

Overview of nutritional strategies to lower enteric methane emissions in ruminants

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Abstract

Since ruminants are capable of utilizing fibrous feeds not digested by mono-gastrics, they represent a valuable natural resource for meeting future increases in global food supply. Ruminants have both local (nitrogen and phosphorus pollutions) and global (greenhouse gases, GHG) environmental footprints. It is estimated that the livestock sector is responsible for 18% of global anthropogenic GHG emissions. Losses of methane represent 30 to 50% of total GHG from livestock production, with the contribution from ruminants accounting for about 80%. Due to the concerns of increases in GHG emissions into the environment and potential effects on global warming, there is a need to develop strategies to lower methane emissions from ruminants as part of an overall requirement to improve the sustainability of ruminant food production systems. Methane is produced as a by-product of anaerobic fermentation in the reticulo-rumen, largely due to the activity of methanogenic *archaea*. Recent research has focused on the potential of novel feed ingredients (probiotics, ionophores, acetogen-based inoculants, bacteriocins, organic acids and plant extracts) or vaccines to lower hydrogen production and/or increase the transfer and utilization of metabolic hydrogen in the production of end-products other than methane in the rumen. Research to date has provided evidence that dietary supplements of plant or marine oils, oilseeds, specific fatty acids and condensed tannins, as well as defaunation, increases in production level or decreases in the proportion of forage in the diet may lower enteric methane production. Even though dietary lipid supplements can be used to lower methane output, in high amounts a decrease in intake and milk production can be expected. While further investigations have demonstrated the efficacy of specific agents on methanogenesis *in vitro*, the effects have not been substantiated *in vivo*. Altering the ratio of H₂ /non-H₂ producing fibrolytic bacteria to lower methanogenesis without altering fibre digestion has been demonstrated under experimental conditions. Furthermore, non-H₂ producing communities have been characterized in the digesta of certain ruminant species. In contrast, stimulating acetogenesis by inoculation with rumen acetogens or non-rumen acetogens have met with limited success *in vitro* and *in vivo*. Research has also concentrated on stimulating the utilisation of metabolic hydrogen by sulphate reducing bacteria, but there remains concern over the toxicity of H₂S in the host ruminant. Investigations of nitrate reducing bacteria which produce more NH₃ and less toxic nitrite, have indicated promising results. Increasing the number of capnophilic bacteria which use CO₂ and H₂ to produce organic acids, succinic acid in particular, may decrease methane production. In isolation, several approaches have been shown to decrease enteric methane emissions, but often part of the changes observed are related to lowered organic matter digestion in the rumen. However, lowering methane production per unit product over the lifetime of an animal should be regarded as the central goal to decrease GHG from ruminant livestock systems. This highlights the need for integrated solutions to improve digestive efficiency, as well as fertility and health. In conclusion, any prospective solution to lower on-farm GHG emissions must be practical, cost effective and have no adverse effect on the profitability of ruminant meat and milk production. Recent research has indicated significant potential, but none of the strategies tested thus far satisfy all of the necessary criteria for immediate implementation.

Key words: Methane, Ruminants, Nutritional strategy

Introduction

The demand for meat and milk is predicted to almost double by 2050 (Steinfeld *et al.*, 2006) due to increases in the global population and increased consumption of these foods in developing countries. Ruminants that are capable of utilizing fibrous feeds, not digested by mono-gastrics, represent a valuable natural resource to meet global food requirements in the future. However, ruminants contribute to both local (nitrogen and phosphorus) and global (greenhouse gases (GHG; collectively CH₄, CO₂ and N₂O) emissions into the environment (Morgavi *et al.*, 2010). Overall, the livestock sector is responsible for 18% of global anthropogenic GHG emissions (Steinfeld *et al.*, 2006). Losses of methane (CH₄) represent 30 to 50% of total GHG from animal livestock production systems, with the contribution from ruminants accounting for about 80% (Gill *et al.*, 2010). Due to the concerns of increases in GHG emissions into the environment and potential effects on global warming, there is a need to develop strategies to lower CH₄ emissions from ruminants, as part of an overall requirement to secure and develop more sustainable ruminant food production systems in the future. Over a wide range of diets, enteric CH₄ accounts for between 2 to 12 % of dietary energy intake (Johnson and Johnson, 1995). In addition to concerns on GHG emissions, it is important to recognize that CH₄ represents a significant loss of energy that could potentially be repartitioned towards tissues or the mammary gland. In attempting to mitigate both local and global emissions into the environment, research should arguably be directed towards lowering enteric CH₄ and CO₂ as well as N₂O per unit product as well as increasing animal longevity. This highlights the need for integrated solutions that do not simply focus on improved digestive efficiency in isolation, but also target improvements in fertility and animal health. In the following short review, both established and emerging nutritional strategies to lower ruminant enteric CH₄ emissions are considered.

Strategies to lower enteric methane emissions

Methane is produced as a by-product of anaerobic fermentation in the reticulo-rumen of ruminants due, in a large part, to the activity of methanogenic *archaea*. Due to the complexity of the rumen microbial ecosystem, other microorganisms also regulate and alter CH₄ production (Morgavi *et al.*, 2010). Existing strategies to lower enteric CH₄ emissions include increasing feed intake, proportion of concentrates in the diet, feeding high-quality forages or dietary supplements of plant and marine oils, oilseeds or specific fatty acids and ionophores. Recent research has focused on the potential of novel feed ingredients (probiotics, acetogens, bacteriocins, archaeal viruses, organic acids and plant extracts), vaccination of host animal against some methanogenic bacteria and the selection of cows with inherently lower losses of CH₄ as a proportion of dietary energy intake (Boadi *et al.*, 2004).

Losses of CH₄ as a percentage of gross energy intake decreases 1.6% for each multiple of maintenance intake (Johnson and Johnson, 1995). The benefits of higher intakes are, at least in part, due to changes in rumen digestion kinetics. Mean retention time in the rumen is thought to explain about 28% of variation in CH₄ production (Okine *et al.*, 1989). Decreases in enteric CH₄ emissions in response to increases in concentrate supplementation are thought to arise from several factors including a reduction in the molar acetate:propionate ratio of rumen volatile fatty acids, decreases in rumen pH and lowered protozoal numbers (Martin *et al.*, 2010).

Supplementing diets with lipids is arguably one of the most practical and effective strategies to lower enteric CH₄ emissions in ruminants. Based on an extensive evaluation of available data, it was reported that lipid supplements decrease enteric CH₄ output on average by 3.8% per 1% dry matter (DM) increase in dietary fat content (Martin *et al.*, 2010). While dietary lipid supplements have been shown to lower CH₄, most studies have been relatively short in duration, and there are few data on the efficacy over an extended period (Martin *et al.*, 2010). A summary of trials conducted in New Zealand reported that dietary supplements of a mixture of sunflower and fish oil (500 g/d) over a 14d period lowered CH₄ by 27%, while no change in CH₄ was observed in cows fed 300 g/d of linseed oil and fish oil for 77d (Woodward *et al.*, 2006). It remains unclear if the inhibitory effects of fatty acids on rumen methanogenesis persist for long periods, or whether microbial communities in the rumen adapt over time. While dietary lipid supplements decrease enteric CH₄ production, feeding oils in high amounts (≥ 50 g/kg diet DM) often

lower feed intake and milk production (Martin *et al.*, 2008; Hristov *et al.*, 2011). Overall, dietary fat addition results in the most consistent decrease in CH₄ relative to changes in the forage:concentrate ratio of the diet or other feed additives, that when fed in moderate amounts can lower GHG without compromising the performance of growing or lactating cattle (Grainger and Beauchemin, 2011).

Ionophores such as monensin cause a moderate but transitory inhibition of rumen methanogenesis. Decreases in CH₄ to ionophores are related to a reduction in rumen protozoal numbers (Guan *et al.*, 2006), and alterations in ruminal bacterial populations, *i.e.* inhibition of the growth of *Ruminococci* without affecting *F. Succinogenes* (Chen and Wolin, 1979). Since January 2006 the use of ionophores in animal feeds has been banned in the European Union. It has been suggested that the relationship between the diversity of cellulolytic microorganisms in the rumen and CH₄ production merits further investigation, based on evidence that metabolic hydrogen and CH₄ production can be decreased in the absence of lowered fibre digestion (Morgavi *et al.*, 2010).

Altering the ratio of H₂ /non-H₂ producing fibrolytic bacteria to lower methanogenesis without altering fibre digestion has been demonstrated under experimental conditions (Morgavi *et al.*, 2010). This concept is supported based on evidence of the occurrence of dominant non-H₂ producing microbial communities in the rumen of certain feral ruminants. Populations of non-H₂ producing fibrolytic bacteria (*Fibrobacter*) were found to be higher and that of methanogens were lower than expected in rumen contents of buffaloes under natural conditions (Morgavi *et al.*, 1994). Furthermore, non-H₂ producing fibrolytic bacteria have been shown to produce less CH₄ *in vitro* (Chaucheyras-Durand *et al.*, 2010).

A meta-analysis concluded that probiotic live yeasts have no effect on CH₄ production (Sauvant, 2005). However, the findings of other studies indicate that probiotic yeasts have variable effects on CH₄ emissions (Doreau and Jouany, 1998; Chaucheyras-Durand *et al.*, 2008), due to functional and metabolic diversity between specific strains (Newbold and Rode, 2006). In light of the significant genetic diversity between yeast strains, the potential of these feed additives to lower CH₄ emissions merits further investigation (Martin *et al.*, 2010).

Certain bacteriocins including nisin and bovicin have been tested *in vitro* or *in vivo*. Most evaluations are based on functional studies *in vitro* with few data *in vivo*, highlighting that much more information on the stability and efficacy of bacteriocins in ruminants is required before these can be used on-farm (Martin *et al.*, 2010). Some time ago, it was suggested that archaeal viruses that act against rumen methanogens could be used to decrease CH₄ production (Klieve and Hegarty, 1999), but thus far, these have not yet been isolated and/or identified in the scientific literature (Martin *et al.*, 2010).

Dietary supplementation of 100 g fumaric acid/kg diet DM in free or encapsulated form was shown to decrease CH₄ by 62% and 76%, respectively in growing lambs (Wood *et al.*, 2009). In contrast, other studies have reported that fumaric acid supplements had no effect on CH₄ emissions when fed at 175 g/d to growing beef cattle (Beauchemin and McGinn, 2006), at 80 g/d to steers (McGinn *et al.*, 2004) or between 4–10 g/100 g (diet DM) in lambs (Molano *et al.*, 2008). Other investigations have examined the potential of organic acids to serve as alternative hydrogen sinks to CH₄ in the rumen. Dietary supplements of DL-malic acid (from 0 to 75 g/kg diet DM) were reported to decrease linearly CH₄ production in beef cattle, changes that were also accompanied by lowered DM intake, total rumen VFA production and molar acetate to propionate ratios (Foley *et al.*, 2009a). It has been speculated that the potential of organic acids to lower CH₄ may depend on the forage to concentrate ratio of the diet (Foley *et al.*, 2009b). Further experiments are required to define conditions that optimize the efficacy of organic acids in the rumen and the persistency of their effects on rumen methanogenesis (Hook *et al.*, 2010).

Three main plant compounds, condensed tannins, saponins, and essential oils, have been identified as effective for lowering CH₄ production *in vitro* (Martin *et al.*, 2010). Tannins are classified into two groups; condensed tannins and hydrolysable tannins. The anti-methanogenic activity of tannins has been attributed mainly to condensed tannins, whereas hydrolysable tannins are considered toxic to the host ruminant (Martin *et al.*, 2010). Two different mechanisms explaining the mode of action of condensed tannins on CH₄ have been described; a direct effect on ruminal methanogens and an indirect effect on hydrogen production due to lower feed degradation in the rumen (Tavendale *et al.*, 2005). Condensed tannins are

found in tropical shrub legumes including *Lotus spp.* and *Acacia spp.* Dietary supplements of plants or extracts of condensed tannins have variable effects on CH₄ production (0 to -30%) in ruminants (Martin *et al.*, 2010). Adding condensed tannins to the diet cannot be assumed to lower rumen methanogenesis, and their use requires further research.

Saponins, a group of secondary compounds, are found in many plants. These glycosides have a direct effect on rumen microbes and decrease protein degradation and increase microbial protein synthesis in the rumen (Makkar and Becker, 1996), changes that lower the availability of hydrogen for CH₄ production. Furthermore, saponins have been shown to increase ruminal concentrations of propionate at the expense of acetate and butyrate (Abreu *et al.*, 2004) that would be expected to decrease CH₄ production.

In the recent years, several investigations have explored the potential of essential oils to lower CH₄ *in vitro*. Essential oils are steam-volatile or organic-solvent extracts of plants (often herbs and spices) containing cyclic hydrocarbons and their alcohol, aldehyde or ester derivatives (Patra and Saxena, 2009).

Essential oils are lipophilic and interact with cell membranes which accounts for anti-bacterial and anti-fungal properties (Patra and Saxena, 2009). Components in essential oils are particularly toxic to gram positive bacteria (Jouany and Morgavi, 2007) and therefore, are capable of influencing rumen fermentation patterns. Garlic oil and some of its constituents have been shown to decrease CH₄ production *in vitro* due to the toxicity of organosulphur compounds such as diallyl sulphide and allicin on methanogens (Busquet *et al.*, 2005; Macheboeuf *et al.*, 2006). Supplementing the diet with 1 g/d of essential oils and spice extracts was demonstrated to have no effect on CH₄ output or alter feed digestibility in heifers (Beauchemin and McGinn, 2006), while further investigations *in vivo* are required to assess the efficacy, persistency and toxicity of these compounds in ruminants (Calsamiglia *et al.*, 2007).

A vaccine against three selected methanogens has been developed in Australia. Immunization in sheep lowered CH₄ production by 8%, while further testing failed to confirm efficacy in other geographical regions (Wright *et al.*, 2004).

It is possible to suppress the activity of rumen methanogenes with chemical agents including halogenated CH₄ analogues. Drenching cows with chloroform resulted in a dramatic initial decrease in CH₄ production and methanogen populations, but rumen methanogenesis gradually recovered from 5 to 39 days of treatment, suggesting resistance or adaptation of affected microbes over time (Knight *et al.*, 2011). However, use of this approach cannot be considered practicable, since chloroform is a known carcinogen and exhibits hepatotoxic properties (Knight *et al.*, 2011).

Repartitioning metabolic hydrogen in the rumen

When methanogenesis is inhibited, H ions must be utilised in other metabolic pathways in the rumen to avoid negative effects on fermentation (Knight *et al.*, 2011). Increasing acetogenesis by natural rumen acetogens or non-rumen acetogens have met with limited success *in vitro* and *in vivo* (Morgavi *et al.*, 2010). The potential of increasing metabolic hydrogen use by sulphate reducing bacteria has been examined, but there is serious concern over the production of H₂S as an end-product due to toxic effects in the host ruminant (Gould *et al.*, 1997). Nitrate is another possible sink for hydrogen produced during rumen carbohydrate fermentation, but the reduction of nitrate results in the production of nitrite, which is both toxic to ruminants and slowly converted to NH₃ in the rumen. Investigations of nitrate reducing bacteria which produce more NH₃ and less toxic nitrite, have indicated promising results (Iwamoto *et al.*, 2002; Sar *et al.*, 2005). Increasing the number of capnophilic bacteria which use CO₂ and H₂ to produce organic acids, succinic acid in particular, may decrease CH₄ production. While further studies have demonstrated the efficacy of specific agents on methanogenesis *in vitro*, but there is insufficient data *in vivo* to confirm the potential of these agents to lower CH₄ in practice.

Conclusions

Proposed strategies to lower on-farm CH₄ emissions must be practical, cost effective, sustainable and have no substantial adverse effect on the profitability of ruminant livestock production in order to be considered viable. Manipulating diet composition to induce changes in rumen fermentation characteristics

remains the most feasible approach to achieve immediate decreases in CH₄ production. However, lowering CH₄ production per unit product over the lifetime of productive ruminants should be seen as the central goal to decrease GHG emissions of ruminant livestock systems. This highlights the need for integrated solutions that not only result in improved digestive efficiency, but also target improvements in fertility and animal health as a means to extend ruminant productive lifetime. While recent research has indicated significant potential, none of the strategies tested satisfy all of the necessary criteria for immediate implementation.

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