Dietary supplementation of medicinal herbs with total mixed ration to mitigate enteric methane emission in sheep

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Abstract
The purpose of this study was to determine the effects of pineapple wastes (Ananas comosus), garlic leaves (Allium sativum), moringa leaves (Moringa oleifera) and their combination on growth, plasma metabolites, meat characteristics and enteric methane emission in sheep. A total of 15 sheep (initial BW: 8.3±0.2 kg; age: ~1 year; non-descript indigenous to Bangladesh) were randomly assigned to one of five dietary treatments with three sheep per treatment in a completely randomized design. Dietary treatments were: (1) CL-diet: a total mixed ration (TMR) pellet based on roadside grass and concentrates with a CP–16.72% and ME–10.7 MJ/kg; (2) PW-diet: CL-diet + 10g DM of pineapple waste (peels, cores, tops, and leaves)/day; (3) GL-diet: CL-diet + 10g DM of garlic leaves/day; (4) ML-diet: CL-diet + 10g DM of moringa leaves /day; (5) HM-diet: CL-diet + 10g DM of herbal mixture (3g pineapples wastes + 3g garlic leaves + 4g moringa leaves)/day. Weekly live weight gain and plasma metabolites did not show any significant variation among the treatments. Compared to the CL-diet group, the herbal supplemented group had 18–34% lower abdominal fat content, and the lowest value was found in the GL-diet group. Furthermore, the methane emission (g/day/sheep) was reduced by 5–13% in herb supplemented groups compared to the CL-diet group. Additionally, the lamb fed GL-diet emitted the lowest enteric methane (21.26 vs 24.07 g/kg DMI) compared to other experimental groups. Overall, garlic and moringa leaves could be added to TMR to minimize fat accumulation and enteric methane emission from sheep.

Keywords: medicinal herbs, bioactive components, enteric methane, plasma metabolites, sheep

Introduction
Concerns regarding the environmental impacts of ruminants farming are emergent. After CO₂, CH₄ is the second most abundant greenhouse gas, accounting for 16% of worldwide greenhouse gas emissions (Edenhofer, 2014). As part of their normal digestion processes, livestock emits CO₂ and CH₄. CO₂ generated by livestock is not considered a net contribution to climate change since ruminants consume plants that utilize CO₂ during photosynthesis (Steinfeld et al., 2006). Moreover, CH₄ once released into the atmosphere, will endure from 9-15 years and trap 25 times more radiation than CO₂ (Ma et al., 2019). Livestock husbandry is an important contributor of CH₄ in the atmosphere. Around 95.5% of CH₄ production in ruminants comes from feed fermentation in the rumen, which results in a loss of 2.3-10.8% of feed energy depending on the food and animal (Guyader et al., 2014; Ma et al., 2019). Sheep generate an average daily 22.15 g CH₄, with around 7-8% of total CH₄ production coming from microorganisms in the rumen and gut fermenting carbohydrates (McGinn et al., 2006 and Nguyen et al., 2018).

Researchers seek ways to reduce enteric methane emissions from sheep while also improving mutton quality through dietary manipulation. Medicinal herbs like hop (Humulus lupulus) having polyphenolic compounds could enhance rumen propionate production (Al-Mamun et al., 2011) and thereby mitigate methane production in ruminants (Narvaez et al., 2011). From economic, safety, and value addition to mutton aspects, phytochemicals from phytogenic feed additives are more sustainable in reducing enteric methane emission than other dietary strategies implications (Beauchemin et al., 2008, Rahman et al., 2021). Tannin as a phytochemical inhibits the growth and development of rumen methanogenic and protozoa which helps to reduce methane production up to 55% (Bodas et al., 2012). Saponin, a bioactive compound found
in herbs, has the potentiality to reduce the multiplication of ruminal bacterial and fungal species and diminish the H2 production for methanogenesis in the rumen, thus mitigating CH4 production (Bodas et al., 2012; Patra et al., 2009). Pineapple waste (PW) consists of the residual pulp, peels, stem and leaves used as a small and large ruminant feed additive, respectively (Schieber et al., 2001; Sruamisri, 2007). PW is an excellent source of phytochemicals that have the ability to represent a strong antioxidant (Oliveira et al., 2009). Moringa (Moringa oleifera) leaves are widely found in tropical and subtropical regions across the globe. They are utilized as supplements in animal feed because of their greater protein, vitamin, and mineral contents (Zhang et al., 2018). It also contains saponins, tannins, and polysaccharides, which have anti-inflammatory, antioxidant, and antibacterial properties (Mendieta-Araica et al., 2011; Azzaz et al., 2016). Garlic (Allium sativum) leaves also consist of greater amounts of flavonoid and polyphenolic components (Chung, 2006). Simultaneously, it includes a number of sulphur compounds (Martins et al., 2016) that influence rumen microbial fermentation and increase sheep performance (Panthee et al., 2017). Redoy et al. (2020) illustrated that sheep receiving 10g DM from garlic leaves significantly improved the antioxidants, immune status and reduced abdominal fat retained. Garlic enriched with 4.8% tannins and 4.3% saponins, which has the potentiality to reduce the enteric methane emission in sheep (Ali and Ebrahim, 2019).

So, the present study was designed to assess the supplementation of pineapple waste, moringa, and garlic leaves and their combination in the total mixed ration (TMR) on growth performance, plasma metabolites, meat characteristics and enteric methane emission in sheep.

Materials and Methods

Animals, diets and sampling

Fifteen non-descript sheep (indigenous to Bangladesh) having initial body weight 8.3±0.2 kg were randomly allocated five dietary treatment groups (3 sheep in each dietary group) in a complete randomized design. Dietary treatments were: (1) CL-diet: a TMR pellet based on roadside grass and concentrates with a CP–16.72% and ME–10.7 MJ/kg; (2) PW-diet: CL-diet + 10g DM of pineapple waste (peels, cores, tops, and leaves)/day; (3) GL-diet: CL-diet + 10g DM of garlic leaves/day; (4) ML-diet: CL-diet + 10g DM of moringa leaves /day; (5) HM-diet: CL-diet + 10g DM of herbal mixture (3g pineapples wastes + 3g garlic leaves + 4g moringa leaves)/day. Feed was offered once daily at 09:00h, and fresh drinking water was available ad libitum. Methane production was measured in vitro after adaptation to these rations. Rumen pH was measured continuously with the view to prevent acidosis. The total experimental period was 60 days, of which the sampling was performed during days 54-60.

Table 1: Ingredients and nutritive value of ration, pineapple (Ananas comosus), garlic (Allium sativum), and moringa (Moringa oleifera )

| Ingredients       | g/kg |
|-------------------|------|
| Local roadside grass¹ | 647  |
| Broken maize      | 89   |
| Wheat bran        | 193  |
| Mustard oil cake  | 55   |
| Salt              | 16   |

| Nutritive value (g/kg) | Ration | Pineapple | Garlic | Moringa |
|------------------------|--------|-----------|--------|---------|
| Dry Matter             | 216    | 159       | 105    | 145     |
| Crude Protein          | 162    | 65        | 248    | 237     |
| Crude Fibre            | 225    | 220       | 187    | 118     |
| Ether Extract          | 32     | 35        | 45     | 38      |
| Ash                    | 93     | 45        | 155    | 90      |
| In vitro organic matter digestibility | 597    | 603       | 663    | 684     |

¹road side grass predominating Axonopus compressus, Panicum repens, Imperata cylindrica, Cynodon dactylon, Cynodon dactylon, Cyperus rotundus species.
Sampling and chemical analysis
Feed samples and leftovers, if any, were collected every seven days interval throughout the experiment. Proximate components of ration, pineapple waste, garlic and moringa leaves were measured in accordance with the guideline of AOAC, (2005). Following proper thawing, plasma was analysed for glucose, triglycerides (TG), total cholesterol (TC), and high-density lipoprotein (HDL-C) using different enzymatic kits (HUMAN, China) in an Urit 810 bio-analysers (URIT Medical Electronic Group Co., Ltd., China) according to the manufacturer's guidelines. Plasma urea was quantified using a urea kit (HUMAN, China) in kinetic mode on an Urit 810 bio-analysers. The methane emission was measured according to the equation developed by Matt Bell et al. (2016) using digestible organic matter (DOMD), ether extract (EE) (both g/kg DM) and feeding level above maintenance intake:

\[ \text{CH}_4 \text{ (g/kg DM intake)} = 0.046 (\pm 0.001) \times \text{DOMD} - 0.113 (\pm 0.023) \times \text{EE} - 2.47 (\pm 0.29) \times (\text{feeding level} - 1) , \]

with concordance correlation coefficient (CCC) = 0.655 and RMSPE = 14.0%.

Statistical analysis
All raw data were arranged in Microsoft Excel software and analyzed in IBM SPSS 20 (USA) statistical tools using one way ANOVA. The significance of differences among means was determined using the Duncan’s Multiple Range Test, and the differences at \( P < 0.05 \) were considered statistically significant. The following model was used in this study.

\[ Y_{ij} = \alpha + T_i + \epsilon_{ij} \]

Where, \( Y_{ij} \), all observed parameters are the dependent variables and \( T_i \), the influence of dietary treatments (pineapple waste, garlic, moringa leaves and their combination) are the independent variable. \( \epsilon_{ij} \) is the error to the \( i \)th replication of the \( i \)th treatments and normally distributed with zero mean and constant variance.

Results

Growth performance
Lamb fed local roadside grass and concentrate based control diet (CL-diet) supplemented with 10g moringa leaves DM/d/lamb (ML-diet) had maximum growth performance compared to other supplemented groups (Figure 1). Growth performance of CL-diet group had the lowest \( (P < 0.05) \) throughout the experimental period. The growth of lamb fed a PW-, GL-, or HM-diets was comparable to each other’s, but was lower than that of the ML-diet group.

\[ \text{Figure 1: Effect of different dietary forage herbs supplementation on growth performance of sheep} \]

Plasma metabolites
The concentration of blood metabolites of lamb was shown in Table 2. However, no significant differences were observed in glucose, TG, cholesterol, HDL, LDL, urea, BUN values of lamb in all dietary supplemented groups. But numerically, the concentration of TG, cholesterol and LDL of lamb was the lowest in GL-diet group than control and other herb supplemented group where HDL concentration was maximum in the lamb of garlic leaves supplemented group (GL-diet). But blood urea level was the highest in CL-diet group and the lowest in GL-diet. Cholesterol and TG concentrations were 11% and 19% lower in GL-diet group than CL-diet, respectively. About 20% low LDL was observed in the blood of lamb where garlic leaves were supplemented group (GL-diet).

Meat characteristics
Significant effect of dietary supplementation of medicinal herbs was not present in the percentage of meat and bone but in case of fat percent, which was \( (P < 0.05) \) maximum 0.55% in lamb of CL-diet group. The lowest fat percent was observed in GL-diet group (0.36) which was about 35% lower than the CL-diet group. In ML-diet and HM-diet groups value for fat percent was close and was more or less 18% lower than control group.
Serum metabolites

Table 2: Effect of different dietary forage herbs supplementation on plasma metabolites in sheep

| Parameters          | CL- diet | PW-diet | GL-diet | ML-diet | HM-diet | SEM | P value |
|---------------------|----------|---------|---------|---------|---------|-----|---------|
| Glucose (mg/dL)     | 73.46    | 63.14   | 62.91   | 63.09   | 63.07   | 3.22| 0.06    |
| Urea (mg/dL)        | 8.87     | 6.17    | 6.13    | 6.14    | 6.18    | 1.04| 0.17    |
| BUN (mg/dL)         | 20.6     | 21.89   | 22.01   | 21.83   | 21.93   | 1.33| 0.74    |
| TG (mg/dL)          | 70.21    | 59.15   | 57.03   | 59.01   | 59.11   | 2.18| 0.14    |
| Cholesterol (mg/dL) | 82.12    | 74.78   | 73.08   | 74.12   | 74.21   | 2.41| 0.08    |
| HDL (mg/dL)         | 49.47    | 53.49   | 54.04   | 54.01   | 53.95   | 1.88| 0.29    |
| LDL (mg/dL)         | 20.21    | 16.81   | 15.98   | 15.02   | 15.61   | 1.80| 0.36    |

*BUN: blood urea nitrogen; TG: triglycerides; HDL: high density lipoprotein; LDL: low density lipoprotein*. CL-diet: local roadside grass and concentrate mixture with a CP=16.72% and ME=10.7 MJ/kg; PW-diet: CL-diet +10g pineapple peels, leaves and nonedible part DM/d/sheep; GL-diet: CL-diet + 10g garlic leaves DM/d/sheep; ML-diet: CL-diet + 10g moringa leaves DM/d/sheep; HM-diet: CL-diet +10g herbs mixture DM/d/sheep (3.0g DM pineapple + 3.0g DM garlic leaves + 4.0g DM moringa leaves); SEM: standard error of means. *a,b,c* Means in the same row with no common superscript differ significantly (*P* < 0.05).

**Methane emission**

There was significant variation in methane emission rate in dietary supplemented group. Methane emission (g/kg DMI) was 21.26 in GL-diet and 21.43 ML-diet which was 11.67%, 10.96% lower than CL-diet group, respectively (Table 4). Methane emission on the basis of DMI was also 22.92 g/kg DMI in PW-diet and 22.01 g/kg DMI which value was close to GL-diet and ML-diet group. Individual methane emission per day of lamb was maximum (p < 0.05) in CL-diet group (8.19 g/day/lamb). 4.76%, 12.57%, 13.19%, 11.84% lower methane emission (g/day/lamb) was noticed in PW-diet, GL-diet, ML-diet and HM-diet group than CL-diet group.

**Discussion**

**Growth performance**

It is generally agreed that herbs can improve energy utilization, resulting in increased growth, by: i) improving fibre digestion; ii) reducing methanogenesis and (iii) type, level and nature of plant secondary metabolites (Cieslak et al., 2014). Both moringa and garlic leaf supplemented lamb demonstrated a similar response in terms of enteric methane emission (Table 4), implying that both groups might exhibit comparable growth performance. However, the lamb showed a 5% greater growth response in the ML-diet group than in the GL-diet group, indicating that some other potential factors may have an effect. The predominance of tannin in both herbs had a distinct molecular weight and chemical structure that might not matter to methanogenic bacteria but may matter to fibrinolytic bacteria (Animut et al., 2008; Wang et al., 2009). The findings of Cieslak et al. (2014) and Khiaosa-Ard et al. (2009) in sheep, *in vivo* and *in vitro* experiments, respectively, support this argument. In addition, moringa leaf's flavonoid profile is superior to those of other herbs supplemented in this experiment (Akbarpour et al., 2021; Siskawardani et al., 2021; Lourenco et al., 2021). The flavonoid component, a natural antioxidant, may trap a greater number of free radicals in lamb fed the ML-diet than others, resulting in less stress in the former groups and promoting growth (Sahoo et al., 2021; Zhang et al., 2021).

**Plasma metabolites**

Herbal supplementation in lamb yielded inconsistent findings in plasma metabolites (Redoy et al., 2020; Reza et al., 2021; Chaves et al., 2008; Shuvo et al., 2017). In this experiment, pineapple waste, garlic and moringa leaves supplementation had no effect on plasma glucose concentration, which is consistent with the findings of Redoy et al. (2020), Panthee et al. (2017) and Akanmu et al. (2020). However, existing research findings has demonstrated that pineapple waste, garlic and moringa have a considerable effect on the plasma lipid profile; however, our trial failed to reflect this (Ososanya et al., 2015; Redoy et al., 2020; Akanmu et al., 2020).
Herbs as anti-methanogenic agent for sheep

Table 3: Effect of different dietary forage herbs supplementation on meat characteristics of lambs

| Parameters (g/100g) | Treatments | SEM | P value |
|---------------------|------------|-----|---------|
|                     | CL-diet    | PW-diet | GL-diet | ML-diet | HM-diet |       |
| Meat                | 12.44      | 13.08  | 13.09  | 12.89  | 12.99   | 0.89  | 0.73   |
| Bone                | 18.70      | 19.47  | 19.43  | 19.01  | 19.02   | 0.55  | 0.50   |
| Fat                 | 0.55a      | 0.39bc | 0.36c  | 0.43b  | 0.45b   | 0.05  | 0.02   |

**CL-diet**: local roadside grass and concentrate mixture with a CP-16.72% and ME-10.7 MJ/kg; **PW-diet**: CL-diet +10g pineapple peels, leaves and nonedible part DM/d/sheep; **GL-diet**: CL-diet + 10g garlic leaves DM/d/sheep; **ML-diet**: CL-diet + 10g moringa leaves DM/d/sheep; **HM-diet**: CL-diet + 10g herbs mixture DM/d/sheep (3.0g DM pineapple + 3.0g DM garlic leaves + 4.0g DM moringa leaves); **SEM**: standard error of means; *a,b,c* Means in the same row with no common superscript differ significantly (*P* < 0.05).

Table 4: Effect of different dietary forage herbs supplementation on methane emission from sheep

| Parameters (g/kg DMI) | Treatments | SEM | P value |
|-----------------------|------------|-----|---------|
| Methane emission      | CL-diet    | PW-diet | GL-diet | ML-diet | HM-diet |       |
| (g/kg DMI)            | 24.07      | 22.92  | 21.26  | 21.43  | 22.01   | 0.761 | 0.037  |

| Parameters (g/day/lamb) | Treatments | SEM | P value |
|-------------------------|------------|-----|---------|
| Methane emission        | CL-diet    | PW-diet | GL-diet | ML-diet | HM-diet |       |
| (g/day/lamb)            | 8.19       | 7.80   | 7.16   | 7.11   | 7.22    | 0.246 | 0.037  |

**CL-diet**: local roadside grass and concentrate mixture with a CP-16.72% and ME-10.7 MJ/kg; **PW-diet**: CL-diet +10g pineapple peels, leaves and nonedible part DM/d/sheep; **GL-diet**: CL-diet + 10g garlic leaves DM/d/sheep; **ML-diet**: CL-diet + 10g moringa leaves DM/d/sheep; **HM-diet**: CL-diet + 10g herbs mixture DM/d/sheep (3.0g DM pineapple + 3.0g DM garlic leaves + 4.0g DM moringa leaves); **SEM**: standard error of means; *a,b,c* Means in the same row with no common superscript differ significantly (*P* < 0.05).

Though the herbal supplemented group’s cholesterol content was substantial lower (*P*=0.08) than the control group. This finding can be justified by the fact that all three herbs possess a diverse phytochemical profile that has been shown to influence ruminal ecology, so enhancing propionic acid production and thereby promoting gluconeogenesis and insulin secretion (Oliveira et al., 2009; Martins et al., 2016; Al-Mamun et al., 2017; Zhang et al., 2018). Additionally, this increased gluconeogenesis might result in a positive energy balance in the experimental animal, resulting in decreased hepatic conversion to triglycerides and ketones in herbal supplemented group.

Meat characteristics

The qualities of lamb meat are determined by both genetic and non-genetic factors. Among non-genetic factors, nutrition has the greatest influence on carcass morphology, physicochemical properties, and meat quality (Odhaib et al., 2021). Additionally, herb supplementation has a significant effect on numerous tissue components, resulting in alterations in the flavor, mutton fat, and fatty acid composition (Odhaib et al., 2021). The lower fat percentage in the herb supplemented groups was consistent with the findings of Redoy et al. (2020) and Qwele et al. (2013), who reported that supplementing lamb and goat with garlic and moringa leaves dramatically reduced the meat fat percentage. Allicin in garlic leaves inhibited adipogenesis and promoted lipolysis in mice (Shi et al., 2019). Additionally, this component stimulates white adipocyte browning and inhibits the Krüppel-like factor 15 signaling cascade, which results in decreased fat accumulation (Shi et al., 2019). Additionally, the high flavonoid content of moringa leaf stimulates leptin expression, which alters cholesterol catabolism and results in decreased fatty acid synthesis and increased fatty acid -oxidation in mice (Bais et al., 2014), which may account for the lower fat percentages in the GL- and ML-diet groups.
Methane emission

During this experiment, lamb supplemented with herbs exhaled 9% less methane per kg DMI than lamb without herbs. The influence of fresh herbs, extracts, or herbal preparations on enteric methane emission in small ruminants, on the other hand, yielded inconclusive findings (Ahmed et al., 2021; Abarghuei et al., 2021; Diaz-Medina et al., 2021; Faniyi et al., 2021; Loza et al., 2021). These discrepancies could be explained by the variation of secondary metabolites in herbs, more especially the tannin content, which has a direct effect on reducing enteric methane production (Loza et al., 2021). The increased tannin content (twofold) of moringa and garlic leaves compared to pineapple waste resulted in a 7 percent reduction in lamb methane production per kg DMI in this experiment (Muhammad et al., 2019; Rabiu et al., 2018; Sultana et al., 2014). Additionally, Faniyi et al. (2021) observed that moringa supplementation significantly decreased methane production as a percentage of total gas production in sheep when compared to control. Additionally, Torres-Fraga et al. (2020) reported the enhanced anti-methanogenic activity of garlic leaf compared to alfalfa hay as solely or partly replacement in beef cattle. All the above-mentioned explanations are in favour with our current experimental findings. The increased tannin content of moringa and garlic leaves might result in a decrease in the overall number of protozoa, entodiniomorphs, and holotrichs in supplemented lamb, resulting in decreased methane production (Cieslak et al., 2014). Additionally, these herbs vastly enhance propionic acid production during ruminal fermentation, which may lead to a decrease in the H₂ sink for methane formation (Abdel-Raheem et al., 2021; Kewan et al., 2021).

Conclusion

To begin, none of the three herbs individually or in combination had a significant influence on growth performance or plasma metabolites under the present experimental conditions. Second, when compared to control, herbal supplementation decreased fat accumulation in sheep. Thirdly, and perhaps most significantly, garlic and moringa leaves significantly decreased enteric methane emission from sheep, which is critical for climate-smart sheep production. Finally, based on the results of this feeding trial, it could be concluded that garlic and moringa leaves can be added to TMR diet with the goal of reducing enteric methane emission from sheep and increasing lean mutton production.

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Conflict of interest

The authors would like to declare that there is no conflict of interest.

References

Abarghuei MJ and AZM Salem (2021). Sustainable impact of pulp and leaves of Glycyrrhiza glabra to enhance ruminal biofermentability, protozoa population, and biogas production in sheep. Environmental Science and Pollution Research 2021:1-11.

Abdel-Raheem SM and EH Hassan (2021). Effects of dietary inclusion of Moringa oleifera leaf meal on nutrient digestibility, rumen fermentation, ruminal enzyme activities and growth performance of buffalo calves. Saudi Journal of Biological Sciences 28(8), 4430-4436.

Ahmed E, B Batbekh, N Fukuma, D Kand, M Hanada and T Nishida (2021). A garlic and citrus extract: impacts on behavior, feed intake, rumen fermentation, and digestibility in sheep. Animal Feed Science and Technology 115007.

Akanmu AM, A Hassen and FA Adejoro (2020). Haematology and serum biochemical indices of lambs supplemented with Moringa oleifera, Jatropha curcas and Aloe vera leaf extract as anti-methanogenic additives. Antibiotics 9, 601-608.

Akbarpour A, B Kavoosi, M Hosseinifarhadi, S Tahmasebi and S Gholipour (2021). Evaluation of yield and phytochemical content of different Iranian garlic (Allium sativum L.) ecotypes. International Journal of Horticultural Science and Technology 8(4): 1-16.

Ali M and IS Ibrahim (2019). Phytochemical Screening and Proximate Analysis of Garlic (Allium sativum). Archive of Organic and Inorganic Chemical Science 4(1):180.

Allam SM, GE Aboul-Fotouh, GM El-Garhy and O Gamal (2015). Use of moringa leaves (Moringa oleifera) in fattening lambs rations. Egyptian Journal of Nutrition and Feeds 18(2):11-17.

Al-Mamun M, A Saito and H Sano (2011). Effect of ensiled hop (Humulus lupulus L.) residues on plasma acetate turnover rate in sheep. Animal Science Journal 82: 451–455.

Al-Mamun M, K Shibuya, M Kajita, Y Tamura and H Sano (2017). Responses of plasma glucose metabolism to exogenous insulin infusion in sheep fed forage herb and exposed to heat. Animal 11(8): 1287-1294.

Animut G, R Puchala, AL Goetsch, AK Patra, T Sahlu, VH Varel and J Wells (2008). Methane emission by goats consuming diets with different levels of...
condensed tannins from lespedeza. Animal Feed Science and Technology 144:212-227.

Azzaz HH, ES Farahat, TA Morsy, HA Aziz, FI Hadhoud and MS Abd-Alla (2016). *Moringa oleifera* and *Echinacea purpurea* as supplements for Rhamani lactating ewes' diets and their effect on rumen characteristics, nutrients digestibility, blood parameters, milk production, composition and its fatty acid profile. Asian Journal of Animal and Veterinary Advances 11(1):684-692.

Bais S, GS Singh and R Sharma (2014). Antiobesity and hypolipidemic activity of *Moringa oleifera* leaves against high fat diet-induced obesity in rats. Advances in Biology 2014,1-9.

Bhatta R, Y Uyeno, K Tajima, A Takenaka, Y Yabumoto, I Nonaka and M Kurihara (2009). Difference in the nature of tannins on in vitro ruminal methane and volatile fatty acid production and on methanogenic archaea and protozoal populations. *Journal of Dairy Science* 92(11): 5512-5522.

Chaves AV, K Stanford, MER Dugan, LL Gibson, TA McAllister, TF Van Herk and C Benchaar (2008). Effects of cinamaldehyde on growth performance and carcass characteristics of growing lambs. *Livestock Science* 117(2-3): 215-224.

Cieslak A, P Zmora, E Pers-Kamczyc, A Stochmal, A Sadowinska, AZ Salem and M Szumacher-Strabel (2014). Effects of two sources of tannins (*Quercus L.* and *Vaccinium vitis ideae L.*) on rumen microbial fermentation: an in vitro study. *Italian Journal of Animal Science* 13(2): 3133.

Diaz-Medina LK, V Collín-Navarro, CM Arriaga-Jordán, L Brunett-Pérez, BR Vázquez-de-Aldana and JG Estrada-Flores (2021). In vitro nutritional quality and antioxidant activity of three weed species as feed additives for sheep in the Central Highlands of Mexico. *Tropical Animal Health and Production* 53(3): 1-9.

Edenhofer O, editor. Climate change 2014: mitigation of climate change. *Cambridge University Press*; 2015 Jan 26

Faniyi TO, MK Adewumi, AA Jack, MJ Adegbeye, MM Elghandour, A Barbabosa-Pileo and AZ Salem (2021). Extracts of herbs and spices as feed additives mitigate ruminal methane production and improve fermentation characteristics in West African Dwarf sheep. *Tropical Animal Health and Production* 53(2): 1-8.

Guyader J, M Eugènè, P Nozière, DP Morgavi, M Doreau and Martin C (2014). Influence of rumen protozoa on methane emission in ruminants: a meta-analysis approach 1. *Animal* 8(11):1816-1825.

Kewan KZ, MM Ali, BM Ahmed, SA El-Kolty and UA Nayel (2021). The effect of yeast (*Saccharomyces cerevisiae*), garlic (*Allium sativum*) and their combination as feed additives in finishing diets on the performance, ruminal fermentation, and immune status of lambs. *Egyptian Journal of Nutrition and Feeds* 24(1): 55-76.

Khiaosa-Ard R, SF Bryner, MRL Scheeder, HR Wettstein, F Leiber, M Kreuzer and CR Soliva (2009). Evidence for the inhibition of the terminal step of ruminal ω-linolenic acid biohydrogenation by condensed tannins. *Journal of Dairy Science* 92(1): 177-188.

Lourenço SC, DA Campos, R Gómez-García, M Pintado, MC Oliveira, DI Santos, VD Alves (2021). Optimization of natural antioxidants extraction from pineapple peel and their stabilization by spray drying. *Foods* 10(6): 1255.

Mendieta-Araica B, R Spörndly, N Reyes-Sánchez and E Spörndly (2011). *Moringa (Moringa oleifera)* leaf meal as a source of protein in locally produced concentrates for dairy cows fed low protein diets in tropical areas. *Livestock Science* 37(1-3):10-17.

Mendis K, Y Wang, Z Xu and T McAllister (2011). Effects of hops on in vitro ruminal fermentation of diets varying in forage content. *Livestock Science* 138:193-201.

Nguyen SH, HD Nguyen, G Bremmer and RS Hegarty (2018). Methane emissions and productivity of defaunated and refaunated sheep while grazing. *Small Ruminant Research* 161:28-33.

Odhaib KJ, QN Al-Hajjar and MH Alallawee (2021). Incorporation of herbal plants in the diet of ruminants: effect on meat quality. *The Iraqi Journal of Veterinary Medicine* 45(1): 22-30.

Oliveira AC, IB Valentim, CA Silva CA, EJH Bechara, MP Barros, CM Mano and MOFG Goulart (2009). Total phenolic content and free radical scavenging activities of methanolic extract powders of tropical fruit residues. *Food Chemistry* 115: 469-475.

O sosanya TO, MK Adegbe and KB Jinadu (2014). Impact of pineapple waste silage on intake, digestibility and fermentation patterns of West African Dwarf sheep. *African Journal of Biotechnology* 13(25): 2575-2581.

Panthee A, A Matsuno and Al-Mamun (2017). Effect of feeding garlic leaves on rumen fermentation, methane emission, plasma glucose kinetics, and nitrogen utilization in sheep. *Journal of Animal Science and Technology* 59(1), 1-9.

Qwele K, A Hugo, SO Oyedemi, B Moyo, PJ Masika and V Muchenje (2013). Chemical composition, fatty acid content and antioxidant potential of meat from goats supplemented with *Moringa (Moringa oleifera)* leaves, sunflower cake and grass hay. *Meat Science* 93(3):455-562.

Rabiu Z, FU Maigari, U Lawan and ZG Mukhtar (2018). Pineapple waste utilization as a...
sustainable means of waste management. In: Zakaria Z. (eds) Sustainable technologies for the management of agricultural wastes. Applied Environmental Science and Engineering for a Sustainable Future. Springer, Singapore.

Redoy MRA, AAS Shuvo, L Cheng and M Al-Mamun (2020). Effect of herbal supplementation on growth, immunity, rumen histology, serum antioxidants and meat quality of sheep. Animal 14(11): 2433-2441.

Reza MM, MRA Redoy, MA Rahman, S Ety, MA Alim, L Cheng and M Al-Mamun (2021). Response of plantain (Plantago lanceolata L.) supplementation on nutritional, endo-parasitic, and endocrine status in lambs. Tropical Animal Health and Production 53(1), 1-7.

Sahoo A, S Sarkar, B Lal, P Kumawat, S Sharma and K De (2021). Utilization of fruit and vegetable waste as an alternative feed resource for sustainable and eco-friendly sheep farming. Waste Management 128: 232-242.

Schieber A, FC Stintzing and R Carle (2001). By-products of plant food processing as a source of functional compounds-recent developments. Trends in Food Science and Technology 12: 401-413.

Shi X, X Zhou, X Chu, J Wang, B Xie, J Ge and G Yang (2019). Allicin improves metabolism in high-fat diet-induced obese mice by modulating the gut microbiota. Nutrients 11(12): 2909.

Shuvo AAS, MRA Redoy and M Al-Mamun (2017). Effect of herbal supplementation to TMR diet on lipid profile of blood and meat in sheep. In: Proceedings of VIII International Scientific Agriculture Symposium, 5–8 October 2017, Jahorina, Bosnia-Herzegovina, pp. 2106–2111.

Siskawardani DD, S Winarsih and K Khawwee (2021). The antioxidant activity of Kelor (Moringa oleifera Lam.) leaves based on drying method. Jordan Journal of Biological Sciences 14(2).

Sruamisri S (2007). Agricultural wastes as dairy feed in Chiang Mai. Animal Science Journal 78: 335-341.

Sultana N, AR Alimon, KS Haque, AQ Sazili, H Yaakub and SMJ Hossain (2014). The effect of cutting interval on yield and nutrient composition of different plant fractions of Moringa oleifera tree. The Journal of Food, Agriculture and Environment 12(2): 599-604.

Torres-Fraga K, J Páez-Lerma and G Pámanes-Carrasco (2020). Substitution of garlic leaves to alfalfa hay and its effect on in vitro ruminal fermentation. Abanico Veterinario 10(1):1-11.

Wang Y, TW Alexander, TA Mcallister (2009). In vitro effects of phlorotannins from Ascophyllum nodosum (brown seaweed) on rumen bacterial populations and fermentation. Journal of the Science of Food and Agriculture 89:2252-2260.

Zhang K, Q Qian, Y Mao, Y Xu, Y Yang, Y Chen and X Wang (2021). Characterization of growth phenotypes and gastrointestinal tract microbiota in sheep fed with caragana. Journal of Applied Microbiology 2021.

Zhang T, B Si, K Deng, Y Tu, C Zhou and Q Diao (2018). Effects of feeding a Moringa oleifera rachis and twig preparation to dairy cows on their milk production and fatty acid composition, and plasma antioxidants. Journal of the Science of Food and Agriculture 98(2):661-666.