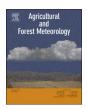
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Determination of methane yield in cattle fed tropical grasses as measured in open-circuit respiration chambers

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ABSTRACT

The objective of this study was to determine methane (CH₄) yield by crossbred cattle fed tropical grasses. A total of 66 individual determinations of dry matter intake (DMI) and 42 determinations of organic matter intake (OMI) in relation to the production of enteric CH₄ were carried out. Methane measurements in heifers were performed in open-circuit respiration chambers. Heifers (*Bos indicus* × *B. taurus*) with an average live weight of 288.5 \pm 55.7 kg fed tropical grasses as basal ration were used. An average intake of 8.22 and 7.80 kg of DM and OM per day were recorded. An average enteric CH₄ production of 88.0 g/heifer/day was determined. It was found that DMI and OMI in relation to CH₄ production have a coefficient of determination (R^2) of 0.73 and 0.70 respectively, to predict CH₄ emissions. It is concluded that the methane yield (18.07 g CH₄/kg DM intake) predicted by regressing DM intake against methane production represents a reliable value to be used for the estimation of enteric CH₄ inventories for cattle grazing in the tropical regions of Mexico.

1. Introduction

Livestock production contribute to the emissions of greenhouse gases (GHG) such as CO_2 , CH_4 and N_2O ; nonetheless, its precise contribution to global warming is still subject to uncertainties derived from methodologies employed for measurement. Usually IPCC (International Panel on Climate Change) methods are being employed for the estimation of inventories of GHG, however for the case of emissions of enteric methane, this has been challenged by different novel approaches (Charmley et al., 2016). The National Institute of Ecology and Climate Change (INECC) of Mexico estimated that emissions from the agricultural sector were 80,169.09 Gg of CO_2eq in 2013 which contributed to 12.0% of total GHG's emissions at national level (INECC, 2016). In tropical Mexico, cattle production systems (meat and milk) are based on grazed pastures containing high contents of neutral detergent fiber and low crude protein, which increase the synthesis of

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methane in the rumen (Moe and Tyrrell, 1979). In addition, about 110 million hectares (53% of national territory) is occupied by 33 million cattle (Servicio de información agroalimentaria y pesquera (SIAP, 2015). There is scarce information in Mexico on the annual inventories of CH₄ generated mainly by ruminal (enteric) fermentation. In this context, Castelán-Ortega et al. (2014) using mathematical models reported that the annual emission of CH₄ in Mexico is 2.02 Tg/ year. Other authors suggested that these values could reach 2.7 Tg/year due to an estimated increase in the cattle herd to 70 million by 2025. It is important to generate the baseline (emission factors and national inventories) of methane emissions in Mexico and to meet the national commitment to reduce GHG's emissions 50% by 2050 (DOF, 2012). Castelán-Ortega et al. (2014) estimated that emissions per cow/year in tropical regions is 116.8 kg and Rendón-Huerta et al. (2013) suggested similar emissions of 115 kg/cow/year. The differences in the estimated emission factors are due to the different sources of variation (measurement method, age of animals, sex, diets, climate and region), which results in predictions of CH₄ inventories in Mexico (Castelán-Ortega et al., 2014; Rendón-Huerta et al., 2013; Beltrán-Santoyo et al., 2016) that have been based on mathematical models containing several assumptions. Thus, it is important to generate equations that allow the prediction of enteric CH₄ emissions more accurately in cattle in a simpler and more reliable way (Blaxter and Clapperton, 1965; Kurihara et al., 1999; Kennedy and Charmley, 2012). Dry matter (DMI) and organic matter (OMI) intakes are the most widely used predictors (R^2) to generate robust enteric CH4 inventories and prediction equations derived from measurements performed in respiration chambers (Charmley et al., 2016; Stergiadis et al., 2016; Jonker et al., 2017). Data from methane emissions in cattle as measured in respiration chambers were collected in order to analyse the relationship between dry and organic matter intakes, and enteric methane emissions to generate an equation to predict this relation (g CH₄ emitted/kg DM or OM intake) in cattle fed tropical grasses.

2. Material and methods

2.1. Animal care

Animals were treated in accordance with guidelines and regulations for animal experimentation at the Faculty of Veterinary Medicine and Animal Science (FMVZ), University of Yucatan, Merida, Mexico.

2.2. Location

The experiment was carried out at the Laboratory of Climate Change and Livestock Production (LACCLIGA) of the Faculty of Veterinary Medicine and Animal Science (FMVZ), University of Yucatan (UADY) (21°15 N 83° 32 W) in Merida, Mexico. The region has a AW0 climate type (according to Köppen as modified by García (1981)) considered warm sub-humid with annual average rainfall of 984.4 mm and temperature of 26.8 °C (García, 1981).

2.3. Experimental animals

Crossbred heifers (Bos indicus × Bos taurus) (n = 33) with an average live weight (LW) of 288 ± 55.2 kg (μ ± SD) were used. Heifers were housed in metabolic crates located in a roofed building with concrete floor and devoid of walls. Before the experimental measurements, animals were dewormed with Ivermectin^{*} (1%, 1 mL per 50 kg LW) and ADE vitamins were administered (1 mL per 10 kg LW). Prior to the experiment, animals were also adapted to enter the respiration chambers for a period of ~3 weeks to reduce the effect of stress on voluntary DMI and behavior in and out of the chambers.

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Table 1

Chemical composition of tropical grasses and commercial concentrate.

Components	Pennisetum purpureum	Megathyrsus maximus cv. Mombasa	Commercial concentrate
OM (%)	93.27	94.26	93.0
CP (%)	6.74	4.40	16.0
NDF (%)	64.70	79.02	40.0
ADF (%)	39.40	53.88	23.0

OM: organic matter; CP: crude protein; NDF: neutral detergent fiber; ADF: acid detergent fiber.

2.4. Description of rations and voluntary dry matter intake

In order to determine methane yields (g CH₄/kg DM intake) cattle were fed a typical basal ration of Southeast Mexico, which consisted of chopped green pasture of *Pennisetum purpureum Schum* cv. Taiwan grass or mature hays of *Megathyrsus maximus* cv. Mombasa grasses. Data (methane production) was obtained from the control treatments (only tropical grass plus a supplement) of several experiments carried out in this laboratory between 2014–2017. In addition, animals were supplemented with commercial concentrate (ground corn, soybean meal and/or wheat bran; 20% of ration DM) (Lorgam^{*} 14% CP), mineral salt and had free access to fresh water. Grass and concentrate was offered separately. The chemical composition of the components of the diets is shown on Table 1.

Heifers were fed *ad libitum* allowing a minimum refusal of 15% of the dry matter (DM) of the ration offered the previous day. The ration supplied an estimated DM intake of 3% of LW. Feed refusals were weighed at 8:00 h the following day. Daily intake was determined as the difference between the amount offered and that rejected in a 24 h period.

2.5. Chemical analysis

Determination of dry matter (DM) of the forage samples offered and rejected was performed in a forced air oven at 55 °C for 48 h (until constant weight) method # 7.007 (AOAC, 1990). Organic matter (OM) was determined by incineration of samples in a muffle furnace at 550 °C for 6 h (AOAC, 1990, Method # 923.03).

2.6. Enteric methane emissions in respiration chambers

Two open-circuit respiration chambers (Fig. 1) of the appropriate volume (9.3 m³) to hold cattle of up to 450 kg were used for methane measurements (Canul-Solis et al., 2017). Respiration chambers dimensions were $2.10 \text{ m} \times 1.60 \text{ m} \times 3.10 \text{ m}$; height, width and length, respectively (Fig. 1). Front and rear doors were refrigerator-type and tightly closed the chambers. Chambers were provided with an air conditioning system to keep temperature (23 °C) within the thermoneutral zone of cattle, while relative humidity was kept at 55% by using a small dehumidifier. Chambers were equipped with a water bowl to supply fresh water at all times and a small fan was fitted to mix the air inside the enclosure. Respiration chambers allowed ambient air to enter into the enclosure and air was pulled through a hose with a dust filter at a rate of up to 450 L/min by the action of mass flow generators (Sable Systems International[®], USA). A multiplexer took a sample of chamber air and directed it to an air-drying column filled with Drierite (W.A. Hammond Drierite Company LTD[®], USA) as a desiccant agent before the air sample entered the infrared analyser (MA-10, Sable Systems International[®], USA). Data was transformed into liters of methane with the ExpeData software (Sable Systems International[®], USA), data was converted to grams for analysis (0.7612 g/L CH₄). Linearity of the methane analyzer was assessed by injecting four different concentrations (1,000; 2500; 5000; 7000 ppm) of methane in synthetic air (Praxair[®], Mexico). Calibrations to test the linearity of the methane analyser were

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