

# GREEN HOUSE GAS CONTROL AND AGRICULTURAL BIOMASS FOR SUSTAINABLE ANIMAL AGRICULTURE IN DEVELOPING COUNTRIES

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## ABSTRACT

Important green house gases (GHG) attributed to animal agriculture are methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O), though carbon dioxide (CO<sub>2</sub>) contributes almost half of total greenhouse effect. Rumen CH<sub>4</sub> production in an enteric fermentation can be accounted as the biggest anthropogenic source. Some of prebiotics and probiotics have been innovated to mitigate rumen CH<sub>4</sub> emission. The possible use of agricultural biomass consisted of non-edible parts of crop plants such as cellulose and hemicellulose and animal wastes was proposed as a renewable energy and nitrogen sources. The ammonia stripping from digested slurry of animal manure in biogas plant applied three options of nitrogen recycling to mitigate nitrous oxide emission. In the first option of the ammonia stripping, the effect of ammonolysis on feed value of cellulose biomass was evaluated on digestibility, energy metabolism and protein utilization. Saccharification of the NH<sub>3</sub> treated cellulose biomass was confirmed in strictly anaerobic incubation with rumen cellulolytic bacteria, *Ruminococcus flavefaciens*, to produce bio-ethanol as the second option of ammonia stripping. In an attempt of NH<sub>3</sub> fuel cell, the reformed hydrogen from the NH<sub>3</sub> stripped from 20 liter of digested slurry in thermophilic biogas plant could generate 0.12 W electricity with proton exchange membrane fuel cell (PEM) as the third option.

**Key words:** GHG, rumen, methane, probiotics, ammonia stripping, biomass

## ABSTRAK

### PENGATURAN GAS RUMAH KACA DAN BIOMASA PERTANIAN UNTUK USAHA PETERNAKAN BERKELANJUTAN DI NEGARA BERKEMBANG

Gas rumah kaca (GRK) yang penting dan berkaitan dengan peternakan adalah metana (CH<sub>4</sub>) dan nitrogen oksida (N<sub>2</sub>O) meskipun karbon dioksida (CO<sub>2</sub>) menyebabkan hampir 50% dari efek rumah kaca. Metana yang berasal dari pencernaan ruminansia merupakan sumber antropogenik yang utama. Prebiotik dan probiotik yang dihasilkan telah mampu menekan emisi metana dari rumen. Pemanfaatan biomasa pertanian yang berasal dari komponen yang tidak dapat dimakan seperti selulosa dan hemiselulosa serta limbah ternak disarankan untuk digunakan sebagai sumber energi dan nitrogen terbarukan. Penjaringan amonia dari limbah ternak dalam usaha pembuatan biogas dilakukan dalam tiga alternatif daur ulang nitrogen untuk mengurangi emisi nitrogen oksida. Pada alternatif pertama, telah diamati pengaruh amonialisasi terhadap nilai biomasa selulosa dari aspek pencernaan, metabolisme energi dan pemanfaatan protein. Sakarifikasi selulosa melalui perlakuan NH<sub>3</sub> dalam inkubasi anaerob menggunakan mikroba selulolitik rumen, *Ruminococcus flavefaciens*, menghasilkan bio-etanol sebagai alternatif kedua dari penjaringan amonia. Pada percobaan baterai berbahan bakar NH<sub>3</sub>, pembentukan hidrogen dari NH<sub>3</sub> yang dijarah dari 20 liter limbah tercerna dalam pembuatan biogas termofilik dapat menghasilkan 0,12 W listrik menggunakan baterai membran tukar proton (PEM = *proton exchange membrane*) sebagai alternatif ketiga.

**Kata kunci:** GRK, metana, rumen, probiotik, penjaringan amonia, biomasa

## INTRODUCTION

The mitigation of anthropogenic six GHG such as carbon dioxide, methane, nitrous oxide, sulphur hexafluoride, hydrofluorocarbons and perfluorocarbons have been established as legally binding commitments in The Kyoto Protocol (IPCC, 1996). Important GHG attributed to animal agriculture are methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O). Rumen fermentation of ruminant livestock and anaerobic fermentation of agricultural

organic waste including animal manures are major contributors of CH<sub>4</sub> emission as anthropogenic sources (MOSS, 1993).

To abate the GHG emission, the development of mitigation methods of rumen CH<sub>4</sub> is the most significant issue in the world ruminant livestock production (VAN NEVEL and DEMEYER, 1996). The prompt increase of atmospheric N<sub>2</sub>O since last century is closely related to abrupt expansion of human and animal population after an innovation of Haber-Bosch

process. Severe environmental pollutions were caused at the same time though the reactive nitrogen withdrawn from atmosphere as stable paired nitrogen brought about prosperous food production. To secure food production, preventing environmental catalyses by global warming, sustainable development of animal agriculture should be sought in not only developed but also developing countries as an alternative way. Inventories of emitters and their abatements are accurately assessed in both GHG to develop “clean developing mechanism (CDM)” in Kyoto Mechanism. The CDM might give highly economical and environmental incentives for the implementation in developing countries. The key element of these recyclings must be low-input for sustainable animal agriculture in developing countries. Carbon and nitrogen recycling in the agricultural biomass as renewable energy and nitrogen resources might contribute mitigation of CH<sub>4</sub> and N<sub>2</sub>O (TAKAHASHI *et al.*, 2003; 2004; TAKAHASHI and UEMURA, 2009).

The present paper deals with perspective on GHG control and possible uses of biomass towards sustainable animal agriculture.

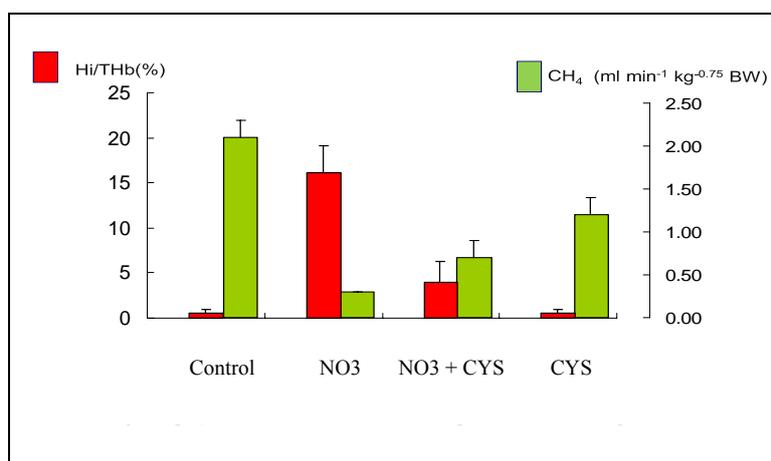
#### MITIGATION OF RUMEN METHANE EMISSION WITH PREBIOTICS AND PROBIOTICS

In the rumen, metabolic H<sub>2</sub> is produced during the anaerobic fermentation of glucose. This H<sub>2</sub> can be used during the synthesis of volatile fatty acids and microbial organic matter. The excess H<sub>2</sub> from NADH is

eliminated primarily by the formation of CH<sub>4</sub> by methanogens, which are microorganisms from the *Archaea* group that are normally found in the rumen ecosystem (Baker, 1999). The stoichiometric balance of VFA, CO<sub>2</sub> and CH<sub>4</sub> indicates that acetate and butyrate promote CH<sub>4</sub> production whereas propionate formation conserves H<sub>2</sub>, thereby reducing CH<sub>4</sub> production (WOLIN and MILLER, 1988). Therefore, a strategy to mitigate ruminal CH<sub>4</sub> emission is to promote alternative metabolic pathway to dispose of the reducing power, competing with methanogenesis for H<sub>2</sub> uptake. The administration of nitrate remarkably suppressed ruminal methanogenesis (TAKAHASHI and YOUNG, 1991; 1992).

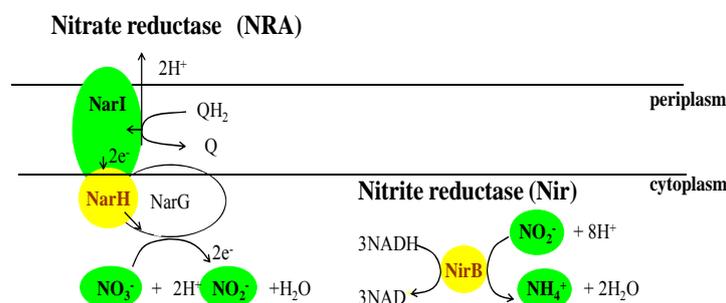
Figure 1 shows that the formation of toxic nitrite reduced from nitrate is successfully prevented by L-cysteine (TAKAHASHI and YOUNG, 1991, 1992; TAKAHASHI *et al.*, 1989, 1998, 2000, 2002), *i.e.* the effective mitigation of ruminal CH<sub>4</sub> emission is safely achieved by simultaneous administration of nitrate and L-cysteine without nitrite intoxication (TAKAHASHI, 2001). Furthermore, *Escherichia coli* modified genetically was developed in an attempt to promote nitrite reduction abating ruminal methanogenesis (SAR *et al.*, 2004a; 2005a; 2005b; 2005c) (Figure 2).

Rumen manipulation with ionophores such as monensin has been reported to abate rumen methanogenesis (MWENYA *et al.*, 2005). However, there is an increasing interest in exploiting prebiotics and probiotics as natural feed additives to solve problems in animal nutrition and livestock production as alternatives of the antibiotics due to concerns about

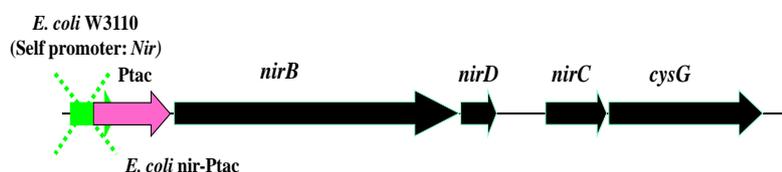


**Figure 1.** The suppressing effect of nitrate (1.3 g NaNO<sub>3</sub> kg<sup>-0.75</sup> body weight) on methane emission and prophylactic effect of L-cysteine (0.21 g S kg<sup>-0.75</sup> body weight) on nitrate induced methemoglobinemia in sheep

### 1. Wild-type *E. coli* W3110



### 2. Construction of *E. coli* nir-Ptac by replacement of self-promoter (*nir*) in *E. coli* W3110 by *tac* promoter (Ptac) (Ajinomoto Co. Inc., Kawasaki, Japan)



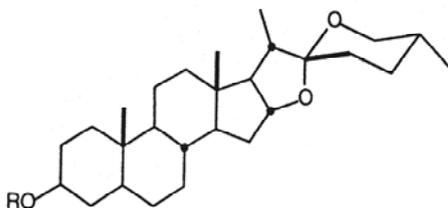
**Figure 2.** Wild-type *E. coli* W3110 and the construction of *E. coli* nir-Ptac

incidences of resistant bacteria and environmental pollution by the excreted active-antibacterial substances (MWENYA *et al.*, 2006). Nisin and saponin-containing extracts of *Yucca schidigera* and *Quillaja saponaria* have been categorized as 'generally recognized as safe (GRAS)' for human consumption by US Food and Drug Administration. Nisin produced by *Lactococcus lactis* subsp. *lactis*, antimicrobial activity against spectrum of Gram-positive bacteria is characterized bacteriocin and performed mitigating effect on ruminal methane emission (MWENYA *et al.*, 2004a; SANTOSO *et al.*, 2004b; SAR *et al.*, 2006). Saponins are natural detergents found in variety of plants. *Yucca* saponins have a steroidal nucleus, whereas *Quillaja* saponins are triterpenoid in structure (Figure 3). Supplementation of saponin-rich plant extracts decreased ruminal protozoa counts and decreased methanogenesis accompanied by decrease in the ruminal acetate/propionate (A/P) ratio *in vitro* and *in vivo* (WALLES *et al.*, 1994; WANG *et al.*, 2000, TAKAHASHI *et al.*, 2000; SANTOSO *et al.*, 2004a; MWENYA *et al.*, 2004a; PEN *et al.*, 2006). However, PEN *et al.* (2007; 2008) showed in recent detailed examination that *Q. saponaria* had no effect on ruminal

methanogenesis and A/P ratio, although it suppressed protozoa number.

Galacto-oligosaccharides (GOS) are non-digestible carbohydrates in nonruminants and have a long history of research as a prebiotics food ingredient. GOS are resistant to gastrointestinal enzymes, but are selectively utilized *Bifidobacteria* (SAKO *et al.*, 1999). In the rumen, *Bifidobacteria* and *Lactobacillus* species utilize fructose, galactose, glucose and starch as substrates to produce lactate and acetate. Lactate is intermediate compound of an acrylate pathway during propionate production in the rumen. Meanwhile, propionate production is indirect competition with methanogens for available hydrogen. As *Bifidobacteria* and *Lactobacillus* species in the rumen can utilize GOS and produce more lactate, ruminal methanogenesis have been suppressed by GOS with or without direct-fed microbe yeasts and lactic acid bacteria (GAMO, 2001; MWENYA *et al.*, 2004b; 2004c; 2004d; 2005; SANTOSO, 2004a; SAR *et al.*, 2002; 2004b; 2004c; TAKAHASHI *et al.*, 2002; 2003). However, the efficacy of GOS with the probiotics on different diets and animal species remains to be elucidated.

*Yucca schidigera*  
(Steroidal saponins)



*Quillaja saponaria*  
(triterpenoid saponins)

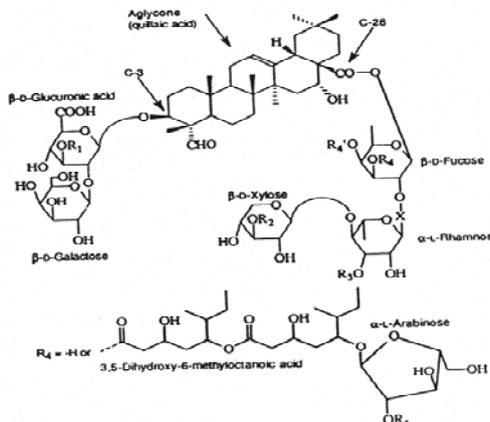


Figure 3. Chemical structure of *Yucca schidigera* and *Quillaja saponaria*

**Creation of renewable energy (biogas) from anaerobic fermentation (biogas plant) of animal manures and the innovative reuse of the digested slurry**

The increased emissions of CH<sub>4</sub> and N<sub>2</sub>O from decomposing unmanaged and bio-based industrial wastes along with the expansion of human activities contribute climate change as GHG. The biogas plant produce biogas including combustible CH<sub>4</sub> as renewable energy using unused resources like animal manures, can provide fuel, heat and electricity (TAKAHASHI *et al.*, 2004; UMETSU *et al.*, 2005, 2006; KOMIYAMA *et al.*, 2006), and minimize the impact on the environment thus reducing the amount of pollutants discharged. Biogas system and its application have been expanded in APEC member economies as a mitigation strategy with an economical incentive

(APEC, 2009). The conventional biogas system based on anaerobic fermentation of the organic wastes, however, is not a nitrogen recycling but a carbon recycling. Therefore, isometric fertilization of the digested slurry after anaerobic fermentation may not be a solution of current issue on excess nitrogen abatement, although nitrous oxide emission is almost completely suppressed during anaerobic fermentation (unpublished data). It causes not only methane emission, but also nitrate leaching and N<sub>2</sub>O emission from soil (TAKAHASHI, 2006).

The introduction of ammonia stripping from digested slurry of thermophilic biogas plant might be a solution to reduce total nitrogen content of the slurry as a liquid fertilizer containing suitable nitrogen and eventually can contribute the mitigation of N<sub>2</sub>O emission as a new concept of biogas system (Figure 4). Furthermore, the stripped ammonia can be put to

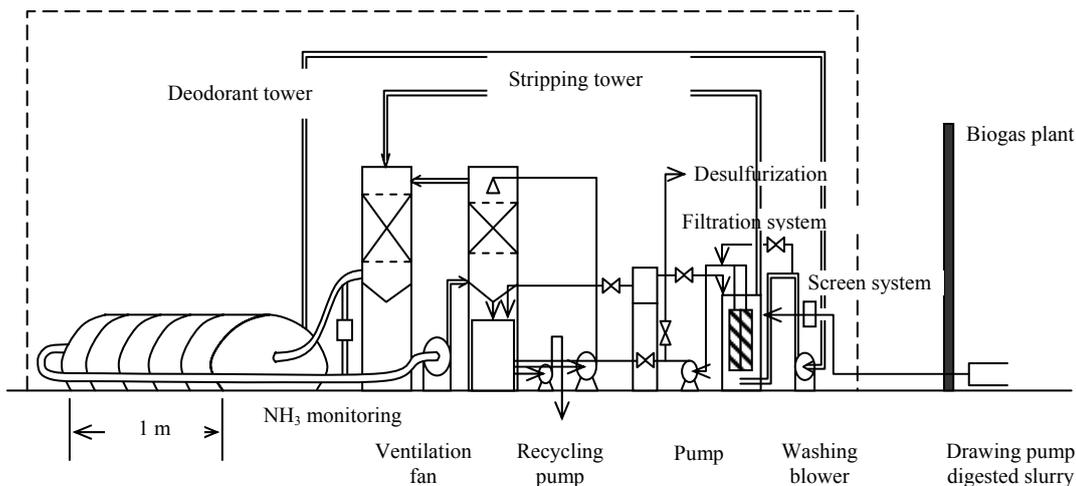
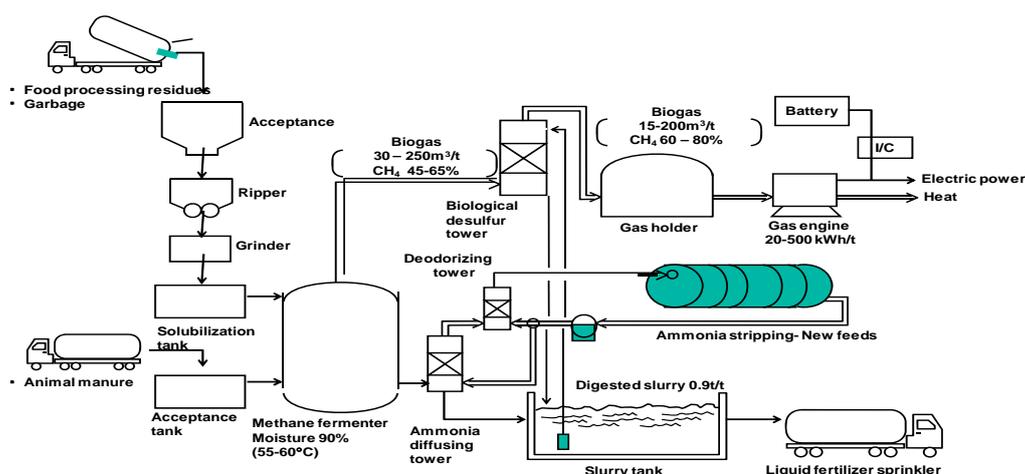


Figure 4. Ammonia stripping apparatus



I **Figure 5.** Biogas system addressed biomass to local society

practical use as a low-input and renewable nitrogen resource without energy supply from outside, because abundant amount of organic wastes exist in developing countries and the energy required for ammonia stripping can be supplied from biogas plant attached to the ammonia stripping apparatus. Figure 5 shows biogas system addressed biomass to local society.

The following three options have been examined for future nitrogen recycling:

1. Production of high quality feed from cellulose biomass in agricultural waste with ammonia stripping process from digested slurry of biogas plant (TAKAHASHI, 2006; 2007).
2. Saccharification of soft cellulose biomass to create bio-ethanol and hydrogen using ammonolysis by stripped ammonia from effluent and hydrolysis of rumen bacteria (unpublished).
3. Ammonia fuel cell with ammonia stripping from digested slurry (TAKAHASHI and UEMURA, 2009).

## CONCLUSION

Controlling the GHG emission from agricultural biomass for sustainable animal production in the developing countries can be conducted by supplementation of feed with probiotics or prebiotics, ammonia stripping from digested slurry of biogas plant; saccharification of soft cellulose biomass to create bio-ethanol and hydrogen; and to develop ammonia fuel cell with ammonia stripping from digested slurry.

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