EFFECTS OF SUPPLEMENTATION OF VEGETABLE OIL AND TANNIN ON METHANE GAS EMISSION IN RAISING CATTLE

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ABSTRACT

The experiments were conducted on 12 Red Sindhi crossbred cattle at 15-16 months of age for the 90-day trial in order to determine methane emission by addition of vegetable oil and tannin into feed. Cattles were housed in farm housing condition then were individually arranged to respiratory chambers to measure methane gas emissions. The diets consisted of basal diets with addition of 0.3 and 0.5% tannin; 1.5 and 3% vegetable oil on dry matter basic. Animals were fed ad libitum with 15 days of diet adaptation prior to testing. Results showed that the effect of different levels of tannin supplementation on the level of CH_4 and CO_2 emissions of experimental cattle was from 168.28 to 180.88 liters/day, there was a significant difference in the amount of methane released ($P<0.05$). Specifically, the control (without added tannin and vegetable oil) had the largest emitted methane at 180.88 liters/day and Treatment 2 (added tannin at 0.5% DM, vegetable oil at 3% DM) has the smallest methane gas emissions of 168.28 liters/day, lower than the control about 6.97%. When increasing tannin and vegetable oil supplement level, methane gas emissions tended to decrease. When supplemented with low tannin 0.3% DM and vegetable oil 1.5% DM (in Treatment 1) the amount of methane emitted was 176.05 liters/day and 125 g/day; However, when supplemented with high levels of tannin 0.5% DM and vegetable oil 3% DM the amount of methane emitted was 168.28 liters/day and 119.48 g/day. It was concluded that different tannin and vegetable oil supplements have a significant effect on the level of methane emissions of experimental cattle, as the supplement level increases, the methane emissions tend to decrease ($P<0.05$). The emission intensity of CH_4 tends to decrease with increasing dietary supplement of tannin and oil, however, when supplementation is at high level, the emission intensity tends to increase.

Use diets supplemented with 0.5% DM of tannin, 1.5% DM of vegetable oil to raise cattle for the best growth. Moreover, it has the good effect by decrease significantly methane gas emissions, contributing to reducing environmental pollution from livestock.

Keywords: *Tannin, Vegetable oil, Methane, Carbon dioxide, emission*

INTRODUCTION

Global warming in recent years is an immerging issue, attracting attention of human because of its effect on ecological balance... It is expected that the earth surface temperature will increase from 1.1 to 6.4^oC by 2100. Global warming is mainly due to the greenhouse effect but the main cause is the emission of CO_2, CH_4, NO_2 etc. from agriculture and livestock production (IPCC, 2007).

The livestock industry is an important sector not only in Vietnam but also in the world. It's contributing 40% GDP of the global agriculture. With a proportion of 26.8% in the structure of agriculture, the livestock activities in Vietnam are developing rapidly in both quality and quantity. Total greenhouse gas emissions (GHG) from livestock industry in Vietnam during the year 2010 about 18.03 million tons of CO_2 equivalent, accounting for 20.4% of total GHG emissions in agriculture and this number has been forecasted to rise to 24.95 million tons, accounting for 24.8% in 2020 (Nguyen Tien Dung, 2015). Of the total methane emissions out of animal husbandry, the amount of methane from ruminant breeding is the largest, approx. 74% (Tamminga, 1992). According to the livestock development strategy to 2020 of the Prime

Minister, the livestock industry has basically changed to new production methods as industrial farm, responding the majority of feed demand, quality assurance for consumption and export.

Livestock contributes about 16% of the planet's total methane gas, after fossil fuels and wetlands, and about 74% of livestock methane is caused by ruminant livestock. Methane (CH_4), carbon dioxide (CO_2), nitrous oxide (N_2O), and halocarbons are greenhouse gases (GHG) which improve the impacts of solar and thermal radiation on surface and atmospheric temperatures and are often expressed on a CO_2-equivalent (CO_{2e}) basis. Various bases are often used to express the share of GHG and CH_4 emissions from livestock farming (Lassey, 2008). Livestock related to methane emissions globally, estimate that CH_4 production will increase by about 60% to 2030. However, changes in feed and fertilizer can change this increase. According to US EPA (2006) is forecasts CH_4 emissions enteric and manure ferment will rise 21% in 2005 and 2020.

According to the evaluation of the Ministry of Science and Technology, the Ministry of Natural Resources and Environment, the Ministry of Agriculture and Rural Development and the Climate Change, Agriculture and Feed Security Programs (CCAFS), the annual total Greenhouse gas emissions from livestock in Vietnam are increasing rapidly. It accounts for over 22% of total greenhouse gas emissions in agriculture and this number is forecast to increase to 24.95 million tons, accounting for 24-25% by 2020.

However, the expansion of livestock scale has many negative impacts on the environment. Methane (CH_4) is produced by fermenting feed in the rumen of ruminant animals, when cattle burp feed to chew then methane is released. In general, add more feed for ruminant occur increase largely methane gas emissions.

In Vietnam, the ruminant (cattle, buffalo, goats, sheepetc.) is one of the main agents for increasing CH_4 due to the fermentation of rumen microorganisms to degrade feed. In the total amount of CH_4 released into the environment from livestock activities in Vietnam (ruminants, cattle, pigsetc.), ruminant husbandry made up about 30%. In order to reduce CH_4 in the rumen of cattle, it is necessary to find ways to reduce the generation of hydrogen, prevent and limit the activity of the CH_4-producing microorganisms.In recent years, there are a lot of research about reducing the methane emissions from livestockof which feeding tannin containing plant materials for ruminants (rumen digestion) is one of the best solution for this issue. From that point, this topic was done with the title: *"Effects of supplementation of vegetable oil and tannin on methane gas emission in raising cattle"*.

MATERIALS AND METHODS

Research station and data collection: Collecting the information of feed source for cattle raising in Le Chi commune, Gia Lam district, Ha Noi city. Research period: From 07/2016 to 07/2017.

Experimental design: The experiments were conducted on 12 Red Sindhi crossbred cattle at 15-16 months of age, with the same weight. They were randomly design in 3 treatments with 4 castles/each experiment. The experimental tracking time is 90 days. These cattle were feed in 15 days to adapt the living condition in the cages. Before taking the experiment, the cattle's were dewormed, vaccinated against pasteurellosis, foot and mouth disease according to the veterinary regulations. Cattles are concentrated in available cages under farmer's house conditions, arranged chambers to measure the methane gas emissions.

Table 1. Experimental layout

Criteria	Unit	Control	Treatment 1	Treatment 2
Number of cattle	Cattle	4	4	4
Weight of cattle (Mean±SE)	Kg/ cattle	153.63±0.24	153.75±0.75	153.25±1.11
Age	Month	15-16	15-16	15-16
Preparation time	Day	15	15	15
Experimental period	Day	90	90	90
Mineral lick (stone)		Optional	Optional	Optional
Tannin supplement	%DM	0	0.3	0.5
Vegetable oil supplement	%DM	0	1.5	3

Table 2. Chemical composition of feed in experiments

Feed	DM	ME	CP	NDF	ADF	CF	EE	Ash	Tannin
	(%)	*(Kcal/kgDM)*				*(%DM)*			
Natural grass	2.0	2,182.3	10.47	65.92	34.21	31.32	2.37	7.87	-
Corn silage	26.9	2,274.7	9.46	65.36	39.64	35.76	2.48	8.52	-
Cassava	87.8	2,757.4	3.55	61.46	41.78	8.74	0.1	1.81	-
Mashed corn	90.0	3,220.4	9.81	36.05	10.79	2.75	4.87	1.51	-
Mixed feed	90.4	3,027.2	15.87	56.03	12.36	9.19	1.61	9.82	-
Vegetable oil	95.2	7,342.5	-	-	-	-	-	-	-
Tea by-products	90.5	2,822.4	21.76	32.85	21.03	18.38	1.95	6.32	25.14

The experimental are used mixed feed according to the actual feed as shown in Table 3 including: Corn silage, natural grass, Cargill concentrated feed, mashed corn, Cassava residue combine with tannin, vegetable oil and by-products. Raw feed was chopped and mixed with cornmeal, cassava pulp and mixed feed before feeding experimental cattle. The experiments used mixed feed according to the following proportions:

Table 3. Mixing experimental diets (Unit:%)

No.	Feed	Control	Treatment 1	Treatment 2
1	Natural grass	24.55	24.06	23.96
2	Corn silage	49.09	48.12	47.92
3	Cassava	4.55	4.46	4.44
4	Mashed corn	10.00	9.80	9.76
5	Mixed feed	11.82	11.58	11.54
6	Vegetable oil	0	1.41	1.41
7	Tea by-products	0	0.58	0.98
8	Total	100	100	100

Table 4. Composition of experimental feed nutrition

Parcel	DM	ME	CP	NDF	ADF	CF	EE	Ash
	(%)	*(Kcal/kgDM)*	*(%DM)*					
Control	43.51	2,457.46	10.23	61.29	32.30	27.00	2.48	7.51
Treatment 1	44.51	2,528.35	10.15	60.26	31.78	26.57	2.44	7.40
Treatment 2	44.70	2,529.95	10.20	60.14	31.73	26.54	2.44	7.39

To study the effect of different levels of tannin and vegetable oil supplementation on CH_4 emissions of experimental cattle, we conducted: Every month, each cattle was put into the respiratory chamber for 1 day and determine the total amount of methane produced. There were 2 treatments for experiment cattle and 1 control for comparison. Every day, the feces, urine and leftovers from cattle in the respiratory chamber were collected to check the amount of methane emission. The determination of total methane production (liters/cattle/day) was determined through the methane analysis system attached to the respiratory chamber. The amount of methane produced per kg of weight gain had calculated when cattle were fed a different diet in each treament.

Indicators index

Chemical composition and nutritional value: Raw energy (GE), exchange energy (ME), Dry matter (DM), crude protein (CP), Insoluble fiber in neutral environment (NDF), Fibers insoluble in acid medium (ADF), crude fiber (CF), total minerals (Ash).

Assessing the effects of tannin and vegetable oil: Total emissions of CH_4 (liters/head/day), CH_4 (g/head/day), Total heat generated (HP) (KJ), CO_2 emission (liters/animal/day).

The monitoring methods

The total methane produced (l/head/day or l/kgDM) was determined through the methane analysis system attached to the respiratory chamber.

The amount of methane produced per kg of weight gain (MPG) of the experimental cattle was determine how much methane produced by a given diet when feeding a given diet, we calculated by the following formula:

$$\text{MPG (l/kg weight gain)} = \frac{\text{Total amount of methane}}{\text{Daily weight gain of cattle}}$$

The amount of CO_2 emission/day was estimated from the total ME intake and the total heat generated by the formula:

A (liter/day) = HP (KJ)/21.75

In which: Total amount of heat generated (HP) (KJ) = ME intake (KJ) - (kg weight x 20,000 KJ / kg weight gain).

Convert methane gas to raw energy by Brouwer method (1965), for every 1 liter of CH_4, it is equivalent to 0.71 grams of CH_4; Equivalent to 0.04 MJ of raw energy (GE).

Data processing methods

The data collected in the study was conducted to process biological statistics of Nguyen Van Thien et al. (2002). Using ANOVA and Tukey of Minitab software to check the degree of statistical discrepancy, the parameters include: Average value, Average error, Compare the average difference P (Sig.)

RESULTS AND DISCUSSION

The level of CH_4 emission of experimental cattle

Results of the effect of different levels of tannin supplementation on the level of CH_4 and CO_2 emissions of experimental cattle was shown in Table 5 and 6: the amount of methane emitted from the experimental parcel was from 168.28 to 180.88 liters/day, there was a significant difference in the amount of methane released ($P<0.05$). Specifically, the control (without added tannin and vegetable oil) had the largest emitted methane at 180.88 liters / day and Treatment 2 (added tannin at 0.5% DM, vegetable oil at 3% DM) has the smallest methane gas emissions of 168.28 liters / day, lower than the control about 6.97%.

Table 5. The level of CO_2 emissions and heat generated of the experimental

Creterial	Control		Treatment 1		Treatment 2		P (Sig.)
	Mean	SE	Mean	SE	Mean	SE	
Heat generated HP (KJ)	$19.417,8^{a}$	73,45	$18.680,6^{bc}$	342,13	$19.886,9^{ac}$	226,19	0.005
CO_2 (liters/day)	$892,77^{a}$	3,38	$858,88^{bc}$	15,73	$914,34^{ac}$	10,40	0.005
CH_4/ CO_2 (liters/day)	$0,20^{abc}$	0,001	$0,21^{ac}$	0,005	$0,18^{b}$	0,002	0.012

Note: Means for the same parameter within same horizontal lines with different letters are significantly different (P<0.05).

As the Table 5 presents so that the heat generated (HP) and the amount of CO_2 increase when adding tannin to the feed of cattle but in low level (P=0.005). The heat generated from 18.680,6 (KJ) in Treatment 1 to 19886.9 (KJ) in Treatment 2 and the amount increase from 858.88 litter/day to 914.34 litter/day. This may effect a little bit to the growth of cattle.

Table 6. The level of CH_4 emissions of the experimental cattle

Creterial	Control		Treatment 1		Treatment 2		P (Sig.)
	Mean	SE	Mean	SE	Mean	SE	
CH_4 (liters / day)	$180,88^{a}$	0,90	$176,05^{b}$	0,95	$168,28^{d}$	0,48	0,000
CH_4 (g / day)	$128,42^{a}$	0,64	$125,00^{bc}$	0,67	$119,48^{d}$	0,34	0,000

Note: Means for the same parameter within same horizontal lines with different letters are significantly different (P<0.05).

Comparing between experimental Treatment 1 and Treatment 2 with tannin and vegetable oil supplement with different levels, it is presented that when increasing tannin and vegetable oil supplement level, methane gas emissions tended to decrease. According to the result, when supplemented with low tannin 0.3% DM and vegetable oil 1.5% DM (in Treatment 1) the

amount of methane emitted was 176.05 liters/day and 125 g/day; However, when supplemented with high levels of tannin 0.5% DM and vegetable oil 3% DM the amount of methane emitted was 168.28 liters/day and 119.48 g/day.

This result shows that the different tannin supplementation levels have a significant influence on the methane gas emission level of the experimental cattle. Compared with the control, adding levels of tannin and vegetable oil at different levels, the amount of methane gas emissions decreased by 6.97% in Treatment 2 (about 3,6 time higher than in Treatment 1).

In recent years, the large amount of methane gas emissions (CH_4) is the negative effects of the livestock industry in general and ruminant in particular to our living environment. Therefore, reducing environmental pollution by reducing methane emissions from dietary changes supplemented with different levels of tannin and vegetable oils is the successful of this research. From the result, we can have some better solution for reduce methane emission in livestock industry with low cost and saving time.

The lowest CH_4 emissions in Treatment 2 is 119.48 g/cattle/day equivalent to 43.61 kg/cattle/year higher than published by Dinh Van Dung et al. (2016).

From the ratio of CH_4/CO_2, it is presented that the experimentalin Treatment 1 (added tannin at 0.3% DM, vegetable oil at 3% DM) had the largest value 0.204; this value is almost the same with the control 0.203. Treatment 2 (added tannin at level 0.5% DM, vegetable oil at 3% DM) has the smallest value of CH_4/CO_2 0.18, ($P<0.05$). Our results was similar to those published by Tran Hiep et al. (2016) indicating that CH_4/CO_2 ratios tend to decrease with increasing tannin and cotton oil levels in the feed for cattle, when adding cotton oil at 3% DM in feed, the lowest CH_4/CO_2 ratio was 0.052 ($P<0.05$).

The intensity of CH_4 gas emission by nutrient intake

Table 7. The intensity of CH_4 gas emissions by nutrients intake

Interpretation	Control		Treatment 1		Treatment 2		P (Sig.)
	Mean	SE	Mean	SE	Mean	SE	
CH_4/DM (l/kg/day)	44,35[a]	0,33	41,95[b]	0,26	39,52[c]	0,13	0,000
CH_4/DM(g/kg/day)	31,49[a]	0,23	29,79[b]	0,18	28,06[c]	0,09	0,000
CH_4/NDF(l/kg/day)	72,37[a]	0,53	69,62[b]	0,43	65,72[c]	0,22	0,000
CH_4/ADF(l/kg/day)	137,32[a]	1,01	132,01[b]	0,81	124,56[c]	0,41	0,000

Note: Means for the same parameter within same horizontal lines with different letters are significantly different (P<0.05)

There was a difference between the cattle's which was feed by cotton oil compared to the cattle which did not feed cotton oil ($P<0.05$), the lowest methane was supplemented with 3% DM of cotton oil in the feed of cattle. From the result of Table 8, it is presented that when adding tannin and vegetable oils to the diet in different level, the methane emission intensity according to cattle weight tended to decrease. However, when adding tannin and oil at a high level of tannin 0.5% DM, vegetable oil 3% DM (Treatment 2), the methane emission intensity tended to increase to 246.1 liters/kg WG ($P<0.05$).

The intensity of CH_4 gas emission is based on the amount of nutrients consumed

Monitoring the intensity of methane gas emissions according to the amount of digestive nutrients including dry matter, organic matter, acid-insoluble fiber and neutral medium insoluble fiber in the Table 9 was show that the intensity of methane emissions tends to decrease with the addition of tannin and vegetable oil at different levels in the diet. Control (no tannin and vegetable oil supplement) has the highest methane emission intensity of 63.0 liters/kgDM digestible and 157.68 liters/kgOM digestible. Next is Treatment 1 (tannin supplement at 1.5% DM, vegetable oil at 3% DM), the value of CH_4/kgDM is 56.60 and value of CH_4/kgOM is 147.83. Treatment 2 (tannin supplement at 0.5% DM, vegetable oil at 3% DM) has the lowest methane emission intensity, 53.94 liters/kgDM digestible and 145.74 liters/kgOM digestion ($P<0.05$).

Table 8. Intensity of methane emissions based on the amount of digestive nutrients

Interpretation	Control		Treatment 1		Treatment 2		P (Sig.)
	Mean	SE	Mean	SE	Mean	SE	
CH_4/DM(l/kg/day)	63,00^{a}	0,73	56,60^{c}	0,61	53,94^{e}	0,40	0,000
CH_4/OM (l/kg/day)	157,68^{a}	1,21	147,83^{ce}	0,49	145,74^{e}	0,71	0,000
CH_4/NDF (l/kg/day)	101,25^{a}	0,42	94,19^{c}	0,54	90,68^{e}	0,52	0,000
CH_4/ADF (l/kg/day)	217,39^{a}	1,61	196,74^{c}	1,83	190,48^{e}	1,36	0,000

Note: Means for the same parameter within same horizontal lines with different letters are significantly different (P<0.05)

When the improved digestion rate increases the amount of nutrients received, leading to the speed of feed transport in the road. Digestion will be faster, reducing the fermentation time of microorganisms in the rumen and resulting in a reduction in methane production. Thus, the addition of vegetable oil in the diet has reduced the loss of feed from methane to the environment. Comparison between experimental parcels supplemented with tannin levels and different vegetable oils, in the direction of increasing dietary supplementation, methane emission intensity according to digestive nutrients.

Raw energy is lost through CH_4 emissions

The effect of different levels of vegetable oil and tannin supplementation on the amount of energy lost through CH_4 emissions was showed in Table 9.

Table 9. Estimate the amount of energy lost through CH_4 emissions

Interpretation	Control		Treatment 1		Treatment 2		P (Sig.)
	Mean	SE	Mean	SE	Mean	SE	
GE received (Kcal)	16.741,6^{a}	94,08	17.231,2^{a}	25,22	17.488,3^{c}	37,36	0,001
GE received (MJ)	70,09^{a}	0,394	72,14^{a}	0,106	73,22^{c}	0,156	0,001
GE lose CH_4 (MJ)	7,24^{a}	0,036	7,04^{cd}	0,038	6,73^{e}	0,019	0,000
GE lose CH_4 (%)	10,32^{a}	0,076	9,76^{b}	0,060	9,19^{d}	0,030	0,000

Note: Means for the same parameter within same horizontal lines with different letters are significantly different (P<0.05)

From the data in the table it is presented that the raw energy (GE) received in three treatments according to the change of feed intake. Treatment 2 (tannin supplement at 0.5% DM, vegetable oil at 3% DM) has the highest average raw energy intake -17,488.3 Kcal / day, equivalent to 73.22 MJ / day. The control (without tannin and vegetable oil supplement) has the lowest average energy intake -16,741.6 Kcal / day, equivalent to 70.09 MJ / day. It's mean that the process of synthesizing methane and emission also lost some energy of experimental cattle.

Calculation of raw energy emitted via methane by the method of Brouwer (1965) 1 liter of methane is equivalent to 0.04 MJ of raw energy. As can be seen that: the results from Treatment 2 have the lowest amount of energy lost through methane emissions: 6.73 MJ/day, corresponding to 9.19% of raw energy received. The control (without supplementation) has the highest energy loss through methane emissions: 7.24 MJ/day, equivalent to 10.32% of the gross energy received ($P<0.05$). The energy loss through reduced methane emissions in the experiment in Treatment 1 is 7.04 MJ/day.

Our results was higher than those reported by Tran Hiep et al. (2016) when adding cotton oil in dairy feed to the ratio of raw energy lost through methane emissions from 4.17-6.28%, when adding oil at 3% DM, the lowest percentage of energy loss ($P<0.05$). Johnson and Johnson et al. (1995) and Johnson and Ward (1996) found that as feed intake increased, the intensity of methane emissions decreased. However, the author also pointed out that the close relationship between dietary digestibility, intake and methane emissions has not been proven. The author also concludes that there is no correlation between the rate of digestible raw energy (GE) and the rate of GE lost as methane.

CONCLUSION

Different tannin and vegetable oil supplements have a significant effect on the level of methane emissions of experimental cattle, as the supplement level increases, the methane emissions tended to decrease ($P<0.05$). The emission intensity of CH_4 tended to decrease with increasing dietary supplement of tannin and oil, however, when supplementation is at high level, the emission intensity tends to increase.

Use diets supplemented with 0.5% DM of tannin, 1.5% DM of vegetable oil to raise cattle for the best growth. Moreover, it has a good effect by decrease significantly methane gas emissions, contributing to reducing environmental pollution from livestock production.

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