1), 0300, 1200, and 1700 h (sampling d 2), and 0000, and 0500 h (sampling d 3). This resulted in an average gas collection period of 40 (8 × 5) min/cow per sampling period. Three GF units

were used and all cows were sampled within 50 min. Breath gas samples were collected for 5 min from each cow followed by a 2-min background gas sample collection. Calibration of the GF units was as described in Hristov et al. (2015b). Bait feed was offered at each of the 8 sampling events for a total of 4 kg/cow over 3 d, which was approximately 5% of the total DMI for each cow during each 3-d sampling period. The bait feed used was a premix containing (as-is basis) 70% ground corn grain, 28% dried molasses, and 2% soybean oil.

For the SF<sub>6</sub> method, permeation tubes containing SF<sub>6</sub> were delivered into the reticulum of each cow using a bolus gun, 1 wk before the first measurement occurred. The mean (±SD) rate of SF<sub>6</sub> release from permeation tubes used in the experiment was 4.38 ± 0.261 mg/d. The SF<sub>6</sub> equipment, sample and canister processing, and analysis of the gas samples for methane and SF<sub>6</sub> were as described in Deighton et al. (2014). Briefly, evacuated canisters were secured on the back of each cow using a harness. Breath gas was continuously collected through tubing extending to the nostrils of the cow. A total of 8 background gas collection canisters were placed on the back of 8 selected cows, evenly distributed in the barn. Background air samples were collected during each day of the SF<sub>6</sub> measurements from the area on the back of the cows. Canisters were replaced every 24 h for an average sampling duration of 3.2 ± 0.10 d/cow and per sampling period. The average gas collection time was 4,620 min/cow per sampling period. An aliquot of the collected gas sample was extracted and analyzed for methane and SF<sub>6</sub> using a gas chromatograph (Varian CP-3800, Varian Analytical Instruments, Walnut Creek, CA) as described in Williams et al. (2011) and Hristov et al. (2015a).

Methane emission data were averaged per cow and sampling period (GF = 8 observations and SF<sub>6</sub> = 3.2 observations per cow and sampling period) and the average values were used in the statistical analysis. Data were analyzed and outliers were removed based on an absolute studentized residual value >3. Descriptive statistics were computed and differences between average SF<sub>6</sub> and GF methane emission data (i.e., SF<sub>6</sub> – GF; g/d) for wk 2 and 12 only were analyzed using the GLIMMIX procedure of SAS (version 9.4; SAS Institute Inc., Cary, NC). The model contained block, treatment, sampling period, and block × treatment and sampling period × treatment interactions. Analyses were carried out separately for each phase and statistical differences were declared at  $P \leq 0.05$ . Additionally, Lin's concordance correlation coefficient (Lin, 1989) and Pearson correlation coefficient were calculated (all study weeks). All computations were done using SAS.

Descriptive statistics of the methane emission data are shown in Table 1. Overall, the mean methane emis-

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