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Research Article
Methane Production Potential and Nutritive Value of Indigenous Browses from mid Rift Valley of Ethiopia

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Abstract

Objective: This study was conducted to evaluate the chemical composition, *in vitro* gas and methane (CH_4) production of eight tannin-containing browses species from Rift Valley of Ethiopia. **Materials and Methods:** Leaves of *Acacia seyal*, *Acacia senegal*, *Acacia tortilis*, *Prosopis juliflora*, *Millettia ferruginea*, *Vernonia amygadalina*, *Croton macrostachyus* and *Cordia africana* were collected, oven-dried and ground to 1.0 mm for *in vitro* gas and chemical analysis. General linier model procedure of SAS, Version 9.2 was used for statistical analysis. **Results:** The highest crude protein (CP) contents of the browses ranged from 13-29% was observed for *Acacia senegal* and lowest for *Acacia seyal*. The nutrient detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were highest for *Cordia africana* and lowest for *Acacia seyal*. The CT content for *Acacia seyal*, *Acacia tortilis* and *Millettia ferruginea* were higher ($p<0.05$) than the remaining species. *Acacia seyal* produced the highest gas volume and *Prosopis juliflora* produced the lowest. The lowest ($p<0.05$) CH_4 percentage of total gas was recorded from *Acacia seyal*. The CH_4 percentage from *Cordia africana* was higher ($p<0.05$) than *Acacia seyal* and *Acacia tortilis*. Inclusion of polyethylene glycol (PEG) significantly improved gas production and organic matter digestibility (OMD) of browses. **Conclusion:** The high gas production and OMD of *Acacia seyal*, coupled with its lowest percent of CH_4 of total gas produced during fermentation would make the browse potential supplement of low quality roughages while reducing enteric CH_4 emissions.

Key words: Chemical composition, condensed tannin, *in vitro* gas production, polyethylene glycol

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Tropical browses are believed to have high protein content, which makes them promising supplements to crop residues and poor quality natural pasture based diets. Tropical browses with high concentrations of CT have been reported to decrease methane emissions directly by inhibiting methanogens' growth and indirectly by decreasing rumen fiber degradability and hence, a reduced H_2 availability for CH_4 production[1]. Research results indicate that tannin-rich tropical browses decreased CH_4 production and, therefore, could be strategically used in diets for decreasing CH_4 emissions from ruminants[1].

In this regard, tannins of lower molecular weights were found to be more effective against methanogens than tannins of higher molecular weight[1]. However, there is huge diversity in tannin structure and concentrations among the browses[2]. This indicates that tannins from different plants might show different response in gas production, digestibility and methane production. Hence, it is important to investigate variations among indigenous browses in terms of gas production, forage digestibility and CH_4 production. Several studies on nutritional values of imported or improved browses have been conducted in different parts of Ethiopia[3-5]. Previous studies have given more emphasis to introduced/improved species than native vegetation in Ethiopia. Little information is available about the potential nutritive value of indigenous browses. Kechero and Janssens[6] evaluated nutritive value of leaves of trees and shrubs from Gilgel Gibe catchments of Southwest Ethiopia. Merga *et al.*[7] studied browse species from Borana rangeland for their anti nutritional factors. Shenkute *et al.*[8] studied chemical compositions of dominant browses from mid Rift Valley. However, *in vitro* gas and methane production potentials of indigenous browse from mid Rift Valley are not yet studied. Moreover, site specific evaluation of indigenous browse species can contribute to further establishment, adaptation and utilization as fodder[6]. It is, therefore, imperative to evaluate nutrient composition, *in vitro* gas and methane production potentials of the indigenous browses in order to screen and recommend best indigenous browses with optimum nutritional value and minimum methane production potentials. The present study, therefore, examines the nutrient composition, *in vitro* gas and methane production and organic matter digestibility of dominant indigenous browses from mid Rift Valley of Ethiopia.

MATERIALS AND METHODS

Description of the study area: Browses samples were collected from Mid Rift Valley area of Ethiopia. The area is located at the center of the Ethiopian Great Rift Valley extending from 7°05'-8°00'N and form 38°20'-38°50'E at an elevation ranging between 1500 -1700 m above sea level. The area is generally characterized by semi-arid and sub-humid climate with mean annual rainfall of 700-1000 mm and range of temperature varying between 20 and 26°C. The long rainy season is from June to September and short rainy season from April to May.

Sample collection: Dominant browse species from the bottom of the Rift Valley (semi-arid region) and top-land of the Rift Valley (sub-humid region) were considered for this study. Three representative sampling sites, in which the experimental browses are dominant, were purposively selected from each top and bottom land. Leaves of *Acacia seyal*, *Acacia senegal*, *Acacia tortilis*, *Prosopis juliflora* were collected from the bottom land and *Millettia ferruginea*, *Vernonia amygadalina*, *Croton macrostachyus* and *Cordia africana* were collected from the top land.

Browse leaves were sampled from 40 (5-from each species) representative trees in the study area at the end of long rainy season (end of September, 2015). During the sampling, fresh plant leaves and twigs (<3 mm) were hand plucked. The harvested samples were pooled for each individual species, shed dried and composite samples were taken, oven dried at 60°C for 48 h and ground to 1.0 mm for chemical analysis.

Chemical analysis: All chemical analysis was conducted in triplicates. Leaves of the browses were analyzed for dry matter (DM), ether extract (EE) and ash according to AOAC[9]. The NDF, ADF and ADL were determined according to the method described by Van Soest *et al.*[10]. Nitrogen was determined by Kjeldhal procedure and CP was calculated as N×6.25. Condensed tannin (CT) was determined using the method described by Makkar[11]. The CT (% in dry matter) as leucocyanidin equivalent was calculated by the equation:

$$\text{CT (\%)} = \frac{\text{A 550 nm} \times 78.26 \times \text{dilution factor}}{\text{Dry matter (\%)}}$$

Determination of *in vitro* gas and methane productions: *In vitro* gas production (GP) was measured according to the procedure described by Grant and Mertens[12]. The rumen fluid was collected from three rumen fistulated Arsi-Bale sheep fed with *Chloris gayana* grass hay *ad libitum* and supplemented with 200 g of concentrate (70% wheat bran and 30% linseed cake) daily. The rumen fluid was collected before morning feed and then prepared and purged with CO_2 to maintain anaerobic conditions[13]. After blending, the rumen fluid was transferred to a large glass beaker inside a 39°C water bath being continuously purged with CO_2 and continuously stirred as recommended by Goering and Van Soest[14]. The media solutions (buffer solution, macro and micro mineral solution) were prepared and utilized as described by Goering and Van Soest[14].

Each sample weighing about 400 mg with or without 400 mg of PEG (molecular weight, 6000) was put into 100 mL calibrated glass syringe together with 30 mL rumen liquid and culture media solution on a1:2 ratio. Syringes were incubated in a water bath at 39°C, where a transparent plastic lid with holes held the syringes upright. Two blank syringes with the rumen liquid and culture media solution were also incubated. The incubation period was 24 h. The initial gas volume was recorded as V_0 and then after at 3, 6, 12 and 24h incubation. The gas production value (mL/400 mg) at t hours was calculated as follows:

$$G_t = \frac{V_t - V_0 - G_0 \times 400}{W_s}$$

where, G_t is the gas production value (mL/400 mg) at t hours, G_0 is the gas production of blank syringes (mL), V_0 is the initial volume in mL, V_t = Volume in mL at t hours, W_s is the weight of dried sample in mg. *In vitro* organic matter digestibility (OMD) at t hours was calculated from the equation:

$$\text{OMD (\%)} = 14.88+0.889\times G_t+(0.448\times \text{CP\%})+(0.651\times \text{Ash \%})^{12}$$

Methane content was measured by absorption of carbon dioxide with 40% of NaOH[15]. The assumption of using this method was that *in vitro* gas mainly constituted methane and carbon dioxide, where the other gases produced during fermentation were insignificant. At the end of incubations (24 h) and after recording the final gas volume as V_1, the lower end of the syringe was connected to the lower end of another syringe containing 5 mL of NaOH (10 M). Then the NaOH was introduced from the latter into the incubated contents, thereby avoiding gas escape. Mixing of the contents with NaOH allowed absorption of CO_2, with the gas volume remaining in the syringe (V_2) was considered being CH_4 [15].

The percentage of $CO_2$was calculated as follow:

$$CO_2\ (\%) = \frac{V1\text{-}V2}{V1}\times 100$$

where, V_1 is the volume of gas before removal of CO_2 (mL) V_2 is the volume of methane and the other gases after removal of CO_2 (mL).

The percentage of CH_4 was estimated from the percentage of CO_2 as follows:

$$CH_4\ (\%) = 100\%\text{-}[CO_2\%+0.2\%\ (\text{other gases})]$$

Statistical analysis: Data were analyzed using general linear model (GLM) procedure of SAS, Version 9.2. One-way analysis of variance (ANOVA) was used to assess the effects of browses on nutrient composition with the following model:

$$Y_{ij} = m+B_i+e_{ij}$$

where, Y_{ij} is an observation, m is the overall mean, B_i is the effect of browses and e_{ij} is the experimental error. The effects of browses, PEG treatment and the interaction effects on *in vitro* gas and CH_4 production were analyzed by two-way ANOVA with the following model:

$$Y_{ij} = m+B_i+P_j+B\times P_{ij}+e_{ij}$$

where, Y_{ij} is an observation, m is the overall mean, B_i is the effect of browses, P_j is the effect of PEG treatment, $B\times P_{ij}$ is the interaction between species and PEG treatment and e_{ij} is the experimental error. All laboratory analyses of each browse species were conducted in triplicates. Multiple correlation analysis was conducted to determine the correlation coefficient (r) of *in vitro* fermentation parameters with chemical and CT composition of browses. The means were separated by Duncan's multiple range test. Differences between means were considered statistically significant if $p<0.05$.

RESULTS AND DISCUSSION

Chemical composition: The chemical compositions of browses are given in Table 1. All browses used in the current study had a CP content above 13% DM. *Acacia senegal* had the highest ($p<0.05$) CP content while *Acacia seyal* had the lowest ($p<0.05$). The NDF and ADF of browses varied from 13.98-48.22 and 9.39-28.98% of DM, respectively. *Cordia africana* had the highest ($p<0.05$) NDF, ADF and ADL

Table 1: Chemical composition (% DM) of leaves of indigenous browses

	OM	EE	CP	NDF	ADF	ADL	CT
Acacia tortilis	93.22^{a}	3.09^{ab}	21.76^{c}	19.48^{f}	12.63^{e}	1.79^{bc}	3.0^{a}
Acacia. seyal	93.52^{a}	3.30^{ab}	13.03^{e}	13.98^{g}	9.39^{f}	0.80^{c}	3.4^{a}
Acacia senegal	89.04^{bc}	4.38^{a}	28.81^{a}	22.04^{e}	12.07^{e}	1.79^{bc}	1.3^{b}
Prosopis juliflora	87.05^{c}	3.07^{ab}	21.33^{c}	30.86^{c}	21.13^{b}	3.16^{bc}	1.0^{b}
Millettia ferruginea	89.38^{b}	3.49^{ab}	22.46^{c}	40.77^{b}	22.42^{b}	7.44^{a}	3.0^{a}
Vernonia amygadalina	87.39^{bc}	2.86^{b}	22.51^{c}	22.08^{e}	14.35^{d}	3.65^{b}	0.4^{b}
Croton macrostachyus	87.46^{bc}	2.92^{ab}	25.94^{b}	27.72^{d}	16.51^{c}	2.97^{bc}	0.3^{b}
Cordia africana	87.03^{c}	1.23^{c}	18.44^{d}	48.22^{a}	28.98^{a}	9.66^{a}	0.3^{b}
SE	0.43	0.29	0.25	0.31	0.29	0.57	0.2

Means in the same column with different superscript differ significantly ($p<0.05$). OMO: Organic matter, EE: Ether Extract, CP: Crud protein, NDF: Neutral detergent fiber, ADF: Acid detergent fiber, ADL: Acid detergent lignin, CT: Condensed tannin

Table 2: *In vitro* gas and methane production (mL 400 mg^{-1}) and organic matter digestibility (%) of indigenous browse leaves after 24 h of incubation

	CT	Gp	OMD	CH_4	CH_4 (%)
Acacia tortilis	3.0^{a}	30.78^{d}	49.98^{c}	7.08^{bc}	25.0^{ab}
Acacia. seyal	3.4^{a}	46.84^{a}	74.87^{a}	4.64^{ef}	10.5^{c}
Acacia senegal	1.3^{b}	43.65^{ab}	70.84^{a}	8.04^{ab}	18.5^{b}
Prosopis juliflora	1.0^{b}	19.94^{e}	33.23^{d}	4.18^{f}	21.0^{ab}
Millettia ferruginea	3.0^{a}	27.24^{d}	44.65^{c}	6.87^{cd}	25.2^{ab}
Vernonia amygadalina	0.4^{b}	35.96^{c}	58.54^{b}	8.08^{ab}	22.5^{ab}
Croton macrostachyus	0.3^{b}	41.84^{b}	67.95^{a}	9.05^{a}	21.5^{ab}
Cordia africana	0.3^{b}	20.31^{e}	33.70^{d}	5.69^{de}	28.0^{a}
SE	0.2	0.98	1.5	0.24	1.6

Means in the same column with different superscript differ significantly ($p<0.05$), Condensed tannin (CT = %) Gas production (GP): mL 400 mg^{-1} DM), Organic matter digestibility (OMD = %), methane production (CH = mL 400 mg^{-1} DM) percentage of CH in gas (CH_4 = %)

concentration while *Acacia seyal* and *Acacia tortilis* had the lowest ($p<0.05$). The highest ($p<0.05$) organic matter (OM) concentrations were observed in *Acacia tortilis* and *Acacia seyal*. Organic matter concentrations of *Cordia africana* and *Prosopis juliflora* were significantly lower than *Millettia ferruginea*, *Acacia tortilis* and *Acacia seyal*. The CT concentration of the browses ranged from 0.30-3.40% of DM. The CT content of *Acacia tortilis*, *Acacia seyal* and *Millettia ferruginea* was significantly higher than the remaining browse species.

***In vitro* gas and methane production and organic matter digestibility:** *In vitro* gas and CH_4 production and organic matter digestibility (OMD) of browses are presented in Table 2. Browse species significantly ($p<0.05$) differed in their gas and CH_4 production at 24 h of incubation. Gas and CH_4 production ranged from 19.94-46.84 and 4.18-9.05 mL 400 mg^{-1} DM, respectively. *Acacia seyal* and *Acacia senegal* produced higher gas volume compared with other species. The highest ($p<0.05$) OMD was observed for *Acacia seyal*, *Acacia senegal* and *Croton macrostachyus* while the lowest ($p<0.05$) was for *Millettia feruginea*. *Acacia seyal* which had high CT, produced relatively lower volume of methane and *Croton macrostachyus* with lower CT content produced significantly higher volume of methane than the rest of browses except *Acacia senegal* and *Vernonia amygadalina*. The lowest ($p<0.05$) CH_4 percentage of total gas was recorded from *Acacia seyal* which had high ($p<0.05$) CT concentration.

Effects of PEG inclusion on gas and methane production and organic matter digestibility: The effects of PEG inclusion on *in vitro* GP, OMD and CH_4 production of browses after 24 h of incubation are shown in Table 3. Both in the presence and absence of PEG there were significant ($p<0.05$) variations among browse species in terms of gas, OMD and CH_4 production. Inclusion of PEG in fermentation of browses resulted in a significant ($p<0.001$) increase in gas profile in *Acacia tortilis*, *Acacia seyal* and *Acacia senegal*, which had relatively high CT content. The percentage increase in gas production in the presence of PEG was 64, 47 and 11% for *Acacia tortilis*, *Acacia seyal* and *Acacia senegal*, respectively. However, no effect of PEG were detected on the gas and CH_4 profiles of *Prosopis juliflora*, *Vernonia amygadalina* and *Croton macrostachyus*, which had relatively low CT content. Inclusion of PEG raised the OMD to 61 and 46% in *Acacia tortilis* and *Acacia seyal*, which had relatively highest CT content while the OMD of *Millettia ferruginea*, which had similar CT content with that of *Acacia tortilis* and *Acacia seyal*, was not improved by inclusion of PEG (Table 3). On the other hand, inclusion of PEG increased GP and OMD by 16 and 15% in *Cordia africana* while the GP and OMD in *Prosopis juliflora*, *Vernonia amygadalina* and *Croton macrostachyus*, which had similar CT contents with that of *Cordia africana*, were not increased.

Table 3: Effect of PEG on in vitro gas production, organic matter digestibility and methane production of browses after 24 h of incubation

	Gp			OMD			CH_4^{-1}		CH_4^{-2}(%)	
Browses	PEG+	PEG-	Increment (%)	PEG+	PEG-	Increment (%)	PEG+	PEG-	PEG+	PEG-
Over all	35.40	31.24	13.3	575.0	509.4	12.9	6.74	6.67	20.4	22.7
SE	0.50	0.50	-	7.0	7	-	0.1	0.1	0.8	0.8
Acacia tortilis	38.23^{a}	23.33^{c}	63.9	617.2^{d}	382.4^{c}	61.4	7.84^{ab}	6.33^{bc}	21^{b}	29^{a}
Acacia. seyal	55.70^{a}	37.98^{ab}	46.7	888.6^{a}	608.8^{ab}	46.0	4.89^{c}	4.39^{d}	9^{d}	12^{c}
Acacia senegal	45.85^{b}	41.46^{a}	10.6	743.7^{b}	673.2^{a}	10.5	7.80^{ab}	8.29^{ab}	17^{c}	20^{b}
Prosopis juliflora	17.71^{h}	22.17^{c}	-20.1	296.7^{h}	367.9^{c}	-19.4	3.94^{c}	4.43^{d}	22^{b}	20^{b}
Millettia ferruginea	27.41^{f}	27.07^{bc}	1.3	449.2^{f}	443.9^{bc}	1.2	5.87^{bc}	7.87^{ab}	21^{b}	29^{a}
Vernonia amygadalina	35.52^{e}	36.40^{ab}	-2.4	578.8^{e}	592.1^{ab}	-2.2	8.88^{a}	7.28^{ab}	25^{a}	20^{b}
Croton macrostachyus	40.98^{c}	42.71^{a}	-4.1	665.6^{c}	693.4^{a}	-4.0	8.78^{a}	9.33^{a}	21^{b}	22^{b}
Cordia africana	21.81^{g}	18.82^{c}	15.9	360.8^{g}	313.3^{c}	15.2	5.95^{bc}	5.45^{cd}	27^{a}	29^{a}
SE	1.3	1.3	-	21	21	-	0.3	0.3	2	2

Means in the same column with different superscript differ significantly ($p<0.05$), PEG: Polyethylene glycol, PEG+: Presence of PEG, PEG-: Absence of PEG. Gas production (Gp = mL 400 mg^{-1} DM), Organic matter digestibility (OMD = g kg^{-1} DM), methane production (CH_4^{-1} = mL 400 mg^{-1} DM) percentage of CH_4^{-2} in gas (CH_4 = %)

Table 4: Correlation coefficient (r) of gas production parameters with chemical composition of browses

Chemical composition	Gas	OMD	CH_4
Ash	-0.003	-0.005	-0.086
CP	0.854**	0.856**	0.658**
NDF	-0.589**	-0.586**	-0.383
ADF	-0.697**	-0.695**	-0.516*
ADL	-0.540*	-0.539*	-0.297
CT	-0.182	-0.187	-0.078

OM: Organic matter, CP: Crud protein, NDF: Neutral detergent fiber, ADF: Acid detergent fiber, ADL: Acid detergent lignin, CT: Condensed tannin, OMD: Organic matter digestibility, GP: Gas production, CH_4: methane, *$p<0.05$ and **$p<0.001$

Correlation of *in vitro* fermentation parameters with chemical composition of browses: The correlation coefficient (r) of *in vitro* gas production parameters with chemical compositions of browses are presented in Table 4. All *in vitro* gas production parameters (GP, OMD and CH_4) were positively correlated ($p<0.001$) with CP content and negatively correlated ($p<0.001$) with CT content and fiber components (NDF, ADF and ADL) of the browses. Methane production showed a negative correlation with CT and fiber components.

The range of CP contents of the browses in the current study is in line with the findings of Njidda and Nasiru[16] and Theart *et al.*[17], who reported 14-21 and 13-25%, respectively. It has been indicated that most tropical browses are high in CP and can be used to supplement poor quality roughages to increase productivity of ruminant livestock in the tropics[16-19] which is consistent with the results obtained in the current experiment.

In this study, the NDF and ADF of browses varied from 14-48 and 9-29% DM, respectively, which is comparable with findings of Fadel Elseed *et al.*[20]. The low NDF, ADF and ADL contents and high value of OM in *Acacia seyal* and *Acacia tortilis* in this study indicate that they have better dry matter intake and high digestibility. On the other hand, the high NDF and ADF contents of *Cordia africana* could be associated with poor dry matter intake and digestibility. The high gas production and OM digestibility values of *Acacia seyal* and the low gas production and OM digestibility values of *Cordia africana* observed in Table 2 of this study supports the above arguments. Schroeder[21] reported the negative correlation of NDF and ADF with dry matter intake and digestibility of browses.

The range of CT concentration in the current study is in line with the findings of Fadel *et al.*[20], who reported values ranging from 0.2-6% of DM for similar browses from Sudan. The CT concentrations of *Acacia tortilis*, *Acacia seyal* and *Acacia senegal* in the current study were comparable with the findings of Mahala and Elseed[22], who reported a CT content of 5, 3 and 3% DM for *Acacia tortilis*, *Acacia seyal* and *Acacia senegal*, respectively. Kechero and Janssens[6] reported CT content of *Millettia ferruginea*, *Vernonia amygadalina*, *Croton macrostachyus* and *Cordia africana* as 4, 0.2, 0.1 and 0.1% DM, respectively, which are similar to the current results. The CT concentrations of browses in this study are less than 4% of DM which are not high enough to reduce intake and digestibility of nutrients. Barry and Duncan[23] reported that plant species with low CI content (<5 of % DM) do not affect voluntary feed intake and nutrient digestibility. Hence, these browses in the current study can be good supplements to low quality roughages.

***In vitro* gas and methane production and organic matter digestibility:** The range of gas and CH_4 production of browses in the current study agrees with findings of Gemeda and Hassen[24] from South Africa, who reported 18.1-50.4 and 3.8-8.24 mL 400 mg^{-1} DM. The high gas production (GP) and OMD for *Acacia seyal* and *Acacia senegal* and the lowest GP and OMD for *Cordia africana* could be attributed to low NDF, ADF and ADL values of the formers and highest NDF, ADF and

ADL values of the later (Table 1). It is well accepted that forage degradation in the rumen is mainly affected by the cell wall content and its lignifications[16]. Theart *et al.*[17] reported negative correlation of NDF, ADF and lignin with *in vitro* digestibility. It is well established that low content of ADF and ADL are indicators of good forage quality[25].

The low CH_4 production for *Acacia seyal* and high CH_4 production for *Croton macrostachyus* could be attributed to high CT contents of the former and low CT contents of the later (Table 1). Theart *et al.*[17] also reported a reduction of CH_4 production with an increase in tannin concentration in browses. Similarly, Sebata *et al.*[26] and Singh *et al.*[27] reported reduction of CH_4 production with the inclusion of tannin in ruminant diets.

The lowest ($p<0.05$) CH_4 percentage of total gas was recorded from *Acacia seyal* which had high ($p<0.05$) CT concentration. Tropical browses with high concentrations of CT have decreased methane emissions[1,28,29]. Lopez *et al.*[30] suggested that methane percentage of total gas produced after 24 h of fermentation can be used to determine the methane reduction potential of any feedstuff and the feedstuffs can be classified in to low, moderate and high potentials based on their methane percentage of total gas produced. Therefore, *Acacia seyal* in the current study, with CH_4 (%) in gas was 11, can be categorized as forage with moderate CH_4 reduction potential.

Effects of PEG inclusion on gas and methane production and organic matter digestibility: In the current study, PEG significantly increased the gas and OMD profile in browse species with high CT contents. However, no effect of PEG on the gas and OMD profiles of browses with low CT contents were detected. Similar results were reported by Theart *et al.*[17], Gemeda and Hassen[24] and Sallam *et al.*[31] from *Acacia* species. Addition of PEG could overcome the adverse effect of tannins on nutrient availability because PEG has a high affinity for tannin and forms PEG-tannin complex which inactivates tannins. Inclusion of PEG during the incubation of tannin rich plants led to an increase in GP upto 100% and has the potential to influence rumen fermentation[32]. The little effect for PEG on GP in *Prosopis juliflora*, *Vernonia amygadalina* and *Croton macrostachyus* could be due to the very low concentration of inhibitory compounds like CT in such browses.

In this study PEG differently affected the GP and OMD of different browse species with similar CT content. This indicates that tannin from different plants might show different response in digestibility and methane reduction. Same concentration of tannins from different plants exhibit variation in their effects[11,33]. The differences in activity may be attributed to differences in the molecular weight and structure of the different tannins[1].

Correlation between gas production parameters and chemical composition of browses: The positive correlation between OMD and CP in this study is in line with the reports of Theart *et al.*[17] from similar browses of South Africa and Njidda and Ikhimioya[19] from grasses and browses in Nigeria. A positive correlation between OMD and CP indicate that as the crude protein content in forages increases, the OMD improves[16,17,19]. The negative correlation between OMD and fiber components observed in this study is similar with the findings of Theart *et al.*[17] and Njidda and Ikhimioya[19]. Njidda and Nasiru[16] reported a highly negative correlation between ADF and OMD of leaves of tropical browses. It is well accepted that forage degradation in the rumen is mainly affected by the cell wall content and its lignifications. The NDF, ADF and lignin are significantly and negatively correlated with OMD[25]. The negative correlation between CH_4 production and CT content observed in this study agrees with the finding of Theart *et al.*[17], who reported a significant and negative correlation between tannin content and CH_4 production of tropical browses. Gemeda and Hassen[24] reported that CH_4 production at 24 h showed negative correlation with phenolic compounds (TP, TT, CT and HT) of browses. Sebata *et al.*[26] and Singh *et al.*[27] reported a reduction of CH_4 production in the presence of tannin in ruminant diets.

CONCLUSION

All browse species studied had high CP contents, sufficient to be considered as high protein forages that can be used as supplements for low quality roughages. The high gas production and OM digestibility of *Acacia seyal*, coupled with its lowest percent of CH_4 of total gas produced during fermentation would make the browse potential supplement of low quality roughages while reducing enteric CH_4 emissions. All browse species could have satisfactory energy value, considering their high OM digestibility.

SIGNIFICANCE STATEMENT

This study discovers the methane reduction potential and nutritive values of some indigenous tannin containing browse species from Ethiopia that can be used by pastorals of developing nations to feed their ruminant animals while reducing enteric methane emission. This study will help the researchers to uncover the significant contribution of tannin-rich natural tropical browses in the reduction of methane emission by ruminant animals. So far many researchers were focused on improved forages and concentrate feeds to reduce methane emission from ruminants. Thus, a new theory on the utilization of locally available, tannin-rich, natural browse species as major ruminant feed in order to reduce methane emission by ruminants may be arrived at.

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