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Determination of gas flux and animal performance test duration of growing cattle in confined conditions

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Abstract

Data from three experiments was analyzed to determine the number of visits and days to assess gas flux (CH_4, CO_2, and O_2), dry matter intake (DMI), and average daily gain (ADG) from growing animals under confined conditions. In experiment 1, 213 animals (461 ± 91 kg initial body weight [BW]) were fed a backgrounding diet and evaluated for 60 d. In experiment 2, 169 steers (488 ± 37 kg initial BW) were fed a finishing diet and assessed for 70 d. In experiment 3, 64 steers (514 ± 42 kg initial BW) were fed a finishing diet and evaluated for 80 d. In each experiment, animals were placed in one pen with one Greenfeed and five SmartFeeds to collect gas flux and feed intake simultaneously. Gas flux was analyzed using data from 161 animals from the three experiments with 100 visits for 2 or more min or 3 or more min. Also, metabolic heat production (MHP) was estimated using the individual gas flux. Daily DMI was calculated as the daily feed intake corrected by the dry matter concentration. ADG was computed as the slope of the regression of the shrunk BW (96% BW) throughout each of the experimental periods. The mean gas flux and MHP were estimated for increasing or decreasing 5-visit intervals starting with the first or the last 5 visits and increasing or decreasing until the full 100-visit dataset was utilized, respectively. Intervals of DMI were estimated for increasing or decreasing 5-d intervals starting with the first or the last 5 d and increasing or decreasing until the end of the experimental period, respectively. Intervals of ADG were estimated for increasing or decreasing measurement period intervals until the end of the experimental period, respectively. Pearson and Spearman correlations were computed between the maximum visits or days and each shortened visit or day interval. The minimum number of visits and days was determined when correlations with the total visits were greater than 0.95. The results indicated that the minimum number of visits needed to quantify CO_2, O_2, and MHP accurately was 40, while CH_4 was 60. A visitation length of 2 min or more or 3 min or more did not modify the gas flux determination. Thus, based on the average daily visitation in these experiments, gas flux data could be collected for 25 d. Additionally, the required days to determine DMI was 30, while ADG could not be assessed in a shorter than 60-d period.

Lay Summary

The evaluation of sustainable practices and the selection of efficient animals requires assessing gas flux (CO_2, CH_4, and O_2) and animal performance (e.g., dry matter intake [DMI] and average daily gain [ADG]). Data from three experiments were used to determine the minimum number of visits and days to determine gas flux, DMI, and ADG from growing animals in confined conditions. The results indicated that the minimum number of visits needed to quantify CO_2 and O_2 accurately was 40, while CH_4 was 55. A visitation length of 2 min or more or 3 min or more did not modify the gas flux determination. Thus, based on the average daily visitation in these experiments, gas flux data could be collected for 25 d. Additionally, the required days to determine DMI was 30, while ADG could not be assessed in a shorter than 60-d period.

Key words: backgrounding diet, carbon dioxide emission, finishing diet, methane emission, oxygen consumption

Introduction

Methane (CH_4) is an important greenhouse gas with 28 times greater global warming potential than carbon dioxide (CO_2; EPA, 2023). Enteric CH_4 represents 40% of the global emissions from the agricultural sector (Gerber et al., 2013) and one-third of the anthropogenic CH_4 emissions (United Nations Environment Programme and Climate and Clean Air Coalition, 2021). Enteric CH_4 is produced during the anaerobic fermentation of organic matter (Ungerfeld, 2020), and it is an energy loss for ruminants that varies between 2% and 12% of the gross energy intake according to the diet characteristics (Johnson and Johnson, 1995).

In the last few decades, there has been a growing interest in quantifying CH_4 emissions under commercial conditions and developing mitigation strategies to decrease CH_4 from ruminants (Hristov et al., 2015; Beauchemin et al., 2022a; Vargas et al., 2022). In this regard, techniques and methodologies to determine precisely and accurately CH_4 emissions are necessary and require field validation (Hristov et al., 2015; Hammond et al., 2016). Additionally, circadian emissions of enteric CH_4 and the variety of production systems impose constraints regarding the selection of technique and methodology.

The automated gas quantification system (AGQS) allows the estimation of CH_4, CO_2, oxygen (O_2), and hydrogen (H_2) gas fluxes using spot-sample measurements (Hegarty, 2013; Hammond et al., 2016). Consequently, animals should visit the AGQS throughout the day to adequately represent the gas flux (Hegarty, 2013). There are different methodological recommendations for using the AGQS regarding the visit length, airflow, and experimental replicates (Arthur et al., 2017; Gunter and Bradford, 2017; Gunter and Beck, 2018;

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Della Rosa et al., 2021). Additionally, there is interest in reducing enteric CH_4 and increasing animal efficiency via genetic selection (Hristov et al., 2013; Beauchemin et al., 2022b). Therefore, animal performance and gas flux collection periods should simultaneously be evaluated. However, there is limited information regarding evaluating gas flux and animal performance in growing animals under confined conditions.

Initial evaluation reported poor concordance on daily CH_4 emissions between AGQS and the respiration chamber, possibly associated with the low representativeness and short visit length of the spot-sample measurements (Hammond et al., 2015). Some authors suggested between 4 and 8 wk to determine CH_4 emissions with more than 3-min length visits (Hegarty, 2013; Cottle et al., 2015; Renand and Maupetit, 2016). However, each experiment varied visitation length and number of visits (Della Rosa et al., 2021). Visit recommendations to determine CH_4 emissions using the AGQS varies between 8 and 50 (Hristov et al., 2015; Manafiazar et al., 2016; Dressler et al., 2023), while visit length ranges from 2 or more or 3 or more min (Hegarty, 2013; Dressler et al., 2023). The optimum visit recommendation will depend on the experimental design, facility characteristics, animal type and physiological state, and data analyses. For example, Dressler et al. (2023) suggested a minimum of 38 to 40 visits to determine mature cows' gas flux under grazing conditions.

The evaluation of animal performance is essential to recognize the variability of an animal population or the effect of a dietary intervention. In this regard, the recommendation period to determine dry matter intake (DMI), average daily gain (ADG), feed conversion ratio, residual feed intake, and water intake was 35 to 43, 56 to 71, 42 to 70, 56 to 63, and 35 d, respectively (Archer et al., 1997; Wang et al., 2006; Culbertson et al., 2015; Ahlberg et al., 2018; Marzocchi et al., 2020; Ryan et al., 2022). In this regard, the experimental length to determine proper gas flow and animal performance is different and must be assessed under various ruminant production systems. Thus, the objectives of this report were to determine the number of visits and the visitation length to assess the gas flow using the AGQS and the number of days to determine the DMI and ADG in growing animals under confined conditions.

Materials and Methods

Ethics Statement

Data from three experiments were analyzed to establish the number and length of visits for determining gas flux and the number of days for assessing DMI and ADG in growing cattle. The studies were conducted at the Climate Smart Research Facility at Colorado State University, CO. All procedures involving animals were approved by the Colorado State University Institutional Animal Care and Use Committee (experiment 1, 1,526; experiment 2, 4,072; and experiment 3, 4,689).

Experimental Designs

Experiment 1: animals, feed management, and experimental conditions. A total of 213 animals were located in one of the six similar pens. Animals consisted of Angus (105 steers, 43 heifers, and 15 bulls; 445 ± 71.2 kg initial body weight [BW]), Hereford (20 heifers and 25 bulls; 525 ± 59.9 kg initial BW), and Wagyu (3 heifers and 2 bulls; 409 ± 37.5 kg initial BW) crossbreds. All animals were offered a backgrounding diet composed of corn silage, alfalfa hay, wheat straw, dry distillers grains, whole corn, liquid supplement, and salt and limestone (19%, 13%, 11%, 20%, 33%, 2.8%, and 1.2% on a dry matter [DM] basis, respectively). Feed samples were weekly collected, dried, composited, and analyzed by a commercial laboratory using a wet chemistry package (Dairy One, Ithaca, NY). Chemical composition was 45.8% neutral detergent fiber (NDF), 10.4% crude protein (CP), 18.7% starch, and 0.86 Mcal/kg DM of net energy of gain (NE_g, Table 1).

Intake and ADG were evaluated for 60 d. Individual intake was recorded daily during the evaluation period. Animals were weighed on days −1 and 0 to obtain the initial BW and on days 59 and 60 to record the final BW. In addition to the initial and final BW, unshrunk BW was obtained every 15 d during the evaluation period.

Experiment 2: animals, feed management, and experimental conditions. A total of 169 Angus steers (approximately 13 mo of age and 488 ± 37.4 kg initial BW) were located in one of the six similar pens. Steers were offered a finishing diet composed of steam-flaked corn, corn silage, dry distiller grains with soluble, Tylan, PMS liquid supplement, and an ionophore (60%, 27%, 4%, 4%, 3%, and 2% on a DM basis, respectively). Feed samples were weekly collected, dried, composited, and analyzed by a commercial laboratory using a wet chemistry package (Dairy One, Ithaca, NY). Chemical composition was 16.8% NDF, 12.8% CP, 55.3% starch, and 1.37 Mcal/kg DM of NE_g (Table 1).

The intake of DM and ADG was evaluated for 70 d. Individual intake was recorded daily during the evaluation period. Also, steers were weighed on days −1 and 0 to obtain the initial BW and on days 69 and 70 to record the final BW. In addition to the initial and final BW, unshrunk weights were obtained during the evaluation period on days 22 and 45.

Experiment 3: animals, feed management, and experimental conditions. A total of 64 Angus steers (approximately 15 mo of age and 514 ± 42 kg initial BW) were stratified by BW in two groups (heavy and light) and located in one of the two similar pens. Steers were offered a finishing diet composed of steam-flaked corn, corn silage, dry distiller

Table 1. Chemical compositions from the diets offered during the experimental periods

Composition (%of DM)	Experiment 1	Experiment 2	Experiment 3
Dry matter, % as fed	66.8	65.2	66.5
Crude protein	10.4	12.8	13.7
Neutral detergent fiber	45.8	16.8	17.5
Acid detergent fiber	29.6	9.9	8.8
Non-fiber carbohydrates	33.0	62.2	61.3
Starch	18.7	55.3	53.8
Ether extract	3.4	4.3	3.1
Ash	7.5	3.9	4.5
Net energy of gain, Mcal/kg of DM	0.86	1.37	1.32

grains with solubles, PMS liquid supplement, Tylan, and an ionophore (65%, 20%, 7%, 4%, 2%, and 2% on a DM basis, respectively). Feed samples were weekly collected, dried, composited, and analyzed by a commercial laboratory using a wet chemistry package (Dairy One, Ithaca, NY). Chemical composition was 17.5% NDF, 13.7% CP, 53.8% starch, and 1.32 Mcal/kg DM of NE_g (Table 1).

Animal performance was evaluated for 52 and 80 d for the heavy and light groups, respectively. Individual intake was recorded daily during the evaluation period. Steers were weighed on days −1 and 0 to obtain the initial BW, and on days 51, 52, 79, and 80 to record the final BW for the heavy and light groups, respectively. In addition to the initial and final BW, unshrunk weights were obtained on day 21 for both groups and day 63 for the light group during the performance evaluation.

Gas Flux, Feed Intake, and BW

In each experiment, animals were located in pens containing five SmartFeed and one GreenFeed (C-Lock, Rapid City, SD) for the simultaneous collection of feed intake and gas fluxes (CH_4, CO_2, and O_2). Before using the units, animals individually received radio frequency electronic ID (RFID, Allflex, USA Inc.,). Animals were exposed to the Greenfeed and Smartfeed units during an acclimation period of approximately 2 wk before data collection. After the acclimation period, cattle panels were used to ensure only one animal at a time had access to GreenFeed.

Steers were allowed to visit the GreenFeed units every 4 h (up to 6 visits per day) and consume up to 6 drops of alfalfa pellet (approximately 35 g/drop) with 30-s spacing between drops. This encourages animals to visit the units throughout the day and ensures animals stay at the GreenFeed for an appropriate gas flux collection.

The emission rate of gases (Qc) was calculated using the following equation (Huhtanen et al., 2015):

$$Qc = [Cp \times (Conc - Bconc) \times Qair] \div 10^6$$

Where, Cp is the fractional capture rate of air, Conc is the concentration of captured gas, BConc is the background concentration of gas, and Qair is the volumetric airflow. Thus, the gas flux (Qm) was calculated using the following equation:

$$Qm = Qc \times 273.1 \div (273.15 + \text{Tair}) \times \text{GD}$$

Where, Tair is the air temperature, and GD is the density of gas at 1 atm and 273.5 K.

To ensure the whole system's performance, CO_2 recovery tests were performed monthly throughout the experiment and at the beginning and end of each experiment. Additionally, zero and span calibrations of the CH_4, CO_2, and O_2 gas analyzers were performed every 3 d via an onboard autocalibration system. Raw collection data were validated by C-Lock Inc., which included checking head proximity, visit length, and airflow and wind corrections. Additionally, data was excluded when the length of the visit was less than 2 min, and the airflow was lower than 26 L/s (Arthur et al., 2017; Gunter and Beck, 2018).

Animals were fed ad libitum and had constant access to fresh water. Individual feed intake was recorded daily for 50, 95, and 54 d using the Smartfeed technology (C-Lock, Rapid City, SD) for experiments 1, 2, and 3, respectively. Additionally, animals were weighed through the experimental period as previously described.

Calculation and Statistical Analysis

Two different databases using the same animals ($n = 161$) were defined to calculate the emissions of CH_4, CO_2, and O_2 using the first 100 visits with 2 or more and 3 or more min of each animal, respectively. Thus, gas flux was evaluated in 27, 122, and 12 animals from experiments 1, 2, and 3, respectively (Table 2). Additionally, metabolic heat production (MHP) was calculated for each visit using Brouwer's equation, omitting the nitrogen excretion (Brouwer, 1965):

$$\text{MHP } (Mcal/d) = 3.866 \times O_2\ (L/d) + 1.2 \times CO_2\ (L/d) - 0.518 \times CH_4\ (L/d)$$

The DMI was calculated as the average feed intake corrected by the DM concentration of the diet during the experimental period. Animal growth was determined by lineal regressions of the shrunk BW (0.96 x BW) against time, and the calculated slope was considered the ADG.

Average CH_4, CO_2, O_2, MHP, DMI, and ADG were calculated following the approach described by Dressler et al. (2023). Briefly, gas flux variables were estimated for increasing (forward) or decreasing (reverse) 5-visit intervals starting with the first or the last 5 visits and increasing or decreasing until the full 100-visit dataset was utilized, respectively. Intervals of DMI were estimated for increasing (forward) or decreasing (reverse) 5-d intervals starting with the first or the last 5 d and increasing or decreasing until the end of the experimental period, respectively. Finally, intervals of ADG were estimated for increasing (forward) or decreasing (reverse) measurement period intervals according to the weighting periodicity in each experiment.

Descriptive statistics were conducted with the SAS 9.4 statistical packages. The residual variance was estimated for each variable according to the interval duration by fitting a mixed model with repeated measurements as follows:

$$Yij = \mu + \text{Interval}i + \text{Animal}j + \varepsilon ij$$

Where, Y_{ij} was the flux of CH_4, CO_2, O_2, MHP, DMI, or ADG of each animal for each interval duration; Interval_i was the fixed effect of the estimated period; Animal_j accounted for the random effect of each evaluated animal, and ε_{ij} was the random residual effect of each observation. The compound symmetry covariance structure was selected according to the lowest Akaike Information Criterion using the Mixed procedure of SAS 9.4 to allow for heterogenous variances over the test duration and correlation among them. A relative change of variance, defined as the percentage difference between the variance obtained from the previous measurement and the current measurement divided by the variance obtained for the first 5-d measurement, was calculated to compare the residual variances among intervals (Wang et al., 2006).

Additionally, Pearson and Spearman phenotypic correlation coefficients were estimated for each interval compared to the total visits or days using the Corr procedure of SAS 9.4.

Table 2. Gas flux (g/d) of CO_2, CH_4, and O_2 from animals visiting 2 or more and 3 or more min in the first 100 visits to the open-circuit gas quantification system during the experimental periods

Visitation length, min	*n*	CO_2, g/d		CH_4, g/d		O_2, g/d	
		Mean	SD	Mean	SD	Mean	SD
Experiment 1							
2 or more	27	5,639	953.7	171	32.1	3,885	746.4
3 or more	27	5,671	974.0	172	31.9	3,921	752.0
Experiment 2							
2 or more	122	9,509	874.2	150	24.8	6,588	557.3
3 or more	122	9,726	890.9	154	25.5	6,725	568.4
Experiment 3							
2 or more	12	10,438	592.6	161	24.9	7,899	401.6
3 or more	12	10,527	478.6	166	23.6	7,875	350.4

Finally, repeatability (r) for gas flow, MHP, DMI, and ADG was calculated as follows:

$$r = \partial^2_{Cow} / (\partial^2_{Cow} + \partial^2_{\mathrm{Re}\,sidual})$$

Where r was the repeatability of each gas or MHP; ∂^2_{Cow} was the variance of each animal; and $\partial^2_{Residual}$ was the variance of the residual.

Results and Discussion

Number of Visits and Visitation Length for Determining Gas Flux and MHP

Gas flux quantification using a spot-sample technique such as the AGQS requires periodical visitation by the animal to represent the circadian dynamic of CO_2 and CH_4 production and O_2 consumption (Cottle et al., 2015; Hristov et al., 2015; Hammond et al., 2016). Additionally, gas flux determination allows the estimation of the MHP (Dressler et al., 2023). In this report, the required number of visits to represent the CO_2, O_2, and MHP was 20, using a Pearson correlation coefficient greater than 0.95, either when visitation length was 2 or more or 3 or more min (Figure 1). However, the required number of visits to represent CH_4 was 55 and 60 when animals visited for 2 or more min and 3 or more min, respectively (Figure 1). Differences in Pearson and Spearman correlation in CO_2, O_2, and MHP suggested a re-ranking of animals across the visit intervals (Ahlberg et al., 2018). However, Pearson and Spearman's correlation was greater than 0.95 when animals visited 40 times the AGQS (Figure 1, Supplementary Tables S1 and S2). Additionally, the phenotypical residual variance decreased among gases and MHP similarly when increasing the visit intervals, stabilizing the phenotypical residual variance after 40 visits (Figure 1, Supplementary Tables S1 and S2).

The literature reports a different number of visits to the AGQS for determining gas flux, which varies according to the experimental design and feeding system (Della Rosa et al., 2021). Thus, the number of visits when mature animals visited the AGQS for 2 or more min under grazing conditions varied between 38 to 40 visits (Dressler et al., 2023), while from growing animals under confined conditions was 45 visits (Arthur et al., 2017). Conversely, when animals visited for 3 or more min, 12 to 15 visits were required to determine gas flux from animals under grazing conditions (Gunter and Bradford, 2017), while 30 were under confined conditions (Arthur et al., 2017). These results are similar to the 20 to 40 visits to determine CO_2, O_2, and MHP from this report (Figure 1).

The circadian variation of CH_4 emissions is influenced by intake level and feed characteristics (Blaxter and Clapperton, 1965). In this regard, CH_4 emissions from animals fed concentrate diets showed greater variation throughout the day than those consumed forages (Hales and Cole, 2017; Gunter and Beck, 2018). Differently, less CH_4 variation showed animals consuming a finishing diet in experiments 2 and 3 than those feeding a backgrounding diet in experiment 1 (Table 2). In this report, the number of required visits to determine CH_4 varied from 55 to 60, using the Pearson correlation coefficient greater than 0.95, and was similar to the 50 visits reported in other studies (Cottle et al., 2015; Manafiazar et al., 2016; Renand and Maupetit, 2016). On the other hand, Arthur et al. (2017) recommended between 30 to 45 visits to determine CH_4 emissions using as a reference the stabilization of the phenotypical residual variance, although the Person correlation coefficient was 0.9, similar to results from this report (Figure 1, Supplementary Tables S1 and S2).

The emission of CH_4 has been determined differently when using the AGQS. Some authors used the average of good visits throughout the day (Hammond et al., 2015; Velazco et al., 2016; Starsmore et al., 2023), while others used the average of predefined hour intervals (Manafiazar et al., 2016; Beauchemin et al., 2022a). Usually, using period intervals allows better representativeness of CH_4 emissions throughout the day when animal visitation is limited or in short experimental periods (Beauchemin et al., 2022a). In this report, the average number of individual visits per hour was 4.2 ± 1.7 and 4.3 ± 2.2 when gas flux was evaluated using 2 or more and 3 or more min, respectively. Additionally, the gas flux was collected during 60, 70, and 80 d, capturing the circadian variability of CH_4 for experiments 1, 2, and 3, respectively.

The repeatability of CH_4 and CO_2 increased when increasing the evaluation period from 0.44 to 0.90 and 0.62 to 0.93, respectively (Arbre et al., 2016; Renand and Maupetit, 2016; Ryan et al., 2022). Similarly, the repeatability of CH_4 and CO_2 increased from 0.54 to 0.77 and 0.62 to 0.85 when increasing the visit intervals, respectively (Supplementary Tables S1 and S2). Differences in repeatability among experiments could be related to the animal type, diet characteristics, and experimental design (Ryan et al., 2022).

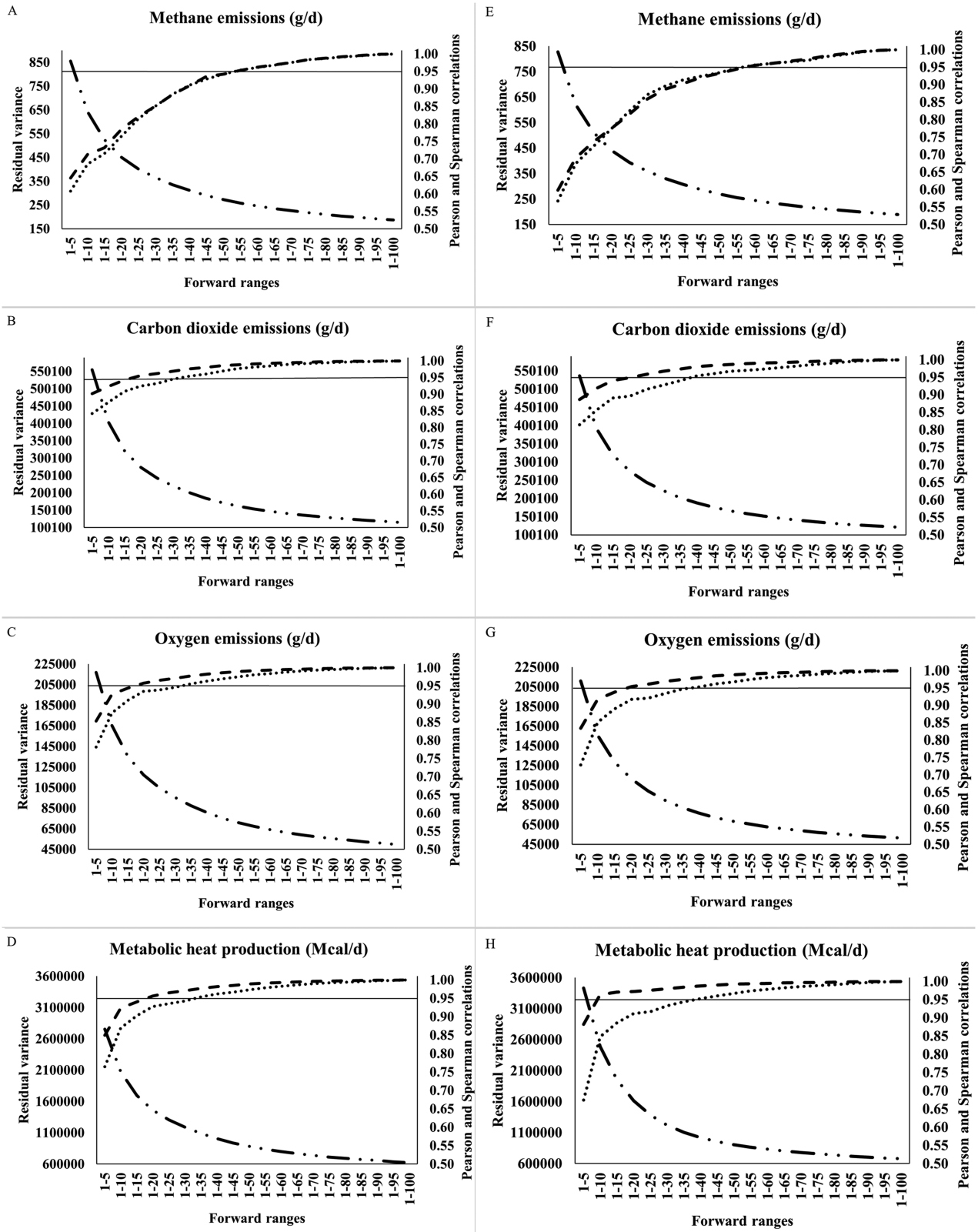

Figure 1. Residual variance (— —) and Pearson (– – –) and Spearman (·····) correlations across visit intervals of gas flux (g/d) and metabolic heat production (Mcal/d) from animals visiting the open-circuit gas quantification system during 2 or more min (A, B, C, and D) or 3 or more min (E, F, G, and H). Continued line represents 0.95.

In this report, the analyses of the visitation length did not affect the required number of visits to determine gas flux and MHP (Figure 1). Previously, gas flux has been evaluated using 2 or more (Ryan et al., 2022; Dressler et al., 2023) or 3 or more min (Hegarty, 2013; Huhtanen et al., 2015; Velazco et al., 2016). Arthur et al. (2017) reported a 50% increase in required visits when gas flux was determined with 2 or more min relative to 3 or more min in growing steers; however, they did not find differences in heifers, possibly associated with different adaptation periods. In this regard, Gunter and Beck (2018) stated that the visitation length did not affect the gas flux estimation after an adequate adaptation period. Experiments accounted for 2 wk of adaptation before the data collection, explaining the similar results when animals visited 2 or more and 3 or more min (Figure 1).

Number of Days for Determining DMI and ADG

Intake of DM plays a significant role in CH_4 emissions (Blaxter and Clapperton, 1965; Molano and Clark, 2008). In this regard, strategies to evaluate sustainability should include the calculation of DMI. However, DMI determination presents challenges because it is affected by multiple factors such as dietary characteristics, animal variation, or feeding management (Forbes, 2005). In this report, the required number of days to determine DMI was 30 for experiments 1 and 3, using the Pearson correlation coefficient greater than 0.95, and 60 for experiment 2. However, the phenotypical residual variance was stabilized after 30 d in experiment 2 (Table S3).

Similarly to the results in this report, the recommended number of days to determine DMI varies between 35 and 42 d (Archer et al., 1997; Wang et al., 2006; Basarab et al., 2013; Culbertson et al., 2015; Renand and Maupetit, 2016; Ahlberg et al., 2018; Marzocchi et al., 2020). In this report, animals from experiment 1 relative to experiment 2 and 3 fed diets with contrasting characteristics, resulting in different reductions of the phenotypic residual variance throughout the visit intervals (Supplementary Table S3). In experiment 1, a backgrounding diet with greater fiber concentration was offered, while in experiments 2 and 3, a finishing diet with greater CP and NE_g was provided (Table 1). Thus, the phenotypic residual variance rapidly decreased when the interval days in animals fed a finishing diet were increased relative to those consuming a backgrounding diet (Supplementary Table S3). Thus, feeding animals with finishing diets requires a longer evaluation period of DMI to reduce the residual variance relative to animals consuming a backgrounding diet.

Strategies to evaluate sustainability should define animal productivity, such as ADG. However, the ADG definition is challenged because multiple factors could affect the accurate determination of BW, such as animal feed and water intake, animal handling, or the proper use of measuring equipment. The recommended days to determine ADG varies from 56 to 71 (Archer et al., 1997; Culbertson et al., 2015; Ahlberg et al., 2018; Marzocchi et al., 2020). Additionally, Wang et al. (2006) reported that ADG showed a Pearson correlation coefficient greater than 0.9 at 63 d of evaluation, although the residual variance did not stabilize.

This report evaluated the ADG following the recommended period evaluation for 60, 70, and 80 d in experiments 1, 2, and 3, respectively (Supplementary Table S4). The Pearson and Spearman's correlation coefficients were lower than 0.95, and the residual variances were not stabilized, suggesting that shorter periods are not recommended to determine ADG, as was indicated in previous experiments (Archer et al., 1997; Culbertson et al., 2015; Ahlberg et al., 2018; Marzocchi et al., 2020).

Conclusion

This report determined the required number of visits and days to assess gas flux, DMI, and ADG of growing animals under confined conditions. Determination of the CH_4 production required 60 visits, while CO_2, O_2, and MHP required 40 visits. Visitation length to the AGQS for 2 or more or 3 or more min did not modify the gas flux determination. Thus, based on the average daily visitation in these experiments (i.e., 2.4 visits/d), gas flux data could be collected for 25 d; however, that will depend upon the degree of visitation per day in a given phenotype assessment. Additionally, the required days to determine DMI was 30, while ADG could not be assessed in a shorter than 60-d period.

Supplementary Data

Supplementary data are available at *Translational Animal Science* online.

Conflict of interest statement

The authors declare no real or perceived conflicts of interest.

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