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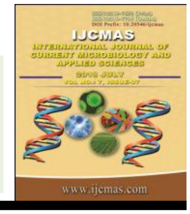

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Methane Emission by Ruminants and Its Measurement- A Review

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Ruminants accounts for up to one-third of the emitted methane worldwide. Over the decade, various methods have been developed with the purpose of measuring and estimating methane emissions from ruminants. These methods have advantages and disadvantages- none of them is perfect: Some are expensive, some cheaper; some suitable for grazing animals, some for housed ruminants; some can handle many animals, some only few. This all will ultimately affect our results and their interpretation. It is therefore, important to know the potential and limitations of each method. In this review, commonly used methods for measuring methane emission have been discussed along with their pros and cons. In addition, this review compares different estimation approaches that are commonly used.

Introduction

Livestock and especially ruminants accounts for up to one-third of the emitted methane worldwide (IPCC, 2007). Methane (CH_4) from agriculture arises primarily from enteric fermentation. Therefore, ruminants are responsible for enteric emissions of CH_4 during the normal digestive process. Among ruminants, cattle are the major contributor to greenhouse effect through CH_4 emission followed by sheep and goats (Charmley *et al.*, 2008). Fermentation CH_4 is the sum of enteric CH_4 and manure CH_4 (Mihina *et al.*, 2012). Enteric fermentation is a large source of methane, which has a global warming potential 28 times as strong as that of CO_2 on a 100-year time horizon (Stocker *et al.*, 2013). It has been proven to be the second most anthropogenic greenhouse gas (Turner *et al.*, 2016) that accounts for a significant energy loss to the ruminants i.e. about 2-15% of feed energy (Hess *et al.*, 2004). It accounts for 17-37 % of anthropogenic CH_4 (Sejian *et al.*, 2011) from ruminants and its emission depends on several factors like animal species, breed, rumen pH, acetate: propionate, dietary composition, etc.

The CH_4 emissions from the ruminants is measured as part of the studies related to ruminal fermentation, energy balance, evaluation of feed additives and most recently, to characterize and reduce the contribution of ruminants to the global CH_4 production. Methodologies for measuring CH_4 emissions range from animal respiration chambers to estimation by model techniques. However, several factors need to be considered in order to select the most appropriate technique like the cost, level of accuracy required and the scale and design of the experiments to be undertaken (Johnson *et al.*, 2000). The commonly used methods for measuring methane emission in ruminants have been discussed in this paper along with their pros and cons. In addition, comparison between them has been done (Table 1).

Respiration calorimeter

It is the classical standard for measuring ruminant CH_4 production. Respiration calorimetry techniques such as whole animal chambers, head boxes, or ventilated hoods and face masks have been effectively used to measure CH_4 emissions in ruminants. There are different designs of calorimeters (Blaxter, 1962), but the most common is "open circuit calorimeter". The basic principle behind "open-circuit indirect-respiration technique" is that outside air is circulated around the animal's head, mouth and nose and well-mixed inside air is collected (Mclean and Tobin, 1987) (Fig. 1). The animal is placed inside open-circuit respiration chamber for a period of several days, the inputs (feed, oxygen, CO_2) and outputs (excretion, oxygen, CO_2 and CH_4) are measured from the chamber. The chamber should be tightly sealed with a slight negative pressure inside. This ensures no net loss of CH_4 from the chamber. Internal ventilation fans efficiently mix the expired air and incoming air. An air pump removes all air from the space through flow meter and gas sensor analysis is done. Difference between outgoing and incoming amount of methane is methane emission. Its pros and cons have been mentioned in Table 2.

Sulphur hexafluoride (SF_6) Tracer Technique

This method is relatively new and was first described in 1993–1994. This method is primarily used to investigate energy efficacy in free ranging cattle (Okelly *et al.*, 1992). The basic principle is that CH_4 emission can be measured if the emission rate of a tracer gas from the rumen is known. For this purpose a non-toxic, physiologically inert (Johnson *et al.*, 1992), stable gas is needed. Additionally, the gas should mix with rumen air in the same way as methane. Therefore, SF_6 was chosen because it fulfills all the above criteria, is cheap, has an extremely low detection limit and is simple to analyze.

For individual animal measurement, a calibrated source of SF_6 is placed in the rumen before conducting an experiment. The source of SF_6 is a permeation tube, and the rate of release of SF_6 is controlled by a permeable Teflon membrane held in place by a porous stainless steel frit and a locking nut. The release rate of the gas permeation tube is calibrated at 39°C by regular weighing for a period, prior to its insertion into the rumen. Each test animal is fitted with a halter, which supports an inlet tube that is placed so that its opening is close to the nose (depicted in Fig. 2). As the vacuum in the sampling canister slowly dissipates, a steady sample of the air around the mouth and nose of the animal is taken. By varying the length and diameter of the capillary tube the duration of sampling may be regulated. The volumes of the canister is about 1.7 and 2.5 litres for sheep and cattle respectively, and the capillary system is designed to deliver half this volume during the collection period of 24 h. An identical apparatus needs to be placed each day to collect an integrated background air sample. After collection of a sample, the canister is pressurized with nitrogen, and CH_4 and SF_6 concentrations are determined by gas chromatography. Methane emission rate is calculated as:

$$QCH_4 = QSF_6 \times [CH_4]/ [SF_6]$$

Where QCH_4 is the emission rate of methane in g/day; QSF_6 is the known release rate (g/day) of SF_6 from the permeation tube; $[CH_4]$ and $[SF_6]$ are the measured concentrations in the canister.

Rumen Simulation Technique (RUSITEC)

In the RUSITEC, solid feeds are confined in nylon bags that are normally replaced by new bags once a day. The amount of ration is small (10-25 g DM/day/L of vessel), and the set points of the liquid dilution rate are also small (25%/h) as compared with the actual *in vivo* values (Bhatta *et al.*, 2007b). Its pros and cons have been mentioned in Table 2.

The gas produced from each fermenter is collected in polythene/rubber bags and the volume of gas is recorded using a dry gas meter. From the gas samples, the concentration of CH_4 is measured in gas chromatograph. The volume of CH_4 gas produced is calculated from the total volume of gas produced after 24 h in the fermenter. The CH_4 production should be converted to STP value (1 atm, 0°C) for comparison with CH_4 measured by other techniques.

Methane in ml (at STP) = (Methane ml) × (273/ (273+25)) × {(atmospheric pressure at the experiment) / (standard atmospheric pressure)}

In Vitro Gas Production Technique (IVGPT)

Menke *et al.*, (1979) developed a feed evaluation system using an *in vitro* gas measuring technique. The amount of gas produced during the incubation of feedstuffs with rumen liquor in 100 ml calibrated syringes is closely related to digestibility and therefore to the energetic feed value of feedstuffs for ruminants.

Therefore, on the basis of gas production, quality of feeds can be evaluated. The concentration of CH_4 (%) in the gas samples was determined by the gas chromatograph. Its pros and cons have been mentioned in Table. 2.

Methane models

During scientific trials using the total national emissions, calculation is not possible. Therefore, methane models have been developed to predict methane production based on existing data, such as animal characteristics (weight, age, and breed), feed characteristics (nutrient and energy content), intake data (dry matter or nutrients) or digested nutrients.

These models frequently uses data derived from experiments conducted with cattle in respiration chambers. The standard model usually used for calculating cattle methane emissions is issued by the IPCC. IPCC (2006) recommends the tier-2 method for estimating CH_4 emissions from enteric fermentation for those countries with large cattle populations. Average daily feed intake (in terms of GE content, MJ/d) and CH_4 conversion rates (Ym) are used to estimate CH_4 emissions in the tier-2 method. For dairy cattle, Ym is 6.5% ± 1% of GE intake and for intensively fed cattle (>90% concentrate), Ym is 3% of GE.

Table.1 Comparison between common methane emission techniques

Method parameter	Respiration Calorimetry	SF_6 technique	IVGPT
Suitability for Individual animals	Yes	Yes	No
Variation within animals	Yes	Yes	No
Variation between animals	Yes	Yes	No
Animal fixation	Yes	Yes	No
Effect of physical form of diet	Yes	Yes	No
Animals to carry equipment	No	Yes	No

Table.2 Pros and cons of methane measuring techniques in ruminants

Technique	Pros	Cons	References
Respiration Calorimeter	Accurate measurement of CH_4 from ruminal and hindgut fermentations	Not suitable for grazing ruminants, restriction of animal movement, construction and maintenance of the chambers is costly	Bhatta *et al.*, (2007a)
Ventilated Hood	Lower cost	Requires restrained and trained animal, hindgut CH_4 can't be measured	Bhatta *et al.*, (2007a)
Facemasks	Simplicity and lower cost, suitable for grazing animals	Underestimates heat production, requires animal cooperation and eliminates their ability to eat and drink	Liang *et al.*, (1989)
Sulphur Hexafluoride (SF_6) Tracer Technique	Allow the animal to move and graze freely, suitable for individual free ranging animals on pasture	Itself it is a GHG with a GWP 23,900 times that of CO_2, presence of residue of SF_6 in meat and milk from farm animals, It is necessary to train the animal to wear a halter and collection canister, less precise, equipment failure	Machmuller and Hegarty (2005)
Rumen Simulation Technique (RUSITEC)	Constructional simplicity and operational easiness, More number of fermenters can be used at a time	Difficulty in obtaining a uniform sample, Protozoa numbers in the effluent gradually decreases as the incubation proceeds and settles at around 3,000/ml after the 8th day for 3.0 %/h dilution rate	Bhatta *et al.*, (2006, 2007a)
***In Vitro* Gas Production Technique**	Possible to screen many different feedstuffs fast and cheaply, easy to control fermentation conditions like pH, animal to animal variation- avoided	It only simulates ruminal fermentation of feed, not emissions and digestibility by the entire animal	Czerkawski and Breckenridge (1977)

Fig.1 Open-circuit calorimeter

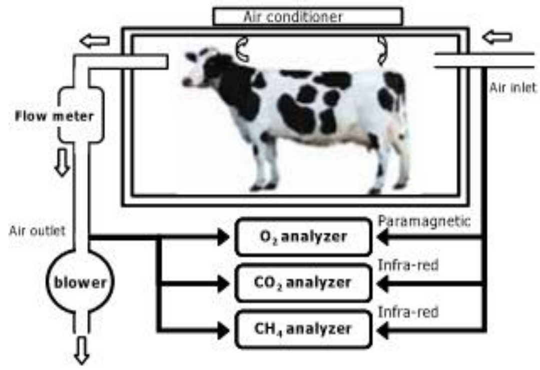
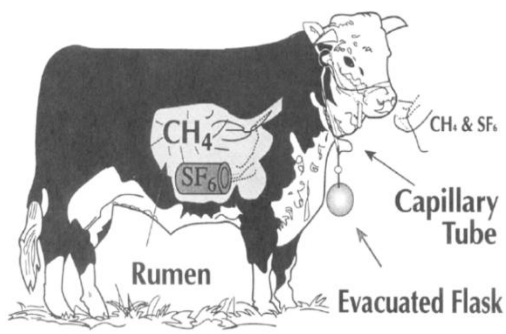


Fig.2 Illustration of SF6 tracer technique (Johnson *et al.*, 1994)



There are 2 types of models:

1) **Empirical (statistical) model**: Relates nutrient intake to CH_4 output directly.

Moe and Tyrrell model (2006): It is an empirical model. It uses data from cattle and relates intake of carbohydrate fractions to CH_4 production

Methane (MJ/d) = 3.41 + 0.51 NFC + 1.74 HC + 2.65 C

where, NFC= non-fibre carbohydrate (kg/d); HC= Hemicellulose (kg/d); and C= cellulose (kg/d).

Dynamic mechanistic model: Attempts to simulate methane emission based on mathematical description of ruminal fermentation biochemistry.

MOLLY Model: It is dynamic mechanistic model of nutrient utilization in cattle. Ruminal CH_4 production was predicted to be based on hydrogen balance.

Advantages of mechanistic model

They are diet-specific, average values are used to emphasize the magnitude of differences in CH_4 emissions when using apparently similar Ym values to estimate national inventory.

Mitigation options implemented at a farm or national level can be assessed for their effectiveness (empirical models- only reductions in emissions can be assessed).

In conclusion, many good methods for measuring CH_4 emissions from ruminants are already in use and new ones are being developed. All these methods require careful consideration before application. In this context, a thorough knowledge of the pros and cons of experimental methods is extremely important. The choice of a method depends on the accuracy. Increased understanding and improved quantification of CH_4 production has implications not only for global environmental protection but also for efficient animal production.

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