International Conference of Agricultural Engineering
AgEng 2014 Zurich 6–10 July

Ref: C0370

Design and test of an artificial reference cow to simulate methane production

Liansun Wu and Peter W.G. Groot Koerkamp, Farm Technology Group, Wageningen University, P.O. Box 317, 6700 AH, Wageningen, The Netherlands

Nico Ogink, Wageningen UR Livestock Research, Wageningen University and Research Centre, P.O. Box 135, 6700 AC, Wageningen, The Netherlands

Abstract

As a strong greenhouse gas, methane emission from dairy cows is a major contributor from livestock production. Methane emission may be mitigated by breeding and feeding strategies, but requires that methane production from individual cows must be accurately and precisely quantified first. Therefore, we need measurement technology capable of evaluating methane emission from a large number of cows in the barns. For developing such measurement techniques, a known reference source that simulates cow exhalation of methane that can be controlled is necessary to improve and validate measurement methods. The reference cow should be suited to operate in cow barns under practical conditions. In this study, we operated an artificial cow that was designed and constructed to simulate methane production and concentration patterns as measured from real cows in practice. The objective of this study was to test the working accuracy and precision of the artificial cow in defined methane exhalation rates. The total methane mass balance of the artificial cow between inputs (produced and inhaled methane) and output (exhaled methane) was tested at methane production rates ranging from 0.0707L/min to 0.4368L/min. Methane was injected by mass flow controllers into a cylinder. It used an actuator to force a piston moving inside this cylinder to simulate exhaling and inhaling cycles. Exchanged air went in and out through separated inlet and outlet tubes connected to the cylinder. The breath frequency, air exchange volume of each exhaling-inhaling cycle, methane production rate and the temperature of exhaling gas could be adjusted between 15 to 40 times/min, 4 to 6 Liter, 0 to maximum 5 L/min and 35 to 37℃, respectively. In addition, an evaluation model was developed to analyse the concentration levels and patterns of the artificial cow at different settings. During the mass balance experiment the exhaled air in the outlet tube was sampled by an FTIR-analyser. The results showed that average methane concentration in exhaled gas changed from 381ppm to 2400ppm with different methane flow rates, and showed a mean of 0.5 ± 2.1% differences between measured and model calculated results. The FTIR measured outputs had a strong positive relationship with the mass flow controller defined inputs. Linear regression analysis resulted in the following equation: $M_{out}=M_{in}\times1.018-0.018$ (R^2=0.999, P<0.001). The total methane mass differed on average 1.8% between the mass flow controller defined inputs and the FTIR measured outputs. The system's methane input and output were almost equal. It is concluded that the artificial cow properly represented the methane exhalation of a cow, and that the system accurately controlled methane concentration and production. This system can be used as a reference source in dairy barns to develop and test practical methane measurement methods.

Keywords: methane, artificial cow, mass balance

1 Introduction

Efforts to mitigate methane emission from dairy cows are critical to reduce the dairy industry's contribution to the production of greenhouse gases and subsequently to global warming. Dairy cows have been identified as the major producer of methane emission as they account for 15% of the global methane emission budget (Lassey, 2007). On average, a dairy cow produces 250 to 400 gram methane per day (Bannink, van Schijndel, & Dijkstra, 2011).The release of methane by dairy cows also represents a loss of energy for the animal. Dairy cows lose 2-12% of their gross energy intake, or 5-14% of their digestible energy intake, in the methane production process (Blaxter & Clapperton, 1965; Johnson & Johnson, 1995). In short, mitigating methane emission from dairy cows will benefit not only the environment but also the cows themselves.

Mitigation of methane emission from dairy cows by nutritional and microbial manipulation has already been extensively studied (Boadi, Benchaar, Chiquette, & Massé, 2004). Recently, a wide interest has developed in reducing methane emission by breeding that is inexpensive and provides a long-term effect. Approximately 10~15% variations of methane emission between and within cows were reported in many studies (Grainger et al., 2007; Johnson & Johnson, 1995), which provide the potential to select dairy cows with low methane yield. To carry out the genetic selection directly in practice, methane production from individual cows must be accurately and precisely quantified first. Therefore, we need measurement technology capable of evaluating methane emission from a large number of cows in the barns.

To develop a methane measurement method, respiration calorimetric chamber is always considered as the reference method because of its accurate measurement results (Blaxter, Brockway, & Boyne, 1972; Derno, Elsner, Paetow, Scholze, & Schweigel, 2009). In this method a cow is kept in a small confined chamber for several days. During this time, methane produced from the cow is calculated as the difference between outgoing and incoming amounts of methane measured in the chamber. However, a small and restricted chamber can modify a cow's behaviour, reduce their feed intake and consequently influence methane production. Moreover, a respiration chamber is limited by the time it takes to train an animal for measurements, the number of animals available, and the large expense of building and maintaining a chamber. In short, a respiration chamber is a time-consuming and costly reference method to manage.

Given this, an artificial mechanical cow would be a good alternative reference method for the following three reasons. First, an artificial cow has known and controlled methane production rates so that different methane emission levels of cows can be created to calibrate and validate measurement methods. Second, it can automatically operate accurately and stable for a long period without adjustment or maintenance because it is a machine. Third, it can produce the same methane concentration patterns under different circumstances.

The aim of this study was to design, construct and validate an artificial reference cow with known methane production rates so that it can simulate methane concentration patterns of real cows. The paper will first describe the schematic and working principle of the artificial reference cow. Then the system's total methane mass balance between input and output will be evaluated in defined methane exhalation rates.

2 Materials and methods

2.1 The working principle of the artificial reference cow

The artificial reference cow (Figure 1) was designed and constructed to simulate a cow's methane production and exhalation procedures during respiration and eructation. The artificial cow consisted of an aluminium cylinder (40cm x 20cm, 12Lv) to provide the cow's tidal and residual volume during respiration. A rubber piston connected to an actuator was placed inside the cylinder. The actuator was operated by about 2bar compressed air and drove the piston horizontally up and down to simulate a cow's inhaling and exhaling processes. The characteristics of real cow's respiration, such as tidal volume and breath frequency vary between cows, which affect the methane exhalation pattern. The artificial cow's tidal volume and breath frequency were determined by the piston's movement time and length, which can be changed by the volume of the compressed air and the stroke length of the actuator. The actuator's stroke length could be adjusted between 0 and 20cm, which means the piston's movement could simulate 0~6L tidal volume. The compressed air volume could be adjusted by the valve connected to the actuator allowing the breath frequency to be simulated between 20 to 40 times per minute.

The artificial reference cow also contained two mass flow controllers (MFC, Bronkhorst high-tech B.V.). These controllers controlled the artificial reference cow's methane and carbon dioxide production rates. Two pure CH_4 and CO_2 cylinders were attached to the MFC to provide the gas source. A 20cm × 62cm silicon heating mat covered the cylinder's left side so that the gas inside was uniformly warmed to the temperature of the cow's breath. A 110cm-long plastic tube was connected to the right side of the cylinder's middle to simulate the cow's respiration tract. At the end of the tube, two 4cm-round openings mimicked the cow's nose. In addition, four methane sensors were located integrated with the artificial reference cow: one inside the cylinder, one in the nose and two on the side of the cylinder. Each methane sensor was set to sound an alarm when the methane concentration was too high because the controls failed or because the artificial reference cow was leaking methane.

The artificial reference cow was controlled by three separate operating systems: Labview, a Programmable Logic Controller (PLC) and a heating controller. Labview operated and processed the main functions of the artificial reference cow, including its piston movement time, CH_4 and CO_2 injecting strategies, gas temperature and the methane sensor's signals. The PLC and heating controller monitored the piston's movement signals and the gas temperature inside the cylinder. The artificial reference cow would shut down gas release if the piston did not move or if the gas temperature was higher than the alarm level.

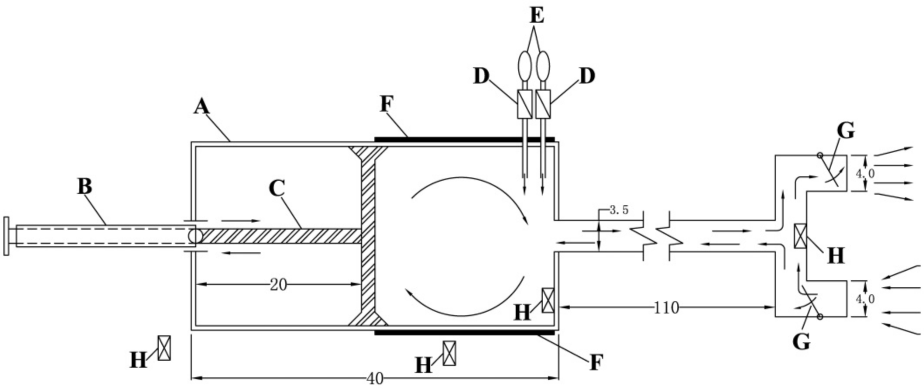

Figure 1. Schematic overview of the artificial reference cow: (A) a 40cm×20cm cylinder, (B) an actuator, (C) the stroke of the actuator that can move the piston between 0 and 20cm, (D) two mass flow controllers, (E) two pure CH_4 and CO_2 cylinders, (F) a 20cm × 62cm heating mat, (H) four methane sensors placed inside the cylinder, in the nose and on the side of the cylinder, (G) two one-way valves in the nose that were only installed and used for the total mass balance experiment.

2.2 Concentration pattern evaluation model

Based on the methane mass balance between input (injected by MFC and inhaled gas) and output (exhaled gas) of the artificial reference cow, an evaluation model was developed to assess the methane concentration pattern produced by the artificial reference cow:

$$\int_0^t dxdt = M + C * \int_0^t dVdt \tag{1}$$

where: x is methane mass accumulated in the cylinder in g/s; M is methane mass injected by MFC in g during time interval t; C is methane concentration in g/l, which is ambient methane concentration during inhaling or concentration inside the cylinder during exhaling; dVdt: gas exchange volume in the cylinder in l/s. The left side of the equation gives the methane mass accumulated in the cylinder. The right side of the equation includes two methane flows of the artificial reference cow: one is the methane injected from MFC and the other one is the methane exchanged during inhaling and exhaling.

The evaluation model can be used to analyze the methane concentration produced by the artificial reference cow in any time resolution. Due to the limitation of gas analysers' response time, methane concentration pattern changes in shorter time intervals than the response time cannot be detected by the gas analyzer. However, such fast changing pattern can be estimated by the evaluation model.

2.3 Methane mass balance of the artificial reference cow

The methane mass balance of the artificial reference cow is one critical factor to evaluate whether the artificial reference cow is working properly. The experimental design to evaluate this aspect is shown in Figure 2. Two one-way valves (Figure 1 & 2) were mounted to the nose of the artificial reference cow. One checking valve was open only during inhalation and the other one was open only during exhalation. Thus, the reference cow's inhalations and exhalations could be analyzed separately. A 3m tube with a 50mm diameter was fixed to the exhaling nose. The exhaled gas was continuously sampled and analyzed for the methane concentration by a portable multicomponent Fourier transform infrared spectroscopy (FTIR) gas analyzer. The FTIR included a gas analyzer (Gasmet DX 4000, Gasmet Technologies Oy, Helsinki, Finland), a portable sampling system, a heating line and a cooling device, which can effectively and accurately measure several gas concentrations simultaneously in two to three seconds. During the experiment, pure methane was injected with fixed flow rates controlled by MFC.

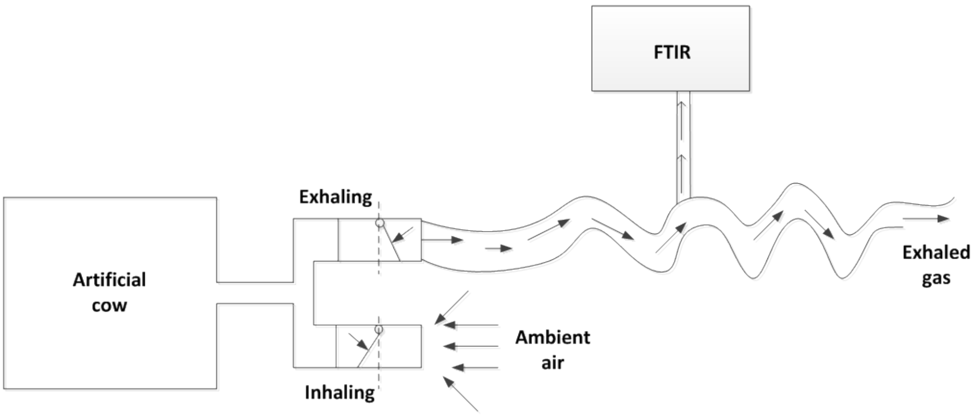

Figure 2. Schematic of the experiment setup to evaluate methane mass balance of the artificial reference cow.

The methane mass balance experiment was conducted in the lab with the artificial reference cow set at different methane injection rates. The methane's injection flow rate was tested at eight levels: 0.05L/min, 0.10L/min, 0.15L/min, 0.20L/min, 0.25L/min, 0.30L/min, 0.35L/min, 0.40L/min. Each level of methane flow rate was conducted for about 8 minutes and repeated four times. During the experiment, the tidal volume and breath frequency of the artificial reference cow were controlled at 6L and 30times per minute.

The methane injected by MFC and inhaled from the ambient environment was the artificial cow's methane input, and the exhaled air was the artificial cow's methane output. The input and output methane mass were calculated according to the following equations:

$M_{in} = (F \times T_e + BF \times T_e \times T_v \times C_a) \times \rho$ (2)
$M_{out} = C \times BF \times Te \times Tv \times \rho$ (3)
where: M_{in}, M_{out} were the artificial cow's methane input and output in g; F was the methane flow rate controlled by MFC in L/min; T_e was the measurement time in minutes; BF was the breath frequency in times per minute; T_v was the tidal volume in L; C_a was the methane concentration in ambient air, which was about 3ppm during the experiment period; ρ was the methane density in g/min; C was the methane concentration in the exhaled gas measured by FTIR in ppm. Therefore, the overall methane mass could be calculated as the expected input and measured output of the artificial reference cow. Input and output could be compared for each defined methane flow rate.

Data was analysed using Genstat software (version16). The relationship between the artificial reference cow's methane input and output was investigated with a linear regression model.

3 Results and discussions

3.1 Measured methane concentration versus predicted results

Methane concentrations measured in the exhaled gas and predicted by estimation model are shown in Table 1. Methane concentration measured in the exhaled gas varied from 376.9±8.4ppm to 2435.6±36.0ppm, which increased along with methane injecting flow rates. With more methane injected, methane concentration in the exhaled gas was higher. The measured methane concentration highly agreed with the values predicted from the estimation model. The difference of average methane concentration between measured and predicted values ranged from -2.2% to 2.9%. However, predicted methane concentration always had larger standard deviation compared to the measured methane concentration. The estimation model predicted the actual methane concentration during each exhalation, whereas measured exhaling gas mixed in the output tube before sampling by FTIR. Moreover, due to the response time of the FTIR, FTIR cannot detect actual methane concentration changes in less than the response time. Therefore, measured methane concentration was the average value of the actual methane concentration during each exhalation, which led to smaller standard deviation compared to the predicted values by the estimation model. In short, the artificial reference cow can precisely simulate different methane concentration levels in exhaled gas with defined methane flow rates.

Table 1. Measured methane concentration in the tube connected to the exhaled nose versus predicted methane concentration at the exhaled nose under different defined methane flow rates

Methane flow rate [L/min]	Measured in 8mins[ppm]		Predicted in 8mins[ppm]		Average difference[b] [%]
	Mean	SD[a]	Mean	SD[a]	
0.05	376.9	8.4	387.3	33.9	2.7
0.10	662.1	26.9	672.2	58.9	1.5
0.15	930.1	12.0	957.2	84.0	2.9
0.20	1218.8	19.4	1242.2	109.0	1.9
0.25	1519.8	20.4	1527.2	134.1	0.5
0.30	1825.2	26.2	1812.2	159.1	-0.7
0.35	2143.4	26.8	2097.2	184.2	-2.2
0.40	2435.6	36.0	2382.2	209.2	-2.2

[a]SD: Standard deviation; [b]Average difference: (Predicted – Measured)/Measured×100%;

3.2 Overall methane mass balance between input and output

The artificial cow operated satisfactorily in terms of producing predicted methane concentrations at defined methane flow rates. The overall methane mass balance between input and output was another crucial factor to evaluate the system's performance. The artificial reference cow's methane input and output at defined methane flow rates were calculated by the equation (2) and (3). The results of total 32 trials are displayed in Figure 3. During the experiment, overall methane mass at input was controlled from 0.14g to 1.69g. The output methane mass was strongly positively related to the input methane mass. In regression analysis, the fitted term with methane input and constant had a smaller standard error (0.0135 versus 0.0160) than with only methane input. Therefore, the best fitted model was expressed as: $M_{out}=M_{in}\times1.018-0.018$ ($R^2=0.999$, $P<0.001$). The model predicted that methane output would rise by 1.018 units with each additional methane input unit. Standardized residuals of the model were evenly distributed around the zero line as depicted in Figure 3.

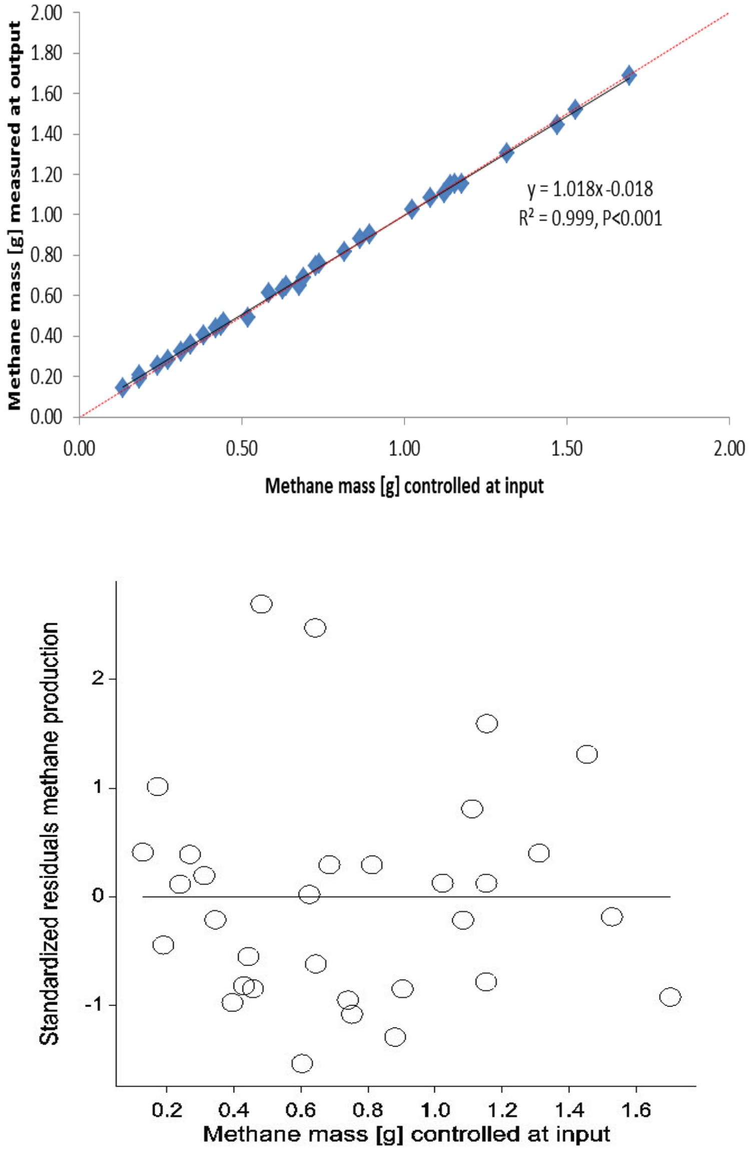

Figure 3. Methane mass [g] controlled at input versus measured at output (above) and standardized residuals (below) versus methane production [g] during each 8 minutes experiment; the dotted line (above) represents the line of equivalence.

The estimates of the parameters in the linear regression model are shown in Table 2. The fitted regression coefficient and constant had standard errors of 0.00498 and 0.00583, both with t probabilities smaller than 0.001 for deviating from 0, indicating that the linear relationship between the system's methane input and out was significant. Confidence intervals of M_{in}

and constant did not consist values of 1 and 0, indicating that the output methane mass was not absolutely equal to the input methane mass. The differences may be caused by the MFC on controlling the methane's injection flow rate and the FTIR on analyzing the methane concentration of the exhaled gas. According to the accuracy of the MFC, the controlled methane flow rate had ±0.5% deviation. Although the exhaled gas had been mixed first in the extra tube before being sampled by the FTIR, inhomogeneous exhaled gas could also affect the measurement results. Yet, with about 1.8% difference between methane input and output, it is demonstrated that the artificial reference cow could accurately control the methane mass production.

Table 2. Estimates of parameters in the linear regression model between the artificial reference cow's methane mass input and output

Parameter	Estimate	S.E.	t pr.	Lower 95%	Upper 95%
Constant	*-0.018*	*0.00498*	*<.001*	*-0.028*	*-0.008*
Regression coefficient	*1.018*	*0.00583*	*<.001*	*1.006*	*1.030*

4 Conclusions

It is concluded that the artificial cow properly represented the main methane production process in a cow, and that the system precisely controlled methane concentration and production. This system can be used as a reference source in dairy barn to develop practical methane measurement methods.

5 Acknowledgements

This study was funded by Wageningen UR Livestock Research, the Ministry of Economic Affairs (through KB Sustainable Agriculture) and the China Scholarship Council. The authors are thankful to Johan Ploegaert of Wageningen UR Livestock Research, Hennie van Dorland of Wageningen UR for constructing the artificial reference cow.

6 References

Bannink, A., van Schijndel, M. W., & Dijkstra, J. (2011). A model of enteric fermentation in dairy cows to estimate methane emission for the Dutch National Inventory Report using the IPCC Tier 3 approach. *Animal Feed Science and Technology, 166-167*.

Blaxter, K. L., & Clapperton, J. L. (1965). Prediction of the amount of methane produced by ruminants. *British Journal of Nutrition, 19(4)*, 511-522.

Blaxter, K. L., Brockway, J. M., & Boyne, A. W. (1972). A new method for estimating the heat production of animals. *Quarterly journal of experimental physiology and cognate medical sciences, 57(1)*, 60-72.

Boadi, D., Benchaar, C., Chiquette, J., & Massé, D. (2004). Mitigation strategies to reduce enteric methane emissions from dairy cows: Update review. *Canadian Journal of Animal Science, 84(3)*, 319-335.

Derno, M., Elsner, H. G., Paetow, E. A., Scholze, H., & Schweigel, M. (2009). Technical note: A new facility for continuous respiration measurements in lactating cows. *Journal of Dairy Science, 92(6)*, 2804-2808.

Grainger, C., Clarke, T., McGinn, S. M., Auldist, M. J., Beauchemin, K. A., Hannah, M. C., . . . Eckard, R. J. (2007). Methane Emissions from Dairy Cows Measured Using the

Sulfur Hexafluoride (SF6) Tracer and Chamber Techniques. *Journal of Dairy Science, 90(6)*, 2755-2766.

Johnson, K. A., & Johnson, D. E. (1995). Methane emissions from cattle. *Journal of animal science, 73(8)*, 2483-2492.

Lassey, K. R. (2007). Livestock methane emission: From the individual grazing animal through national inventories to the global methane cycle. *Agricultural and Forest Meteorology, 142(2-4)*, 120-132.