

## Nutritive Value of Several Tropical Legume Shrubs in Hainan Province of China

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**Abstract:** The aim of this experiment was to evaluate the nutritive value of tropical legume shrub species *Cratylia argentea*, *Leucaena leucocephala*, *Flemingia macrophylla*, *Cajanus cajan*, *Dendrolobium triangular*, *Cassia didymobotrya*, *Cassia bicapsularis* and *Acacia farnesiana* in Hainan province, China based on their chemical compositions and *in vitro* Gas Production (GP). The Crude Protein (CP) contents of legume shrubs ranged from 13.43% (*D. triangular*) to 18.44% (*C. argentea*). The Acid Detergent Fiber (ADF) and Neutral Detergent Fiber (NDF) contents varied between 20.73% (*S. didymobotrya*) to 48.61% (*D. triangular*) and 21.11% (*S. didymobotrya*) to 55.27% (*D. triangular*), respectively. The Total Tannins (TT) ranged from 0.78% (*C. argentea*) to 2.67% (*A. farnesiana*). The Relative Feed Values (RFV) of tropical legume shrubs ranged from 85.9% (*D. triangular*) to 321% (*C. didymobotrya*). GP of tropical legume shrubs was determined 0, 3, 6, 12, 24, 48, 72 and 96 h after incubation and their kinetics were described using the equation  $y = a + b(1 - \exp^{-ct})$ . The calculated *in vitro* Dry Matter Degradability (IVDMD), Organic Matter Digestibility (OMD) and Metabolizable Energy (ME) ranged from 36.91% (*D. triangular*) to 79.30% (*C. bicapsularis*), 40.70% (*F. macrophylla*) to 72.70% (*C. bicapsularis*), 5.41 MJ kg<sup>-1</sup> (*D. triangular*) to 11.26 MJ kg<sup>-1</sup> (*C. bicapsularis*), respectively.

**Key words:** Chemical composition, relative feed value, *in vitro* gas production, *in vitro* dry matter degradability, organic matter digestibility, metabolizable energy

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### INTRODUCTION

Hainan province situated at the Southernmost tip of China is the 2nd largest island of China. It covers a land with area of 35.4 km<sup>2</sup> and has a tropical monsoon climate with very clear rainy and dry seasons each year. The rainy season runs from May to October and the dry season from November to April. There are calculated 9×10<sup>6</sup> Hainan black goats (*Capra hircas*) in the island. Most of them belong to different smallholder farmers.

These goats live largely on the natural pasture and crop residues and are often short of feeds during the long dry season. Thus, the major constraint of goat productivity in this area is inadequate nutrition. Most tropical legume shrubs are evergreen plants, which contain high protein content and Gross Energy (GE) (Blair, 1990; Leng, 1997; Rubanza *et al.*, 2003; Salem *et al.*, 2006; Mahipala *et al.*, 2009) and play an important role in ruminant feeding systems in many tropical areas around the world. The *in vitro* Gas Production (GP) technique which provides empirical equations to estimate the digestibility and Metabolizable Energy (ME) content of

animal feeds (Menke and Steingass, 1988) has been widely used to evaluate the nutritive value of animal feeds such as forage, straws, byproducts and tropical feeds (Getachew *et al.*, 1998a, b; Makkar *et al.*, 1999; Mould, 2003; Negesse *et al.*, 2009; Camacho *et al.*, 2010). Feeding legume shrubs could improve the nutritional status of goats during the dry season. But the chemical compositions and feeding values of legume shrubs in Hainan are largely unknown. Therefore, the present study was aimed to evaluate the nutritive value of legume shrubs through measurement of their chemical compositions, *in vitro* GP kinetics and some estimated parameters such as ME and Organic Matter Digestibility (OMD).

### MATERIALS AND METHODS

**Sample preparation:** The foliage of *Cratylia argentea*, *Leucaena leucocephala*, *Flemingia macrophylla*, *Cajanus cajan*, *Dendrolobium triangular*, *Cassia didymobotrya*, *Cassia bicapsularis* and *Acacia farnesiana* were procured from the legume shrubs grown

at the Tropical Pasture Research Center (TPRC) of Chinese Academy of Tropical Agricultural Sciences (CATAS). TPRC is located in Danzhou city, Hainan, China and has latitude of 19°30'N, longitude of 109°30'E and altitude of 149 m. The legume shrub species were selected based on ease of collection and their wide occurrence in Hainan province, China. Samples were oven dried at 65°C for 48 h and milled through a 1.0 mm screen for *in vitro* studies and chemical analysis.

**Chemical analysis:** Dry Matter (DM), Ash contents (XA) and Nitrogen (N) content were measured by the Kjeldahl method (AOAC, 1990). Crude Protein (CP) was calculated as  $N \times 6.25$ . Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) were determined by the method of Van Soest *et al.* (1991). Gross Energy (GE) content was determined by PARR6300. Total Tannin (TT) content was determined by the method of Makkar (2003). All chemical analyses were carried out in triplicate.

**Relative feed value:** Relative Feed Value (RFV) developed by the Hay Marketing Task Force of American Forage and Grassland Council (Rohweder *et al.*, 1978) (Table 1) is the most widely used index for forage quality in the marketing of hays (Rohweder *et al.*, 1978). RFV of legume shrubs was calculated from the estimates of Dry Matter Digestibility (DMD) and Dry Matter Intake (DMI). Following are the used equations:

$$\text{ADF} = \text{Acid Detergent Fiber (DM\%)}$$

$$\text{DMI} = \text{Dry Matter Intake (BW\%)}$$

$$\text{DMD (\%)} = 88.9 - (0.779 \times \text{ADF\%})$$

$$\text{DMI (BW\%)} = 120 / \text{NDF\%}$$

$$\text{RFV} = (\text{DMD\%} \times \text{DMI\%}) / 1.29$$

where, BW is the body weight.

**In vitro gas production:** The rumen fluid was collected from three healthy mature Hainan black goats using with permanent rumen cannulae ( $\varnothing = 50$  mm) for measurement of *In vitro* Dry Matter Digestibility (IVDMD) and GP. Rumen fluid was strained through gauze and mixed with buffer solution as described by Menke and Steingass (1988). About 200 mg samples were placed in polyester/polyethylene bags (size 5×3 cm; pore size 25  $\mu\text{m}$ ), incubated at 39°C with 35 mL rumen liquor-buffer mixture in 100 mL glass syringes and measured after 0, 3,

Table 1: Legume, grass and legume-grass mixture quality standards

Quality standard <sup>a</sup>	CP	ADF (DM%)	NDF (DM%)	RFV <sup>b</sup>
Prime	>19	<31	<40	>151
1	17-19	31-40	40-46	151-125
2	14-16	36-40	47-53	124-103
3	11-13	41-42	54-60	102-87
4	8-10	43-45	61-65	86-75
5	<8	>45	>65	<75

<sup>a</sup>Standard assigned by Hay Market Task Force of American Forage and Grassland Council; <sup>b</sup>Relative Feed Value (RFV)- Reference hay of 100 RFV contains 41% ADF and 53% NDF

6, 12, 24, 48, 72 and 96 h incubation. After finishing the *in vitro* digestion trials, bags were gently rinsed with cold tap water and dried at 65°C for 48 h to determine IVDMD. The residues were analyzed for Organic Matter (OM) and Organic Matter Digestibility (OMD). Each measurement was performed in triplicate. GE of feeds was obtained using the method described previously (ARC, 1965) and Metabolizable Energy content (ME) value was calculated using the following equation:

$$\text{ME} = \text{GE} \times \text{IVDMD} \times 0.815$$

The exponential Eq:

$$y = a + b(1 - \exp^{-ct})$$

Where:

y = The gas production at time t

a = The gas production from the immediately soluble fraction (mL)

b = The gas production from the insoluble fraction (mL)

c = The gas production rate constant

a+b = The potential gas production (mL)

t = The incubation time (h)

proposed by Orskov and McDonald (1979) was used to determine characteristics of gas production.

**Predictive methods:** The ME ( $\text{MJ kg}^{-1}$  DM) and OMD of samples were calculated using equations of Menke *et al.* (1979) as follows:

$$\text{ME (MJ kg}^{-1} \text{ DM)} = 2.20 + 0.136 \text{ GP} + 0.057 \text{ CP}$$

$$\text{OMD (\%)} = 14.88 + 0.889 \text{ GP} + 0.45 \text{ CP} + 0.0651 \text{ XA}$$

Where, ME (24 h GP) and OMD (24 h GP) were calculated by using 24 h net Gas Production (GP) ( $\text{mL} / 200 \text{ mg}$ ), CP(%) and XA(%). Similarly, the ME (48 h GP) and OMD (48 h GP) as well as ME (96 h GP) and OMD (96 h GP) were calculated using GP, CP and XA measured 48 and 96 h after incubation, respectively.

**Statistical analysis:** Chemical composition and *in vitro* GP were analyzed using the General Linear Models (GLM) procedure of SAS (1996) in a completely randomized design to test differences among plant species. Differences of the means within species of grass or legume and between grass and legume were compared using probability of difference. The means were compared for statistical significance by using Duncan's multiple range test (Duncan, 1955). Mean differences were considered significant at  $p < 0.05$ . Standard errors of means were calculated from the residual mean square in the analysis of variance. As there were great differences in the chemical composition and *in vitro* kinetics among the Legume shrub species, Pearson correlation coefficients ( $r$ ) between chemical composition and GP and between some estimated parameters within legume shrubs were estimated.

## RESULTS

**Chemical composition:** The chemical composition and TT contents of the shrub species are shown in Table 2. The DM contents of shrubs ranged between 25.02 (*S. bicaularis*) to 37.12% (*A. farnesiana*). CP contents ranged between 13.43% (*D. triangularis*) to 18.44% (*C. argentea*). EE contents varied between 2.36 (*F. macrophylla*) to 5.06 % (*C. bicaularis*). ADF and NDF contents varied between 20.73% (*C. didymobotrya*) to 48.61% (*D. triangularis*) and 21.11% (*C. didymobotrya*)

to 55.27% (*D. triangularis*), respectively GE contents varied between 17.37 MJ  $\text{kg}^{-1}$  (*C. argentea*) to 19.57 MJ  $\text{kg}^{-1}$  (*A. farnesiana*). TT ranged from 0.78% (*C. argentea*) to 2.67 % (*A. farnesiana*).

**Relative Feed Value (RFV):** DMD, DMI and RFV of the shrub species are shown in Table 3. DMD of shrubs ranged from 51.03% (*D. triangularis*) to 72.75% (*C. didymobotrya*). DMI ranged from 2.17 (*D. triangularis*) to 5.68% (*C. didymobotrya*). RFV ranged from 85.9% (*D. triangularis*) to 321% (*C. didymobotrya*). The grades of *Cratylia argentea*, *Leucaena leucocephala*, *Flemingia macrophylla*, *Cajanus cajan*, *Dendrolobium triangularis*, *Cassia didymobotrya*, *Cassia bicaularis* and *Acacia farnesiana* were 2, prime, 2, 1, 4, prime, prime and prime, respectively. In another word, the grade of shrubs ranged from 4 to prime according to the standard assigned by Hay Market Task Force of American Forage and Grassland Council given in Table 1.

The order of shrub species containing RFV from high to low was *Cassia didymobotrya*, *Cassia bicaularis*, *Acacia farnesiana*, *Leucaena leucocephala*, *Cajanus cajan*, *Cratylia argentea*, *Flemingia macrophylla* and *Dendrolobium triangularis*. Except *Dendrolobium triangularis*, all the observed legume shrubs are high grade fed for ruminants.

**Gas production:** The GP profiles for the shrub species examined are shown in Fig. 1. There were differences

Table 2: Chemical composition of Legume shrubs, DM (%) basis

Species	DM	CP	EE	ADF	NDF	Ash	GE	TT
<i>Cratylia argentea</i>	25.98 <sup>d</sup>	18.44 <sup>a</sup>	2.98 <sup>cd</sup>	30.69 <sup>cd</sup>	49.19 <sup>b</sup>	9.25 <sup>a</sup>	17.37 <sup>c</sup>	0.78 <sup>d</sup>
<i>Leucaena leucocephala</i>	31.25 <sup>c</sup>	18.19 <sup>a</sup>	4.15 <sup>b</sup>	27.19 <sup>d</sup>	37.66 <sup>c</sup>	8.27 <sup>a</sup>	17.89 <sup>bc</sup>	2.34 <sup>a</sup>
<i>Flemingia macrophylla</i>	33.67 <sup>bc</sup>	14.22 <sup>c</sup>	2.36 <sup>d</sup>	39.43 <sup>b</sup>	45.27 <sup>b</sup>	5.40 <sup>bc</sup>	17.98 <sup>b</sup>	1.91 <sup>b</sup>
<i>Cajanus cajan</i>	36.26 <sup>ab</sup>	15.29 <sup>bc</sup>	4.43 <sup>ab</sup>	34.29 <sup>c</sup>	43.52 <sup>b</sup>	4.80 <sup>c</sup>	19.16 <sup>a</sup>	1.29 <sup>c</sup>
<i>Dendrolobium triangularis</i>	35.98 <sup>ab</sup>	13.43 <sup>c</sup>	3.31 <sup>c</sup>	48.61 <sup>a</sup>	55.27 <sup>a</sup>	5.85 <sup>bc</sup>	18.01 <sup>b</sup>	1.16 <sup>c</sup>
<i>Cassia didymobotrya</i>	26.69 <sup>d</sup>	15.49 <sup>bc</sup>	4.73 <sup>ab</sup>	20.73 <sup>e</sup>	21.11 <sup>e</sup>	7.19 <sup>ab</sup>	17.59 <sup>bc</sup>	1.82 <sup>b</sup>
<i>Cassia bicaularis</i>	25.02 <sup>d</sup>	17.19 <sup>ab</sup>	5.06 <sup>a</sup>	21.94 <sup>e</sup>	23.75 <sup>e</sup>	8.32 <sup>a</sup>	17.42 <sup>c</sup>	1.38 <sup>c</sup>
<i>Acacia farnesiana</i>	37.12 <sup>a</sup>	18.05 <sup>a</sup>	4.66 <sup>ab</sup>	21.87 <sup>e</sup>	31.27 <sup>d</sup>	4.22 <sup>c</sup>	19.57 <sup>a</sup>	2.67 <sup>a</sup>
Mean	31.49	16.29	3.96	30.59	38.38	6.66	18.12	1.45
SE	4.99	1.94	0.96	9.83	12.19	1.85	0.81	0.59

DM: Dry Matter (%), CP: Crude Protein (%), EE: Crude fat (%), NDF: Neutral Detergent Fiber (%), ADF: Acid Detergent Fiber (%), GE: Gross Energy (MJ  $\text{kg}^{-1}$ ), TT: Total Tannin (%). Means within the same column with different letters are significantly different ( $p < 0.05$ ). SE: Standard Error

Table 3: The effect of species on dry matter digestibility, dry matter intake and the relative feed value

Species	DMD (%)	DMI (%)	RFV	Quality standard
<i>Cratylia argentea</i>	64.99 <sup>bc</sup>	2.44 <sup>d</sup>	123.0 <sup>c</sup>	2
<i>Leucaena leucocephala</i>	67.72 <sup>b</sup>	3.19 <sup>c</sup>	167.0 <sup>bc</sup>	Prime
<i>Flemingia macrophylla</i>	58.18 <sup>d</sup>	2.65 <sup>d</sup>	119.0 <sup>c</sup>	2
<i>Cajanus cajan</i>	62.18 <sup>c</sup>	2.76 <sup>d</sup>	133.0 <sup>c</sup>	1
<i>Dendrolobium triangularis</i>	51.03 <sup>e</sup>	2.17 <sup>e</sup>	85.9 <sup>d</sup>	4
<i>Cassia didymobotrya</i>	72.75 <sup>a</sup>	5.68 <sup>a</sup>	321.0 <sup>a</sup>	Prime
<i>Cassia bicaularis</i>	71.81 <sup>a</sup>	5.05 <sup>a</sup>	281.0 <sup>a</sup>	Prime
<i>Acacia farnesiana</i>	71.86 <sup>a</sup>	3.84 <sup>b</sup>	214.0 <sup>b</sup>	Prime
Mean	65.07	3.47	181.0	-
SE	7.66	1.29	84.0	-

DMD: Dry Matter Digestibility (%), DMI: Dry Matter Intake (%), RFV: Relative Feed Value. Means within the same column with different letters are significantly different ( $p < 0.05$ ). SE: Standard Error

among species in the gas volume produced from digestion throughout the incubation period: gas volume produced from *A. farnesiana* was the lowest and from *C. bicapsularis* was the highest. The GP kinetics and some estimated parameters are shown in Table 4. GP parameters a, b, a+b and c of shrubs ranged from -4.57% (*A. argentea*) to 0.21% (*A. farnesiana*), 12.01% (*A. farnesiana*) to 37.04% (*C. didymobotrya*), 12.22% (*A. farnesiana*) to 38.31% (*C. bicapsularis*) and 0.04% (*F. macrophylla* and *D. triangulare*) to 0.08% (*C. bicapsularis*), respectively. The GP at 96 h incubation ranged from 12.29 mL (*A. farnesiana*) and 40.07 mL (*C. bicapsularis*). The IVDMD of the shrub species ranged from 36.91-79.30%. The order of shrub species which had GP parameters a+b, GP 96 h and IVDMD from high to low was *Cassia bicapsularis*, *Cassia didymobotrya*, *Cratylia argentea*, *Leucaena leucocephala*, *Cajanus cajan*, *Flemingia macrophylla*, *Dendrolobium triangulare* and *Acacia farnesiana*.

#### Metabolizable energy and organic matter digestibility:

The observed and calculated values of ME and OMD are shown in Table 5. The ME values of shrubs ranged from

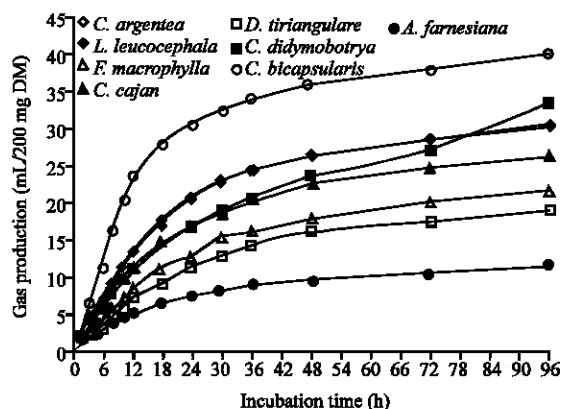


Fig. 1: Cumulative gas production profiles (mL gas/200 mg DM) from *in vitro* fermentation of the shrub species >96 h

5.41 (*D. triangulare*) to 11.26 MJ kg<sup>-1</sup> (*C. bicapsularis*) in *in vitro* observation from 3.23% (*A. farnesiana*) to 6.34% (*C. bicapsularis*) at 24 h incubation, from 3.54% (*A. farnesiana*) to 7.07% (*S. bicapsularis*) at 48 h and from 3.78% (*A. farnesiana*) to 7.64% (*C. bicapsularis*) at 96 h, respectively. The OMD values of shrubs ranged from 40.70% (*F. macrophylla*) to 72.70% (*C. bicapsularis*) in *in vitro* observation, from 21.66% (*A. farnesiana*) to 41.96% (*C. bicapsularis*) at 24 h, 23.68% (*A. farnesiana*) to 46.72% (*C. bicapsularis*) at 48 h, 25.21% (*A. farnesiana*) to 50.48% (*C. bicapsularis*) at 96 h, respectively. The observed values of ME and OMD were higher than the predicted and the predicted increase with the incubation time. The wide variation in the predicted values of ME and OMD is evident when compared to those observed *in vitro*.

#### Correlations among gas production, some estimated parameters and chemical composition:

The correlation coefficients among chemical composition, GP and some estimated parameters of legume shrub species are shown in Table 6. There were strong positive correlations among GP, ME, IVDMD and OMD contents (data not presented). CP and EE contents were positively correlated with GP, ME, IVDMD and OMD ( $p > 0.05$ ). XA were positively correlated with GP ( $p < 0.05$ ), ME ( $p > 0.05$ ), IVDMD ( $p < 0.05$ ) and OMD ( $p > 0.05$ ). ADF and NDF contents were negatively correlated with GP ( $p > 0.05$ ), ME ( $p < 0.05$ ), IVDMD ( $p < 0.05$ ) and OMD ( $p < 0.05$ ). GE and TT contents were negatively correlated with GP, ME, IVDMD and OMD ( $p > 0.05$ ). ADF/CP and NDF/CP contents were negatively correlated with GP ( $p > 0.05$ ), ME ( $p < 0.05$ ), IVDMD ( $p < 0.05$ ) and OMD ( $p < 0.05$ ).

#### Predictive equations of ME, IVDMD and OMD for tropical legume shrubs:

Predictive equations were screened with multiple linear regression analysis of several chemical compositions and GP at 96 h of incubation which is significantly correlated with ME,

Table 4: Gas production kinetics and some estimated parameters

Species	a	b	a+b	c	GP	IVDMD
<i>Cratylia argentea</i>	-4.57 <sup>b</sup>	35.83 <sup>b</sup>	31.26 <sup>bc</sup>	0.06 <sup>b</sup>	31.68 <sup>bc</sup>	66.22 <sup>b</sup>
<i>Leucaena leucocephala</i>	-0.87 <sup>a</sup>	30.77 <sup>c</sup>	29.90 <sup>bc</sup>	0.05 <sup>bc</sup>	30.19 <sup>bc</sup>	60.67 <sup>b</sup>
<i>Flemingia macrophylla</i>	0.01 <sup>a</sup>	21.90 <sup>d</sup>	21.90 <sup>d</sup>	0.04 <sup>c</sup>	21.46 <sup>d</sup>	43.00 <sup>d</sup>
<i>Cajanus cajan</i>	-0.24 <sup>a</sup>	27.71 <sup>c</sup>	27.47 <sup>c</sup>	0.05 <sup>bc</sup>	27.78 <sup>c</sup>	50.36 <sup>c</sup>
<i>Dendrolobium triangulare</i>	-0.61 <sup>a</sup>	20.73 <sup>d</sup>	20.11 <sup>d</sup>	0.04 <sup>c</sup>	19.31 <sup>d</sup>	36.91 <sup>e</sup>
<i>Cassia didymobotrya</i>	-2.85 <sup>b</sup>	37.04 <sup>b</sup>	34.19 <sup>b</sup>	0.06 <sup>b</sup>	35.08 <sup>b</sup>	76.16 <sup>a</sup>
<i>Cassia bicapsularis</i>	-3.66 <sup>b</sup>	41.97 <sup>a</sup>	38.31 <sup>a</sup>	0.08 <sup>a</sup>	40.07 <sup>a</sup>	79.30 <sup>a</sup>
<i>Acacia farnesiana</i>	0.21 <sup>a</sup>	12.01 <sup>e</sup>	12.22 <sup>e</sup>	0.05 <sup>bc</sup>	12.29 <sup>e</sup>	46.49 <sup>cd</sup>
Mean	-1.57	28.49	26.92	0.05	27.23	57.39
SE	1.85	9.93	8.44	0.01	9.06	15.66

Means within the same column with different letters are significantly different ( $p < 0.05$ ). SE: Standard Error

Table 5: Metabolizable Energy (ME, MJ kg<sup>-1</sup> DM) values and Organic Matter Digestibility (OMD (DM%)) for the shrub species evaluated by different methods

Species	ME				OMD			
	<i>In vitro</i>	24 h GP	48 h GP	96 h GP	<i>In vitro</i>	24 h GP	48 h GP	96h GP
<i>Cratylia argentea</i>	9.36 <sup>b</sup>	5.03	5.79	6.36	60.10 <sup>b</sup>	33.37	38.37	42.06
<i>Leucaena leucocephala</i>	8.83 <sup>b</sup>	4.99	5.79	6.31	55.65 <sup>b</sup>	33.17	38.38	41.76
<i>Flemingia macrophylla</i>	6.30 <sup>d</sup>	3.97	4.65	5.15	40.70 <sup>d</sup>	26.48	30.91	34.19
<i>Cajanus cajan</i>	7.85 <sup>c</sup>	4.50	5.27	5.77	47.94 <sup>c</sup>	29.93	34.93	38.22
<i>Dendrolobium triangulare</i>	5.41 <sup>a</sup>	3.77	4.40	4.78	34.75 <sup>e</sup>	25.15	29.29	31.73
<i>Cassia didymobotrya</i>	10.89 <sup>a</sup>	4.62	5.77	6.73	70.68 <sup>a</sup>	30.73	38.25	44.49
<i>Cassia bicapsularis</i>	11.26 <sup>a</sup>	6.34	7.07	7.64	72.70 <sup>a</sup>	41.96	46.72	50.48
<i>Acacia farnesiana</i>	7.39 <sup>c</sup>	3.23	3.54	3.78	44.53 <sup>cd</sup>	21.66	23.68	25.21
Mean	8.41	4.56	5.29	5.82	53.38	30.31	35.07	38.52
SE	2.08	0.95	1.08	1.22	13.83	6.21	7.05	7.96

Means within the same column with different letters are significantly different (p<0.05). SE: Standard error

Table 6: The correlation coefficients between GP, ME, IVDMD and OMD and chemical composition of Legume shrubs

Parameters	GP	ME	IVDMD	OMD
CP	0.23	0.54	0.50	0.49
EE	0.34	0.61	0.53	0.56
ADF	-0.41	-0.81*	-0.75*	-0.77*
NDF	-0.44	-0.76*	-0.72*	-0.74*
Ash	0.77*	0.66	0.74*	0.69
GE	-0.18	-0.42	-0.47	-0.45
TT	-0.57	-0.14	-0.23	-0.19
ADF/CP	-0.42**	-0.80**	-0.74**	-0.76**
NDF/CP	-0.47**	-0.84**	-0.78**	-0.80**

\*Significance at 5% level; \*\*significance at 1% level

IVDMD and OMD. About three approaches were developed to predict ME, IVDMD and OMD for tropical Legume shrubs as:

$$\text{ME} = 9.52 - 0.94 \times \text{ADF} + 0.41 \times \text{NDF} + 0.16 \times \text{GP} + 10.06 \times (\text{ADF/CP}) - 5.17 \times (\text{NDF/CP})$$

$$(R^2 = 0.9973)(p < 0.01) \quad (1)$$

$$\text{IVDMD} = 53.10 - 10.57 \times \text{ADF} + 7.07 \times \text{NDF} - 0.04 \times \text{XA} + 1.28 \times \text{GP} + 151.63 \times (\text{ADF/CP}) - 113.26 \times (\text{NDF/CP})$$

$$(R^2 = 0.9975)(p < 0.01) \quad (2)$$

$$\text{OMD} = 55.12 - 8.60 \times \text{ADF} + 5.09 \times \text{NDF} + 1.12 \times \text{GP} + 114.54 \times (\text{ADF/CP}) - 77.60 \times (\text{NDF/CP})$$

$$(R^2 = 0.9978)(p < 0.01) \quad (3)$$

Where, the units of ME and GP were MJ kg<sup>-1</sup> DM and mL/200 mg DM at 96 h of incubation, respectively the unit of IVDMD and OMD was %; the unit of CP, ADF, NDF and XA was DM%.

## DISCUSSION

**Chemical composition:** Chemical compositions of some tropical legume shrubs have been reported previously. Kexian *et al.* (1998) reported that in Colombia, N (CP), ADF and NDF contents were 3.37, 36.73 and 58.87% for *C. argentea* and 2.86, 36.80 and 48.56% for

*F. macrophylla*, respectively. Tiemann *et al.* (2008) reported that in the same region of Colombia, N (CP), ADF and NDF contents were 41.3, 232 and 423 g kg<sup>-1</sup> DM for *C. argentea* and 38.1, 167 and 431 g kg<sup>-1</sup> DM for *L. leucocephala*, respectively. However, their results of N contents (CP) (3.37% and 41.3 g kg<sup>-1</sup> DM, respectively) for *C. argentea* were higher than that of 18.44% in this study.

The ADF and NDF contents for *C. argentea* in the study were lower than that reported by Kexian *et al.* (1998) and higher than that by Tiemann *et al.* (2008). Compared to the present study, Kexian *et al.* (1998) reported lower N (CP) and comparable NDF and ADF contents for *F. macrophylla* and Tiemann *et al.* (2008) reported higher N (CP), higher NDF and lower ADF contents for *L. leucocephala*. Getachew *et al.* (2002) analyzed leaf samples of tropical browse species collected from the International Livestock Research Institute (ILRI) Forage Seed Multiplication Centre, Zwai (Ethiopia) and reported higher CP contents, 194.7, 171.8 and 265.5 g kg<sup>-1</sup> DM for *L. leucocephala*, *F. macrophylla* and *C. cajan*, respectively, compared to the present study. Fondevila *et al.* (2002) reported in Brazil, N (CP), ADF and NDF contents of 33.7, 198 and 390, respectively for *L. leucocephala* and 30.2, 328 and 523 g kg<sup>-1</sup> DM, respectively for *C. cajan*.

Compared to the present results, their results of N and NDF contents are higher and of ADF contents are lower. Ramirez *et al.* (2000) and Ramirez and Lara (1998) reported higher level of CP, ADF and NDF contents (20.2, 23.4 and 39.5%) for *A. farnesiana* in Mexico than the results. Interestingly, *D. triangulare* has been reportedly used as tropical forage legumes in marginal smallholder farming systems of the sub humid and humid tropical regions (Schultze-Kraft *et al.*, 1989; Van der Maesen, 1996; Kretschmer Jr. and Pitman, 2001). *C. bicapsularis* and *C. didymobotrya* has also been reportedly used as medicine in Africa because their extracts have diseases or parasites-resistant activity (Deograciousm and Innocent,

2006; Cyrus *et al.*, 2008). The minimum CP in diet required for lactation and growth of ruminants are 12% and 11.3%, respectively (ARC, 1984). *L. leucocephala*, *C. argentea* and *A. farnesiana* showed comparable CP content with *Medicago sativa* (NRC, 2007). Overall, the mean CP content in the legume shrub species was 16.29%, obviously higher than the threshold CP requirement, indicating these eight legume shrub species could be considered as potential CP source to supplement poor quality basal diets. TT of legume shrubs has been reported. For example, Kexian *et al.* (1998) reported that the total condensed tannin content were 0% for *C. argentea* and 5.13% for *F. macrophylla* which is higher than that in the present study.

Tiemann *et al.* (2008) reported a higher TT level for *C. argentea* and *L. leucocephala* compared with that of the current study. Getachew *et al.* (2002) reported TT were 44.8, 85.3 and 11.4 g kg<sup>-1</sup> DM for *L. leucocephala*, *F. macrophylla* and *C. cajan*, respectively; only *C. cajan* had lower TT compared to the present study. Fondevila *et al.* (2002) reported that *L. leucocephala* and *C. cajan* had higher total condensed tannins than the present study. Ramirez and Lara (1998) reported lower TT (1.8%) in *A. farnesiana* compared to the present study. Barry *et al.* (1986) suggested the level of tannins in forages for ruminants should be 30-40 g kg<sup>-1</sup> DM in order to improve the efficiency of nitrogen digestion. Low level of tannins could protect protein from microbial degradation, thus increasing the amount of undegraded proteins in the small intestine.

TT contents of all legume shrub species in the current experiment were lower than the suggested level. However, variations in the chemical composition of legume shrub species in current study have been observed in other studies on the same shrubs which are largely affected by plant species, planting location, plant morphological fraction, environmental factors, part of plants and maturity stage (Chikagwa-Malunga *et al.*, 2009; Camacho *et al.*, 2010). Overall, legume shrubs in the current study are potential high quality feed resource for goats because of its higher CP content and lower tannin contents in tropical area in China.

**Gas production and estimated parameters IVDMD, OMD and ME:** Kexian *et al.* (1998) reported that IVDMD were 48.42% for *C. argentea* and 22.87% for *F. macrophylla*. Bakshi and Wadhwa (2007) reported higher net GP (132.4 mL g<sup>-1</sup> DM/24 h) and OMD (70.1%) for *L. leucocephala* compared to the present study. Fondevila *et al.* (2002) reported high GP (mL g<sup>-1</sup> DM/24 h) and OMD and low DMD for *L. leucocephala* (163, 47.3, 55.6%, respectively) and *C. cajan* (146, 41.2, 55%, respectively). Ramirez *et al.* (2000) and Ramirez and Lara (1998) reported the DMD of *A. farnesiana* in *in situ* study

with sheep and in *in vitro* study using effective DMD at solid outflow rates as 2, 5 and 8% h<sup>-1</sup> were 56.3, 47.7 and 43.4%, respectively as well as 55.5, 46.7, 41.5%, respectively.

Krishnamoorthy *et al.* (1995) reported ME for *L. leucocephala* K8 and *L. leucocephala* K72 were 8.2 and 7.9 MJ kg<sup>-1</sup>, respectively. The means of their results were comparable to the present study. GP and estimated parameters IVDMD, OMD and ME of the legume shrub species in this study were different from those reported in the literature for other tropical shrubs. The wide variations of GP and estimated parameters were mostly caused by their variable nutrient contents.

Overall, the mean ME content of the legume shrub species was 8.41 MJ kg<sup>-1</sup> which is sufficient to meet the maintenance ME requirement (8.42 MJ kg<sup>-1</sup> DM) for goats fattening in China (Yang, 2003). Moreover, *C. argentea*, *C. didymobotrya* and *C. bicapsularis* were relatively adequate to the ME requirements of growing goats with 30 kg body weight (NRC, 2007). In addition, the legume shrubs studied were high in ME, thus they except *F. macrophylla* and *D. triangulare* would be able to support animal production as a sole diet.

**Correlations among gas production, some estimated parameters and chemical composition:** Kexian *et al.* (1998) reported IVDMD was positively correlated with N and negatively with ADF, NDF and CT for three tropical shrub legumes. Among them, the correlation between IVDMD with N, ADF and CT were significant. Other researchers have reported negative relationships between the *in vitro* fermentation parameters such as IVDMD, OMD, ME, shrub fiber fraction and TT (Rubanza *et al.*, 2003; Salem *et al.*, 2007; Salem, 2005; Casler and Jung, 2006; Camacho *et al.*, 2010).

Mahipala *et al.* (2009) reported that AX was positively correlated with GP, OMD and ME and TT was significantly negatively correlated with GP, OMD and ME in leguminous browse species. Consistent relationships occurred between the contents of CP, NDF, ADF, AX, TT, *in vitro* GP and some estimated parameters in the present study. Cone reported incubation of casein produced 32% GP compared to carbohydrates. Tan reported the dietary ratio of structural and nonstructural carbohydrates could affect rumen degradability.

Tang *et al.* (2005) reported that characteristic of *in vitro* GP and its positive correlation with NDS/CP (p<0.001) depended upon the balance of non-structural carbohydrate and CP. In present study, NDF/CP and ADF/CP were negatively correlated (p<0.01) with GP, IVDMD, OMD and ME. This could be explained as

follows; the carbohydrates in substrate was the energy source for microorganisms in rumen in which gas was largely produced from GP and utilization efficiency of structural and nonstructural carbohydrate was different. CP in substrate provided nitrogen for the microorganisms and was only used to produce small part of GP. Sub-strate balance between carbohydrates and protein was the key factor for *in vitro* fermentation. However, further researches on the optimal ratio of carbohydrates to protein and the ratio of structural and nonstructural carbohydrates in the diet for ruminant are necessary.

**Predictive equations:** Menke *et al.* (1979) suggested ME values of feeds can be predicted from GP by *in vitro* incubating samples with rumen fluid in combination with a few chemical components. In addition, a strong correlation between ME values measured *in vivo* and predicted by GP. Robinson *et al.* (2004) reported six unified single equations to predict *in vivo* ME values of feeds such as forages, grains, protein meals and other by 0.99 products in sheep. These results suggested that no equation was able to consistently discriminate ME value of individual feed. The goodness of fit ( $r^2$ ) for the two NRC approaches and UC Davis approach were 0.61, 0.72 and 0.84, respectively.

Therefore, the precision of these prediction approaches is lower than the ME prediction equation in present study ( $r^2 = 0.9973$ ). Magalhaes *et al.* (2010) evaluated several methods for ME estimation of some tropical forages and compared with that measured in *in vivo* digestion. Their results indicated that *in vitro* systems underestimated ME values of most tropical forages and suggested that the accurate estimate systems for predicting ME of tropical feeds should base upon chemical composition such as NRC (2001), Detmann 1 and 2.

Overall, the predicted ME values with different prediction equations were mostly inconsistent with those determined *in vivo*. On the other hand, predictive equations should vary dependent upon local conditions, specific objectives and feedstuffs. The equations proposed in the present study to predict the values of ME, IVDMD and OMD have high accuracy and precision and should be recommended to evaluate the nutritive value of tropical legume shrubs.

The tropical legume shrubs studied except *D. triangulare* have high contents of RFV, CP, ME, IVDMD and OMD and moderate content of TT, most of which meet the nutrition requirements for small ruminants (NRC, 2007). Thus, legume shrubs have higher potential nutritive values for goats in Hainan province of China. The predictive equations proposed in the present study for ME, IVDMD and OMD based on the

*in vitro* gas production technique and some chemical composition are suitable for tropical legume shrubs in China.

## CONCLUSION

These results indicate that the above nutrition indices of the tropical legume shrubs studied except *D. triangulare* meet the requirement proposed by Hay Market Task Force of American Forage and Grassland Council and are potential high nutrition feeds for goats in Hainan province of China.

In addition, this study found that ADF, NDF, ash content, ADF/CP and NDF/CP were significantly correlated with IVDMD, OMD and ME and proposed new, more accurate and precise predictive equations for ME, IVDMD and OMD.

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