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Characteristics Of Some Potential Forages In Indonesia In Reducing Methane (Ch4) Emission From Ruminants: *Benefits And Limitations*

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Abstract

Animal production can be more efficient and also sustainable if we reduce CH4 production from ruminal fermentation. One option is to find alternative forages that modify rumen fermentation. CH4 is not only harmful the environment but also means loss to the animals. All of the aspects of the issue is related to the condition of ruminant's farm in Indonesia. Some other forages that are mainly fed as protein source to ruminants, are: cassava leaves, sweet potato leaves, soya bean leaves, tofu waste, leaves of *Artocarpus heterophyllus*, *Musa paradisiaca L*, *Ipomea batatas*. Roughage sources are hays of *Panicum maximum*, *Pennisetum purpureum*, and *Setaria sphacelata* and the concentrate sources mainly corn, rice bran and cassava waste, and corn cobs. However, there are very limited studies in finding alternative forages that can both increase animals productivity and also reduce CH4 production. Only forages relevant to Indonesia that have been studied *in vitro* is reviewed in this article, about its potential in reducing CH4 production from rumen fermentation. Even though some forages reduce CH4, it could negatively influence digestibility, hence less productivity. Some studies indicated that it was due to the fat content of the forage while others indicated that the concentration of the bioactive compounds such as condensed tannin influence the side effect of low CH4 ruminal production.

Background

Among the Green House Gases (GHG) produced by the ruminants' animals, methane (CH_4) is the most important contributor to global warming (Beauchemin et al., 2010; Wood and Rowlings, 2011). A previous study indicated that CH_4 accounts for 50% of GHG emission from agricultural sector, with indications that problem could become worse with the projected of 40-55% increase in human population, and therefore food consumption, exacerbated by a relative increase in the demand for animal products, particularly in Asia (Vergé et al., 2007). In another study, it was estimated that approximately 80 million tonnes of CH_4 per year are produced around the globe by ruminant-based industries, particularly if the animals consume a diet comprising slowly digested fibrous diet (Jayanegara, 2010). Seijan (Sejian et al., 2011) reported that CH_4 emissions varied among the ruminant species, with goats contributing 13.7 g/animal daily (7-fold less than cattle, but the output depends on the type and composition of the feed in the ration).

Importantly, CH_4 emissions are a loss to the animal. Normally, the final products of ruminal fermentation are CO_2, CH_4 and volatile fatty acids (VFA), with VFA being the source of energy for bodily functions (Banik et al., 2013). Normally, the total VFA concentration in the rumen is 70-150 mM, where higher values indicating better feed degradability (Bergman, 1990). Most CH_4 is synthesized in the rumen by methanogens that have an enzyme that utilize the H2 that is produced largely by protozoa, combining it with CO_2 (Murray et al., 1976) (Morgavi et al., 2011). However, H_2 reduction can be switched to fumarate pathways that form propionate, a major substrate for gluconeogenesis and thus energy production (Mitsumori and Sun, 2008). Therefore, avoiding CH_4 formation allows the animal to retain extra energy, the equivalent 2–12% ATP equivalents (Johnson and Ward, 1996). Importantly, especially from the perspective of the recent project, H_2 reduction pathways can be changed by manipulating the diet (Martin et al., 2010; Mitsumori and Sun, 2008; Morgavi et al., 2008).

All the aspects of the CH_4 issue, the GHG emissions and the energy inefficiency, are directly relevant to Indonesia. It is a developing agricultural country with a population growth rate of 1.01% per annum (Talib, 2007), with a rapidly increasing demand for ruminant products such as meat and milk (Talib, 2007), especially during the peak periods where animals are sacrificed for moslem religious occasions. This is obviously a market opportunity for local ruminant farmers, but they rarely fully realize the opportunity because there is insufficient supply of high quality forages. One possible solution to such problem is the application of integrated farming systems, but there are questions around whether ‘sustainable intensification’ can offer the best outcome for the ruminants, the environment or mankind (Eisler et al., 2014). Livestock will continue to be a prominent source of food for humans (Wood and Rowlings, 2011) and they also provide many other resources (Eisler et al., 2014), so we need to find ways to mitigate the environmental impact of livestock production.

Indonesia, a developing agricultural country with the population rate of 1.01% per annum and the total population of 223 millon people in 2006 has been a highly prospective market target for the well developed countries, particularly in dairy and meet products (Talib, 2007), due to the higher local market demand, brings the small ruminant farm convincing to be well developed in the near future. In fact, there is wider opportunity to export goat’s meat overseas such as to Malaysia, Brunei Darussalam and the middle east countries; instead of fulfilling market demand domestically (Diwyanto, 2008).

Thus, it is required feed that can be long lasting, safe for both the environment and the animals, and efficient, in order to achieve the target of reducing methane emission and increase animal production. This paper will review some research about types of forages in Indonesia that have been studied for their potential in reducing methane production from rumen, with the benefits and limitations that affects the end result to the animals productivity.

Traditional Ruminant Forages in Indonesia

There are some traditional plants used as ruminants feed, either through the ‘cut-and-carry’ method or through integrated farming, such as: *Pennisetum purpureum, King grass, Setaria decumbens, Caliandra calothrysus, Kudzu grass, Sesbania grandiflora, Imperata cylindrica Div, Leucaena leucocephala*, *Calopogonium, Centrosema pubescens, Pterocarpus indicus, Gliricidia maculate.* Some are recently fed to ruminants: *Medicago sativa L, Mutingia calabura L, Albizzia falcate, Arachis hypogaea, Cassia siamea, Trifolium repens, Erythrina lithosperma, Psophocarpus tetragonolobus, Albizzia procera*; the leaves of *Spondeas lutea, Moringa oleifera*, *Eugenia aquena, Ceiba petandra, Mangifera indica, Hibiscus rosa-sinensis, Lannea grandis, Dendrophthoe pentandra, Desmanthus virgatus* (Mink, 1983; Quattrocchi, 2006; Services, 2013; Soerjani et al., 1987), and *Brachiaria humidicola* (Delima et al., 2015). However, only few of these forages have been studied systematically, with data published in academic sources, so little solid information is available, even for their effects on animal productivity. No information is available regarding their effects on methane production in small ruminants, only a few have been studied in large ruminants.

There are also some other good quality plants that are potentially but less commonly used as feed for ruminants, such as: *Brachiaria subquadripara, Eleusine indica (L) Gaerta, Eragrostis amabilis , Eragrostis brownie, Eulalia trispicata, Saccharum officinarum, Leptochloa chinensis, Brachiaria reptan, Hymenachne acutigluma*, *Paspalum gueonarum, Stylosanthes guianensis, Caliandra calothyrsus,* and *Bracharia humidicola* (Delima et al., 2015). Some of these plants have not been academically published.

Some other forages that are mainly fed to ruminants, categorized as protein sources are mostly leaves of crops, and legumes, such as: cassava leaves (241%), sweet potato leaves (19,2%), leaves of soya bean (16,7%) tofu waste (30.3%), (Administrator, 2012; Ashari et al., 1999); leaves of *Artocarpus heterophyllus*, *Musa paradisiaca* L, Ipomea batatas (Nusantara, 2009). Roughage sources are hays of *Panicum maximum*, *Pennisetum purpureum*, and *Setaria sphacelata* and the concentrate sources mainly corn, rice bran and cassava (Ashari et al., 1999), the leaves of *Manihot esculanta* (Ashari et al., 1999; Sirait and Simanuhuruk, 2010) , corn cobs with the nutrition contents: water, dry matter, crude protein and fiber is 29,54%; 70,45%; 2,67% dan 46,52% respectively in 100% dry matter (Ashari et al., 1999).

Some of those forages have anti nutrition compounds or less palatable for the ruminants. The treatment such as ensilage can be done to increase its palatability (Ashari et al., 1999). However, to eliminate the HCN content, the leaves should be air dried prior to fed to the animals or also can be in the form of silage (Sirait and Simanuhuruk, 2010). The leaves of *Morus sp* contains high protein (15,0–35,9%), and gas fermentation production is 35,4 –60,8 ml/200mg with the ME is 7,7 –12,3 MJ/kg DM (Ashari et al., 1999).

The compilation of the use of recent forages for ruminants in Indonesia is shown in the table as follows:

Tabel 1. The effect of using recent forages for the small ruminant's productivity in Indonesia

Species	**Optimum level of feeding**	**Effect on production**	**Sources**
The leaves of *Manihot utilissima*	1500 g/head/day, + natural grass *ad libitum*	31 g/h/d	(Wargiono and Sudaryanto, 2000)
***Panicum maximum* : concentrate**	20% : 80%	No report on production but decreasing CH4	(Gustiar et al., 2014)
Chopped Sweet potato	*Ad libitum*+ consentrate, eggplant and Penisetum purpureum	44 g/h/d in goats	(Katongole et al., 2009)
Manihot esculenta	30% + natural grass *ad libitum*	109 g/h/d in sheep	(Sirait and Simanuhuruk, 2010)
Murbei (*Morus alba*)	1,5%BW + King Grass (basal diet)	101 g/h/d in sheep	(Yulistiani, 2015)
	30% of ration + ammoniated hay basal diet	75.4 g/h/d	(Yulistiani et al., 2007)

The highest body weight gain of the sheep is when using *M.esculenta* 30% as a supplement on the basal diet natural grass fed ad-libitum (Sirait and Simanuhuruk, 2010). Whereas to the goat is by feeding chopped sweet potato, mixed with concentrate, eggplants and *P.purpureum* (Katongole et al., 2009). However, it still requires further study on how efficient the formula is and whether or not it can be sustainable for the animals and the environment.

Characteristics of some prospective plants / additives in Indonesia in reducing ruminal methane production

There are a few plants and herbs that has been studied in Indonesia for their potential in reducing CH_4 emission from ruminants.

1. *Hibiscus tiliaceus L*

This plant grows maximum 1700 m above the sea level, can grow 5 – 15 meters high, preferable on the fertile soil, and drought resistant. It has straight stem and small leaves but on the unfertile sand its stem is curved with diameter 40 – 50 cm, and wider leaves, branched and brown, and the underside leaves is hairy (Syamsuhidayat and Hutapea, 1991). The roots of *H. tiliaceus* can be used to cure fever, cough, diarrhea, and also to grow hair. The flower is used to cure trachoma and colds (Syamsuhidayat and Hutapea, 1991). Chemical content of leaves and roots of *H. tiliaceus* are saponins and flavonoids, tannins and polyphenol (Syamsuhidayat and Hutapea, 1991); alkaloid, amino acid, carbohydrate, organic acids, fatty acids, and steroid (Bandaranayake, 2002). The nutrition content is:

ash 10,79%; protein 17,08%; fat 3,45%; fiber 22,77%; carbohydrate 45,91% (Bimasmaraputra, 2011). *H.tiliaceus* supplementation in ruminants feed 10%, didn't negatively affect goat's productivity, where rumen pH is 7,05, NH_3 concentration 37,96 mg/100ml; protozoa 9,25 x 10^4/ml, propionate 35,01mM and acetate (115,90Mm) the highest of all level of supplementation (Bimasmaraputra, 2011).

2. *Gliricidia sepium* (Gamal)

Gamal originally comes from Brazil and can grow easily on the 1200 meter above sea level. It was firstly introduced by a Dutch in Medan, Indonesia to be used as shading plants of tea tree. It is a leguminous plant that can grow rapidly in dry areas. Its basic habitat is in tropical forests, adaptable in less fertile and acid soil, and drought resistant (Chadhokar, 1982; Nusantara, 2009). The leaves are oval with the arrangement similar to leucaena or turi. Gamal flowers appear in summer and a butterfly-shaped collected at the end of the rod (Natalia et al., 2009). It has high branched with a height of 2-15 cm, 15-30 cm stem diameter, panicle-shaped flowers, pink, and the leaves will fall in the dry season (Nusantara, 2009).

It is known as high protein source fodder, with protein content 23% (Natalia et al., 2009) and 25,7% (Hartadi et al., 1993), Fiber = 0,7% (Natalia et al., 2009) and 13.3% (Hartadi et al., 1993) , dry matter = 29,1% and more than 17% digestible by ruminants (Natalia et al., 2009). It also contains anti nutrition (toxic compound) such as dicaumerol and Acid Cyanide (HCN), Nitrite, and Tannin. Therefore, it should be air dried prior to mixed with other grasses in feeding the ruminants, to release the toxin and to increase the quantity of feed intake (Natalia et al., 2009).

3. *Sapindus rarak*

Sapindus rarak originally comes from South East Asia and now can be easily found in Asia and Africa. The fruits have soft and brown pericaps that are used as a washing soap. There has never been an *in vivo* study of using it as feed additive in ruminants, even though it has potential to increase the effectiveness of ruminal fermentation due to its saponin content (Hamburger et al., 1992; Haryanto and Thalib, 2009; Thalib, 2004).

The extract of Lerak fruit, fatty acids long-chain unsaturated, ferric ions and sulphate ions as well as acetogenic bacteria preparations can be used to reduce enteric methane emissions (Haryanto and Thalib, 2009).

4. *Calliandra calothyrsus*

The plant is originally from Central America and Mexico and is found from southern Mexico to Central Panama (NFTA, 1988). It tolerates infertile soil, light acidic soil, and poorly aerated but not in alkaline soils (Orwa et al., 2009) and also drought tolerance (NFTA, 1988). It can grow in many different kind of soils but does not tolerate water logging and not particulate tolerant of shade (International, 1999).

It is a protein source fodder, with the 22% of protein content DM, and highly digestible for ruminants (60 – 80%). This legume potentially reduces CH4 production from ruminants gut (Tiemann et al., 2008), because it contains condensed tannin up to 11%. However, if it is fed a lot to ruminants, it may reduce protein digestibility to 40% (Orwa et al., 2009). Drying of *Calliandra calothyrsus* was shown to have a negative effect on the voluntary feed intake, which was associated with lower *in-sacco* digestibility found. However, there have not been found any problems with acceptability, when fed as a supplement (30 – 40%) (Maasdrop et al., 1999).

5. *Indigofera zollingeriana*

Indigofera was firstly introduced by European around 1900, drought tolerance, and salinity. It contains crude protein 26 - 31%; fibre 15,25%; calcium 0,22% and phosphor 0,18%. It can grow until1650 above sea level and prefer fertile soil and good drainage. Thus, it can be fed to ruminants (Hassen et al., 2007), both as a basal diet or supplement, due to its high digestibility (77%), particularly for the lactation period. It has low tannin around 0,6 - 1,4 ppm therefore it is highly palatable (Abdullah et al., 2012).

6. *Garlic Oil*

Garlic oil derives from crushed garlic cloves (*Allium sativa*), then is heated prior to distillation process. It contains many secondary plant products including allicin ($C_6H_{10}S_2O$), diallyl sulfide ($C_6H_{10}S$), diallyl disulfide ($C_6H_{10}S_2$), and allyl mercaptan (C_3H_6S) (Lawson, 1996). A study indicated that it can eliminate CH4 production out of the rumen, by decreasing acetate but increase propionate and butyrate. However, it is not influence N-NH3 concentration. Garlic oil mainly responsibles on metabolism of carbohydrate. In fact, the energy metabolism derived from carbohydrate fermentation probably impacts N-NH3 availability (Busquet et al., 2005).

Bioactive Compounds in the Plants that can eliminate methane production: benefits and limitation

There has been some research in Indonesia on bioactive compounds of the plants or herbs that can be used in ruminants feed. This part will discuss some overseas studies on the bioactive of the plants that can be found also in Indonesia, and discuss the benefits and the limitations.

Mostly, bioactive compounds that can eliminate ruminal methane production are tannins and saponins. However, under different concentration it can be harmful to the animals regardless their ability to reduce protozoa population and thus change the pathways of the fermentation temporary products (Wina, 2012). Some research showed that saponins and tannins able to alter the fermentation pathway to reduce the waste so that more energy will be more available for the animals (Benchaar et al., 2008). Basically, they are potential in manipulating rumen microorganism, by inhibiting the activity of certain species of rumen bacteria and reducing the number of ciliate protozoa and methanogens, so the cellulolytic bacteria can be actively fermented the carbohydrates in to energy for the animals and for the bacteria itself to convert it into microbial protein. Hence, it will increase the percentage of microbial protein absorbed in the intestine and eventually it will increase feed utilization for the ruminants (Kamra et al., 2006). However, the effect of saponin on protozoa is temporary (Ding et al., 2012; Jayanegara et al., 2011; Wina, 2012).

Saponin can function as surfactant to kill protozoa due to the chemical reaction between those compounds with the cholesterol in the membrane of protozoa (Ding et al., 2012), or can be as feed additive for the manipulation of rumen microbes to reduce CH4 production out of rumen (Kamra et al., 2006).

The fruit extract of lerak (*Sapindus rarak*) contains high level of saponin thus, can be used as defaunating agent (Thalib, 2004; Thalib et al., 2010; Wina, 2012). The extract of *S. rarak* fruit pericarp has been proved to eliminate the methanogenic activity and to increase sheep average daily gain by 40% (Wina et al., 2005). The population of protozoa in the rumen is directly related to the production of CH_4 reduced if population of rumen protozoa also reduced (Thalib, 2008).

Garlic (*Allium sativum*) extract has the potential to reduce ruminal methane formation without affecting the total fermentation in the rumen (Patra et al., 2006) due to its methanol and ethanol compounds which has been reported can obstruct methanogenic activity in the rumen (Kamra at al., 2006). During the rumen fermentation process, there is the gradual decrease of the content of the garlic oil and garlic extract (Busquet et al., 2005). The effect of garlic oil (30 and 300 mg / L) indicated a lower proportion of acetate and the higher proportion of propionate and butyrate (Busquet et al., 2005).

Ethanol and methanol extracts of cloves, garlic, and fennel potentially inhibit the production of methane out of rumen. All extracts of garlic and fennel decreased the proportion of acetate and the ration of acetate to propionate (Patra et al., 2006). Ethanol extract of *Sapindus mukorossi* completely hampered methane production *in* *vitro* along with a significant decrease in the number of protozoa and ratio of acetate / propionate (Kamra et al., 2006).

For plants that contain tannins, anti-methanogenic activity has been attributed mainly to condensed tannins. Two models of tannins on methanogenesis: a direct impact on digestibility of rumen methanogens and indirect effects on the production of hydrogen due to reduced feed quality is lower (Tavendale et al. 2005). Further *in vivo* studies are required to determine the optimal dose of the active compounds, in relation to the microbial adaptation, the presence of residues in animal products and anti- nutritional potential side effects of such molecules (Calsamiglia et al., 2007).

The use of saponin as the rumen modifier of the extract fruit of Sapindus rarak function as defaunator could increase average daily gain of sheep to 44% and improve FCR to 20%, reduce CH4 20%. The function of Sapindus rarak can be partially or fully combined with legume *sesbania* and *albizia* that contain protein 26,3% and 24,0% respectively (Thalib et al., 2010). Other recent in vivo study in cattle using higher concentrate portion than grass (*Panicum maximum*) indicated that on the level of 80% concentrate and 20% grass, CH4 production is the lowest compare to other level (440ppm or decrease 28.5%) and the highest TDN (45.42%) (Gustiar et al., 2014). An in vitro study using mixture of grass (*Panicum maximum*) and legume as the protein source (Caliandra, Gliricidia and Leucaena) 60 : 40%, indicated that Gliricida produced more VFA per unit ODM with acetate : propionate and butyrate is 61,5 : 32,5 : 6,0 respectively.

The compounds in garlic (*allium sativum*) such as allicin ($C_6H_{10}S_2O$), diallyl sulfide ($C_6H_{10}S$), diallyl disulfide ($C_6H_{10}S_2$) and allyl mercaptan (C_3H_6S) (Lawson, 1996), has the ability to increase the ratio of acetate : propionate thus can reduce methanogens (Busquet et al., 2005). The other way that had been studying in decreasing CH4 production out of rumen is through supplementation of fatty acids such as: sun flower oil, coconut oil, canola oil, and kernel (Dohme et al., 2000; Macmüller and Kreuzer, 1999). Those compounds work as defaunating agents that kills protozoa and influence the H2 pathway not to be used by methanogens (Johnson and Johnson, 1995). However, some limitations have been reported in using canola oil, not only reducing CH4 ruminal production but also reducing feed intake and fibre digestibility, thus negatively influence animal's performance (Beauchemin and McGinn, 2006). While in coconut oil, no limitations published (Machmüller et al., 2003; Soliva et al., 2003).

Another study using supplementation of garlic powder and coconut oil of 100 g/day and 7% respectively, and concentrate 0,5% BW and rice straw as the basal diet in cattle, could increase propionate, reduced acetate, CH4 production by 9%, respectively. This ration could reduce 68 – 75% of protozoa population and increase the population of rumen bacteria (Kongmun et al., 2011).

The details info about the potential of some plants that could be found in Indonesia in reducing methanogenesis, and the effects in rumen characteristics:

Table 2. Some potential plants in Indonesia in reducing CH4 production, and the effects in rumen characteristics *

Species	Effects on digestibility	Effects on Rumen characteristics	Rumen Fermentation	Bioactive Compounds	Researcher
Acacia villosa	increase OMD	pH = 7.49; decrease protozoa; increase bacteria	Very low total gas and CH4; and low NH3	Phenols and condensed tannin	(Jayanegara et al., 2011)
Eugenia aquea	increase OMD	pH = 7.44; decrease protozoa; increase bacteria	Low total gas very low CH4 and low NH3	Phenols and condensed tannin	(Jayanegara et al., 2011)
Coconut oil 12% (Ding et al.,	No report	Decrease Methanogens, fungi,	Low CH4	No report	(Ding et al., 2012; Sitoresmi et al.,

2012); 5% (Sitoresmi et al., 2009)		and *F.succinogenes*; increase *R.flavefaciens*			2009)
***Mimosa pigra* (100g/d)**	Higher DMD	Decrease methanogens	Low CH4	Condensed tannin	(Silivong et al., 2011)
Sapindus rarak	Improve DMD and FCR	Decrease methanogens	Low CH4	Condensed tannin	(Thalib, 2004)
Garlic powder	No report	Decrease methanogens and protozoa	Low CH4	Allicin, diallyl sulfide, allyl mercaptan	(Kongmun et al., 2011)
Canola oil	Reduce fiber digestibility and feed intake	Decrease methanogens	Low CH4	No report	(Beauchemin and McGinn, 2006)
Sweet Potato	Reduce digestibility	Decrease methanogens	No significant reduction in CH4	No report	(Bhatta et al., 2008)

*Source: compiled from research in Indonesia and overseas.

Those plants can be found in Indonesia and is prospective to be analyzed of their potential in reducing CH4 emission from ruminants in *in vivo* procedure. The previous research have indicated that plant secondary compounds in the forages such as tannins and saponins can modulate rumen fermentation so there will be a reduction of CH4 (Bodas et al., 2012; Guo et al., 2008; Wallace et al., 2002). However, the effect was varied in other study. Some indicated that there was the decrease in feed intake and digestibility (Beauchemin and McGinn, 2006; Bhatta et al., 2008). Further study is required particularly on how the plants compound influence the activity of the total methanogens in the rumen and how it can be balanced with the nutrition intake so that the negative effects on the animal can be reduced. Hence, there can be dual benefits, animal production can be increased and CH4 emission can be eliminated.

Conclusion

Animal production can be more efficient and also sustainable, if we reduce methane production from ruminal fermentation. One option is to manipulate rumen fermentation pathways through diet manipulation. However, not all the potential plants is safe for the animal production due to their plant secondary compounds, that in one side can reduce CH4 production from the rumen but the side effect is reducing the digestibility and feed intake.

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