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Review

METHODS OF METHANE MEASUREMENT IN RUMINANTS

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ABSTRACT

This review is devoted to methodology, which can help direct and indirect measurement of methane emissions. This paper will be useful for expanding the knowledge base of researchers, farm planners, and policymakers as they work to develop and maintain sustainable environment conditions for farming systems in Slovakia. The following methods like respiration chamber, SF_6 technique, alternative methods, micrometeorological methods, proxy methods, in vitro gas production technique, and models for predicting methane production are described. Above mentioned methods are compared and their advantages and disadvantages are enlisted.

Key words: methane; emission; method

INTRODUCTION

Animals contribute to global warming by releasing of greenhouse gas emissions. The major greenhouse gas produced from enteric fermentation of ruminants during the normal digestive process is methane (CH_4). Fermentation CH_4 is the sum of enteric CH_4 and manure CH_4 (Veysset *et al*., 2010; Mihina *et al*., 2012). Enteric fermentation from livestock is a large source of methane, which has a global warming potential 23 times that of carbon dioxide (Bhatta *et al*., 2007; Loh *et al*., 2008). Methane from agriculture arises primarily from enteric fermentation; therefore, ruminants (especially beef and dairy cattle) are mainly responsible for enteric emissions of CH_4 (Kebreab *et al*., 2006). Enteric CH_4 from ruminant livestock accounts for 17-37 % of anthropogenic CH_4 (Beauchemin *et al*., 2010; Sejian *et al*., 2011).

Methodologies for measuring CH_4 emissions range from animal respiration chambers to estimation of model techniques. While chambers provide a simple measurement technique that is ideal for testing treatment differences there are disadvantages, too as only a small area or number of animals may be studied (McGinn *et al*., 2008; van Haarlem van *et al*., 2008; Flesch *et al*. 2007). The latest technology developed to estimate CH_4 more accurately is the micrometeorological mass difference technique (Harper *et al*., 1999; Sejian *et al*., 2011).

Emission of CH_4 in ruminants differs depending on factors like animal species, breed, pH of rumen fluid, ratio of acetate: propionate, methanogen population, composition of diet and amount of concentrate fed. Among the ruminant animals, cattle contribute the most towards the greenhouse effect through methane emission followed by sheep, and goats, respectively (Charmley *et al*., 2008; Bhatta *et al*., 2008).

The purpose of the current study was to describe new methods for direct and indirect measurement of methane emissions.

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Respiration chamber

The principle of the chamber is to collect exhaled CH_4 emissions from all sources of enteric fermentation (mouth, nostrils, and rectum) from the animal and to measure the concentration. Chambers are divided into two types, the closed-circuit and the open-circuit. The closed-circuit system is almost not used and preferred are open-circuit chambers. An air pump removes all air from the space through a flow meter and gas sensors in the open-circuit system. Each chamber is fitted with internal ventilation fans for efficient mixing of expired gases and incoming air. Air inlet is located at the front and an air outlet at the back. Fresh air to chamber is directly drawn from outside or through an air conditioning system to control humidity and temperature. The chamber is equipped with sensors for measuring relative humidity, temperature and barometric pressure. These allow air flow data to be adjusted for dry, standard temperature and pressure conditions. Outlet gas from each chamber is continuously sampled for analysis. Air flow is ducted via flexible polyurethane hoses. Air circulation is provided throughout the chambers at continuous but adjustable flow rates (usually 100-250 L.min^{-1}) (Chagunda *et al*., 2011; Storm *et al*., 2012).

Methane emission is calculated from flow and gas concentration in inlet and outlet air from the chamber. The difference between the outgoing and incoming amount of methane expresses the methane emission (Muñoz *et al*., 2012). Outlet gas from each chamber is continuously sampled for analysis. A multigas analyser with capability for measurement of methane and other gasses as carbon dioxide, and oxygen is used for the gas analyses (Pinares-Patino *et al*., 2008a; Chagunda *et al.*, 2011).

SF_6 tracer

The principle is that methane emission can be measured if the emission rate of a tracer (non-toxic, physiologically inert, stabile) gas from the rumen is known (Hegarty, 2013). SF_6 was selected from many comparisons, because it has an extremely low detection limit (Muñoz *et al.*, 2012). The gas should mix with rumen air in the same way as methane. The SF_6 technique involves the use of a SF_6 permeation tube dosed into the reticulo-rumen (Lassey *et al*., 2001). The calculation of daily CH_4 emission is based on the CH_4:SF_6 ratio of concentrations (adjusted for background concentrations) and the specific pre-calibrated permeation rate of SF_6 from the particular permeation tube deployed in the animal.

SF_6 is filled into small permeation tubes. The rate of diffusion of SF_6 out of the permeation tubes is measured by placing them in a 39°C water bath and measuring the daily weight loss until it is stable. The permeation tube containing ultra-pure SF_6 is placed in the rumen of an animal before the experimental period (Martin *et al*., 2008). The sampling apparatus consists of a collection canister, a halter and capillary tubing. A representative of breath gas sample, containing respired and eructated gas is collected through a capillary tube placed at the nose of the animal, fitted to a halter, or behind the head and connected with the evacuated canister (approximately 2.5 L); the tubing regulates the sampling rate for 24 hours (Lassey *et al*., 2001). This strategy requires two suites of canisters (the one removed became free once the collected samples were transferred to the analysis laboratory) (Bárbaro *et al*., 2008). The concentration of SF_6 and CH_4 in the canister is determined then by gas chromatography. The methane emission is calculated from the release rate of SF_6 and concentration of SF_6 and CH_4 in the containers in excess of background level (Storm *et al*., 2012).

Pinares-Patińo, Clark (2008) and Laubach *et al*. (2008) recommended the use of SF_6 method in grazing cattle involving large herds. The tracer technique is now widely used in New Zealand and many other countries for CH_4 emission measurements on grazing and pen-fed cattle, sheep, deer and alpacas (Pinares-Patińo *et al*., 2008b). CH_4 emission estimates SF_6 method revealed slightly lower (by 5-10 %) than the respiration chamber measured values. However, other studies with cattle using hoods or respiration chambers (Grainger *et al*., 2007) reported SF_6 tracer estimates slightly higher (by 1-2 %) than calorimetric estimates.

Alternative methods

More applications of alternative methods are combined with milking and feeding. The animals entering in automatic milking or feeding system are recognized and concentrations of CH_4 and CO_2 are measured. Air is continuously pumped through the equipment to quantify flow and thereby CH_4 and CO_2 emitted during milking and feeding.

Garnsworthy *et al*. (2012a) developed a novel technique based on sampling air released by eructation during milking. Methane analyzers are installed in automatic milking stations. Belching frequency and methane released per eructation are used to estimate methane emission rate. Air is sampled continuously from the feed mangers in the milking stations at 1 L.min^{-1} via an 8-mm diameter polyethylene tube, approximately 3 m in length, connected to the gas inlet port of the infrared methane analyzer with a range of 0 to 10.000 mg.kg^{-1}.

The same authors (Garnsworthy *et al.*, 2012b) recorded methane emissions of cows during milking using methane analyzers installed in automatic milking stations, modified as respiration chamber. Methane concentrations in air released by eructation are measured continuously at each milking and eructation data are used to calculate individual daily means for methane emission rate during milking. Air blows through the instrument by the pump between the gas inlet port and analyzer. Air is sampled continuously during the stay in the milking stations via a polyethylene tube, connected to the gas inlet port of analyzer. The port for the exhaust air from the analyzer is vented into the space at least 3 m from any sampling point.

Hegarty (2013) describes the device patented in USA called Emission monitoring unit, which measures emissions from individual cattle repeatedly over short timed periods whenever they visit the unit to consume a delivered mixture. Air is continuously drawn into the space where cattle received feed, and CH_4 and CO_2 flux are calculated continuously by multiplying the CH_4 or CO_2 concentration by the flow rate of air.

Other methods under development include the micrometeorological technique, combined feeder and CH_4 analyzer. An additional method for estimating methane emissions from livestock is based on the use of CO_2 as a tracer gas. Instead of using externally some gas, the naturally emitted CO_2 is used to quantify CH_4 emission (Madsen *et al.*, 2010). The exhaled air contains both the gases CO_2 and CH_4 (Laubach *et al.*, 2004).

The calculations are the similar as for the SF_6 tracer technique (just replacing SF_6 with CO_2). Corrections can be made for growing and lactating animals. The CO_2 method can be used to quantify methane production under different circumstances, for example from a dairy cow's barn and individual estimates for cows visiting an automated milking system (Storm *et al.*, 2012). Lassen *et al.* (2012) recorded individual methane (CH_4) and CO_2 production repeatedly on high number of dairy cows during milking also in an automatic milking system. They used a portable air sampler and analyzer unit based on transform infrared detection. The ratio between CH_4 and CO_2 was used as a derived measure with the idea of using CO_2 in breath as a tracer gas to quantify the production of methane. The repeatability was sufficient. The results of their study suggested that the CH_4 to CO_2 ratio measured using the non-invasive method is suitable and may be useful in both management and genetic evaluations. The instruments combined with automatic milking system may be useful to generate large data for genetic evaluation of CH_4 production in dairy cattle.

Micrometeorological methods

Micrometeorological methods are defined as measuring fluxes of gas in the free atmosphere and relating these fluxes to animal emissions. The methods are based on measurements of wind velocity and methane concentration, but the number of measuring points and the theories used to calculate emission rates differ between methods. The external tracer ratio technique can be used, where a tracer gas is released in the paddock or barn, and the concentrations of tracer and methane are measured in the surroundings (Harper *et al.*, 2011). This category of methods also includes the technique of mass balance in enclosed barns, where ventilation rate and concentrations in inlet and outlet are used to estimate the emission. While it is relatively easy to estimate emission rates from mechanically ventilated closed barns, naturally-ventilated buildings are problematic because of difficulties with measuring air exchange rates (Derno *et al.*, 2009). These types of buildings are commonly used for cattle since they are not especially susceptible to draughts and temperature changes and no extra heating is required. Air exchange rates in these buildings depend on the temperature gradient, temperature humidity index, and the air velocity. In this case, the release rates of harmful gases may also depend on external and uncontrollable parameters such as wind speed and the other parameters of outside environment. This method is particularly important in the current period; the present trend in milk production in Europe is to change to systems with loose housing in naturally-ventilated buildings (Ngwabie *et al.*, 2009).

Bjorneberg *et al.* (2009) used an open-path spectrometer operating in the monostatic mode for measuring methane. In this instrument, radiation from an incandescent silicon carbide source is collimated and passed into an interferometer. The exit ray from the interferometer leads onto an external beam splitter, so half the radiation is conducted into a 250 mm telescope that expands the beam due to magnification of its collimation. The diameter of the expanded beam at a distance of 50 m from the telescope is less than 400 mm. A cube-corner retro reflector is mounted at an appropriate distance from the telescope (usually between 150 and 250 m) and is aligned so that the reflected beam is returned to the telescope. The telescope reduces the beam back to a diameter of about 40 mm. The beam is driven from the telescope to the external beam splitter, which passes the beam to a cooled mercury cadmium telluride detector. Interferograms are measured at 70 s intervals. Quantitative determinations of CH_4 concentrations (also NH_3 and N_2O) are performed by partial least squares regression of the open-path spectra (Bjorneberg *et al.*, 2009).

A significant improvement in methane measurement accuracy is contributed by micrometeorological techniques which allow accurate emission estimates from agricultural sources via a dispersion technique (also called inverse dispersion technique) (Flesch *et al.*, 2005). This method has the advantages, which include non-interference, and the ability to incorporate the measurement footprint over larger areas. Inverse-dispersion methods have been used with success in several studies of feedlot gas emissions (Flesch *et al.*, 2007; Loh *et al.*, 2008; McGinn *et al.*, 2011). However, there are several limitations to using inverse dispersion methods including wind conditions and the need for source homogeneity (van Haarlem van *et al*., 2008).

Lagrangian Stochastic (bLS) method, belonging to category of dispersion techniques (but also in the category of micrometeorological techniques), is usually used in conjunction with global positioning system information from individual animals, to evaluate CH_4 emissions from pens of cattle (Laubach *et al*., 2005). CH_4 concentration is measured using an open-path laser. Each laser path is located at a height of 1.5 m about 1 to 1.5 m outside the perimeter of the pens (McGinn *et al*., 2009). The gas dispersion model contains vertical concentration profiles (Laubach *et al.*, 2008).

Methane emissions from grazing cattle are determined in a field experiment using paddock-scale (also belonging to micrometeorological) methods. The paddock-scale methods exploit how the gas, once emitted from the cattle, is transported and dispersed by the wind. Therefore, the emission rate may be calculated from measurements of wind speed, wind direction and turbulence, as well as CH_4 concentration upwind and downwind. The paddock-scale methods include a mass-budget approach, flux-gradient method and gas dispersion model. Accuracy is dependent on certain conditions, particularly whether the place is usually windy and free of obstructions that alter the turbulent airflow (Laubach *et al*., 2008).

Loh *et al*. (2008) applied open path spectroscopic concentration measurements and a bLs dispersion model for evaluation of methane and total greenhouse gasses in situ from feedlot beef production for the first time. Their results are consistent with other studies using a similar approach to measure emissions on a farm scale.

Proxy methods

Proxy methods were developed with the purpose of examining many animals at a same time without complex and expensive equipment. Close relationship of methane emissions with parameters that can be measured in easily obtainable from samples of milk or feces is used (Dehareng *et al*., 2012). Usually, the fatty acid profiles of milk are examined for correlations with methane production of the cows. The principle is that some fatty acids or fats in the milk or feces are correlated with either the feed composition or the amount of methanogens in the rumen (Vlaeminck *et al*., 2006; Chilliard *et al*., 2009).

The two challenges in using short-term breath measures as a proxy for measures of emissions are collecting data for an adequate period to provide a repeatable estimate of emission rate and scaling up from a short-term emission rate to methane production for whole day. These efforts resulting from the fact that the measurement is not entirely reliable and that a short term enteric methane emission measurement is not identical to a measure of daily methane production made in a respiration chamber.

Use of spectometry to predict the CH_4 emission of dairy cows has got high potential, too. (Dehareng *et al*., 2012) investigated the feasibility to prognosticate CH_4 emissions using milk mid infrared spectra. The experiments aimed to induce a large variation in CH_4 emission by feeding different diets (fresh grass and sugar beet pulp; maize silage and hay; grass and corn silage with cracked corn, soybean meal and dried pulp). Milk sample of 50 ml was collected from each cow and analyzed by spectrometry. Results suggest the feasibility of direct CH_4 prediction from milk mid infrared spectra. This alternative method could be useful to predict the CH_4 emissions at farm level or at the regional scale and it also could be used to identify cows with low CH_4 emission.

***In Vitro* gas method**

The gas measuring technique has been widely used for evaluation of nutritive value of feeds. More recently, the increased interest in the efficient utilization of roughage diets has led to an increase in the use of this technique due to the advantage in studying fermentation kinetics. Gas measurement provides a useful data on digestion kinetics of both soluble and insoluble fractions of feedstuffs (France *et al.*, 2000). This method has been modified for methane creation (Navarro-Villa *et al*., 2011; Storm *et al*., 2012).

The principle is to ferment feed under controlled laboratory conditions by natural rumen microbes. Feedstuffs are incubated at 39°C with a mixture of rumen fluid, buffer and minerals for a certain time period. The amount of total gas produced during incubation is measured and its composition analyzed, to obtain data on the *in vitro* production of methane. The method requires access to fresh rumen fluid, which is typically obtained from fistulated cows or other ruminants. The calculations are the same as for the CO_2 tracer technique.

Pellikaan *et al*. (2011) showed the gas production equipment which offers the possibility to determine total gas production, as a measure of organic mater fermentation, and methane synthesis simultaneously. With this system the maximum level of total gas production and methane synthesis can be determined, as well as the kinetics of synthesis. A fast screening of feedstuffs and additives for methane synthesis and total gas production is possible.

Models for predicting methane production

In many cases of scientific trials using the total national emissions calculation is not possible. Therefore there is an interest in being able to predict methane production using models based on existing data, such as animal characteristics (weight, age, breed), feed characteristics (nutrient and energy content), intake data (dry matter or nutrients) or digested nutrients. Such models often use data derived from experiments conducted with cattle in respiration chambers, but not techniques for measuring methane which were applied in recent years. Tremendous progress has been made in the field of designing simulation models for predicting CH_4 emissions, and the latest integrated farm system models offer greater scope to accurately predict greenhouse gas emissions with the incorporation of climatic and management information (Ellis *et al*., 2009; Sejian *et al*., 2011). Dry matter intake (DMI), metabolizable energy intake, neutral detergent fibre, acid detergent fibre, ether extract, lignin, and forage proportions were considered in the development of models to predict CH_4 emissions (Ellis *et al*., 2007).

Majority of methane models were developed from measurements obtained in respiration chambers. Some models require the proportion of roughage in the ration, while the other models require digested amounts of different nutrients. Total CH_4 production (L/d) in the cattle data set has been closely related to dry matter intake. Ramin and Huhtanen (2013) concluded that feed intake is the main determinant of total CH_4 production and that gross energy intake is negatively related to feeding level and dietary fat concentration and positively to diet digestibility, whereas dietary carbohydrate composition has only minor effects. CH_4 production was positively related to diet digestibility and negatively related to dietary fat concentration, whereas dietary carbohydrate composition had only minor effects. When authors expressed as a proportion of gross energy intake, CH_4 production was negatively related to feeding level and dietary fat concentration and positively related to diet digestibility and dietary concentrations of non-fibre carbohydrate and neutral detergent fibre.

A comparison of the above mentioned models leads to large differences in the estimates of methane emission. The model estimates are also associated with errors. The best equations developed by Ellis *et al*. (2007) for beef cattle, dairy cattle, and cattle in general had prediction errors of 14.4, 20.6 and 28.2 %, respectively. When models were evaluated with independent datasets, the prediction errors were increased.

The results of Ramin and Huhtanen (2013) indicate that CH_4 production can be predicted accurately from a set of variables that are available at the time of prediction. Equations predicting CH_4 production per unit of feed intake (gross energy or dry matter) are biologically more valid, and therefore it is recommended that CH_4 production is predicted as intake of gross energy (GE) or dry matter (DM) × production per unit (MJ of GE or kg of DM) of intake.

Methods of choice for estimating enteric methane emission depend on aim, equipment, knowledge, time and money available, but interpretation of results obtained with a given method can be improved if knowledge about the disadvantages and advantages are used in the planning of experiments (Ramin and Huhtanen, 2013). The prediction models should use to predict emissions for each strategy (Legesse *et al.*, 2011; Aljaloud *et al*., 2011; Kebreab *et al*., 2006, 2008).

An inverse dispersion model was utilized to calculate CH_4 emissions from a commercial cattle feedlot and an adjacent runoff retention pond. The feedlot measurements were collected within the interior of the feedlot enabling a near continuous emissions record over the 12 d of the study period (van Haarlem *et al*., 2008).

There have been several attempts to formulate mathematical models to predict CH_4 emissions from cattle. The models can be classified into 2 principal groups: empirical (statistical) models that relate nutrient intake to CH_4 output directly and dynamic mechanistic models that attempt to simulate CH_4 emissions based on a mathematical description of ruminal fermentation biochemistry (Kebreab *et al*., 2008; Alemu *et al*., 2011). A synthesis of the available literature suggests that the mechanistic models are superior to empirical models in accurately predicting the CH_4 emission from dairy farms. The latest development in prediction model is the integrated farm system model which is a process-based whole-farm simulation technique (Sejian *et al*., 2011).

The model proposed by Moe and Tyrrell (cit. Kebreab *et al*., 2006) is an empirical one developed using data from cattle, and the model relates intake of carbohydrate fractions to CH_4 production as follows: 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). In cases in which NFC values were not available, it was calculated as NFC = 100 − (CP + ether extract + ash + NDF), where CP = crude protein and NDF = neutral detergent fibre.

MOLLY model is a dynamic mechanistic model of nutrient utilization in cattle. Ruminal CH_4 production was predicted based on hydrogen balance. Excess hydrogen produced during fermentation of carbohydrates and protein to lipogenic volatile fatty acids (acetate and butyrate) is partitioned between use for microbial growth, biohydrogenation of unsaturated fatty acids, and production of glucogenic volatile fatty acids (propionate and valerate). The assumption is made that the remaining hydrogen is used solely and completely for methanogenesis (Kebreab *et al*., 2004).

The rumen model of Dijkstra *et al.* (cit. Kebreab *et al*., 2006) is the basis for the mechanistic model used in the present evaluation. The model is based on a series of dynamic, deterministic, and nonlinear differential equations. Kebreab *et al.* (2004) incorporated the rumen model to a whole animal model that included nitrogen and phosphorus utilization. Bannink *et al.* (2011) developed a new stoichiometry for fermentation within the rumen based entirely on experimental observations with lactating dairy cows; therefore, model COWPOLL was modified to accommodate these stoichiometric coefficients. One of the fundamental differences in estimating CH_4 emissions between MOLLY and COWPOLL is the representation of microbes in the rumen and the coefficients of fermentation for transformation of substrate to volatile fatty acids. The MOLLY model uses 1 group of microbes, whereas COWPOLL separates the microbial community into 3 groups: amylolytic, cellulolytic bacteria, and protozoa (Kebreab *et al*., 2008).

Charmley *et al*. (2008) described a modelling approach that estimates cattle methane emissions for various bioregions. The approach incorporates a metabolizable energy based model of animal production linked to a property herd economic model. This provides a flexible tool to evaluate animal and property herd dynamics on regional methane yields and live weight productivity, as well as to assess financial impacts. The model predicts that an important determinant of methane output per unit of product is reduced days to market. Reduced days to market may be achieved through a range of energy supplementation and marketing strategies. The modelling framework can be applied to a wide range of production, management and marketing scenarios to generate information on possible changes in methane emissions and financial gross margins. While these changes can be quantified, the output should be considered in light of the data deficiencies (Charmley *et al.*, 2008).

Many governments have implemented policies to reduce greenhouse gas emissions from agriculture and significant efforts are now being directed towards developing animal husbandry methods that lower enteric CH_4 emissions (Beauchemin *et al*., 2010). To adequately assess greenhouse gas mitigation strategies, it is necessary to use a whole system modelling approach (Beauchemin *et al*., 2010).

Three primary areas require refinement and relate to a better understanding of the forage base that makes up the major component of the diet. They include estimation of diet quality under selective grazing conditions; estimation of dry matter intake under heterogeneous grazing conditions; and precision of predicting methane yield from cattle grazing forages (Charmley *et al*., 2008).

Mathematical models allow us to predict CH_4 production from cattle without undertaking extensive and costly experiments. The models used can be classified as either statistical models, which relate nutrient intake to CH_4 production directly, or dynamic mechanistic models, which estimate CH_4 production using mathematical descriptions of rumen fermentation biochemistry (Kebreab *et al*., 2004, 2006). Although many statistical models have been fairly successful in predicting CH_4 production, many have inputs that are not commonly measured and some may have difficulty predicting CH_4 production outside the range of values on which they were developed. These problems may be addressed by using commonly measured equation input variables and by developing models on expansive data sets compiled from multiple sources (Ellis *et al*., 2007).

Advantages and inefficiencies of methods

Respiration chambers are regarded as the standard method for estimation of CH_4 methane emission from ruminants, because the environment can be controlled and the reliability and stability of instruments can be measured. However, results obtained in chambers cannot be extrapolated to loose housing animals, nor on pasture. This method is extremely slow and expensive (Hegarty, 2012), requires trained animals, restricted animal movement, causes stress, and have a high labour input (Pinares-Patińo, Clark, 2008). Respiration chambers are not used for determining methane production on farm.

The SF_6 method can be used to investigate nearly all aspects of feeding and nutrition, effect of chemical and physical composition, restricted or *ad libitum* feeding, different additives and grazing. However, using the method for investigation of dynamics of methane emission may be problematic. The following cons are maintaining a constant release rate from permeation tubes, effect of release rate upon emission rate of methane, background level determination, inconsistency between CH_4 measurements determined in chambers and with SF_6 (Storm *et al*., 2012; Hegarty, 2013). The SF_6 method gives more variable results of methane emission than chamber measurements. The method is the only available method for measuring individual free ranging animals on pasture (Muñoz *et al*., 2012). The number of animals is limited to 30 (Laubach *et al*., 2008). The CO_2 technique is a newly developed approach for estimation of methane emissions from ruminants. It can be used under different conditions on large numbers of animals or for the overall estimation of herd emissions. However, this method is less precise than the respiration chamber methods.

The micrometeorological methods are still new and further development and documentation on reliability is needed, but the methods are valuable in evaluating whole dairy systems and interactions between animals and landscape. Unfortunately, all these methods are influenced by instabilities like non-steady state wind or movement of point-emission sources (McGinn *et al*., 2008). It is also difficult to relate the CH_4 production to feed intake for grazing animals.

A disadvantage of *In Vitro* gas production technique is that it only simulates the ruminal fermentation of feed, not emissions and digestibility by the entire animal. Furthermore, under normal conditions it does not include long-term adaptation of the ruminal microorganisms to the tested feedstuffs. During live animal experiments it is usually a practice to have adaptation periods to new feeds of at least 14 days and animals' output is not considered stabile in this method (Pellikaan *et al*., 2011). Results should therefore always be interpreted with care (Storm *et al.*, 2012). Fortunately, the method can easily be applied to many animals making it possible to reduce the standard error of means from experiments. It is possible to determine *in vitro* degradation of the feedstuffs and find if the reduction in methane production is at the cost of total feed degradation. Screening large amounts of feeds and additives is the best application of the *in vitro* method. This method has a large capacity, making it possible to test many different combinations of feedstuffs.

The mathematical models are essential for estimating national or global emissions. They are easy to apply and will give estimates of the average emission of the unit in question. The models are based on experimental data and as such are limited in their application. However, a model based on respiration chamber experiments can therefore not be directly applied to free ranging cattle. Also, our understanding of ruminal digestion is not yet complete. Therefore a continuous need exists for more data to increase our knowledge of this complex system.

CONCLUSION

Many suitable methods for CH_4 measuring are already in use and new ones are being developed. Some, however, are only useful for a particular environment. It is extremely important to compare several methods for accurate assessment. Further research is needed to better understand the CH_4 measurement and evaluation in progressed managements.

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