

Effect of different processing methods on the nutrient composition and anti-nutritional factors of *Gmelina arborea* leaves in Anwai community, Delta State, Nigeria

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Abstract. The effect of various processing methods on phytochemicals and nutrient composition of Gmelina arborea leaves was studied. Leaves of G. arborea leaves (GAL) were collected and processed as fresh (T1), chopped (T2), sun-dried (T3), air-dried (T4) and boiled-dried (T5). Samples were used for proximate, mineral and phytochemical analyses to determine the nutrient profile and anti-nutrient composition. The data collected were subjected to one-way analysis of variance and means were separated by Duncan's Multiple Range Test. Nutritional analysis of the processed G. arborea leaves showed that samples contain major nutrients like carbohydrates, protein, fat, minerals and vitamins for ruminants optimum physiological performance. The value of crude protein obtained in this study was 18.05-20.95%. Mineral profile revealed the presence of iron, zinc, calcium, potassium, phosphorous, magnesium, copper and sodium which varies with processing methods. The phytochemical analysis revealed the presence of tannin, alkaloid, oxalate, flavonoid, saponnin and steroid depicting potential toxicity of the feed resources. The boiled-dried method had a better reduction effect on aniti-nutrients, followed by air-dried and sun-dried, compared to the fresh method. Processing tremendously improved the nutritive value of G. arborea leaves as feed for ruminants. The processing methods adopted in the study showed that they do not reduce the nutritional values but could reduce the anti-nutritional components.

Key Words: browse plants, mineral profile, phytochemicals, proximate composition.

Introduction. The economic consequences of plant toxicity have provided the impetus for concerted research efforts particularly in countries where extensive livestock faming is practiced. In the light of this, D'Mello (2000) reported that the world list of poisonous plants includes about 1,500 species. Interestingly, majority of these plant species are readily acceptable and relished by ruminants and they are often sources of tannins (Bruneton 1999) as well as saponins, which make them somewhat medicinal due to their pharmacological properties (Agbugui et al 2010). For this reason, farmers and researchers alike do undermine the possibility of anti-nutritional factors (ANF) present in the fodder plants, even when they are capable of eliciting physiological response in ruminants (Bruneton 1999). Tropical browses have been shown to contain varying quantities of condensed tannin and other anti-nutritional substances in their biomass which affect their optional utilization by animals (Onwuka 1996). The utilization of Gmelina arborea leaves also has the attendant problem of high ANF (Bruneton 1999) and information on the various processing techniques to detoxify these anti-nutritional factors before it can be fully utilize as feedstuff for ruminants is still limited. As a result of these, there is the need to search for an effective technique of processing G. arborea leaves into a more nutritious feed to enhance its utilization as feed for ruminant animals, by detoxifying its anti-nutritional factors. Against this backdrop some processing methods such as chopping (i.e. fresh and succulent), air-drying, and sun-drying as well as boiling and drying methods of processing needs to be investigated. The objective of this study

therefore, is to examine the effect of different processing methods on the phytochemical properties, nutrient composition, and the anti-nutrient contents of *G. arborea* leaves.

Material and Method

Experimental location. The study was conducted at the sheep and goat unit of the Teaching and Research Farm and the Animal Science laboratory of the Department of Animal Science, Faculty of Agriculture, Delta State University, Asaba Campus, Nigeria. Delta State falls within the humid tropics of Nigeria and Asaba precisely lies between longitudes 6° E and 8° E and Latitude 06° 49`N of the Equator. Asaba has its raining season from March to September with a mean annual rainfall of 1,500– 1,849.3 mm. It has a moderate climate with very high temperature during the dry season (October – February) with its mean annual temperature and precipitation of $28^{\circ}\pm6^{\circ}$ C and 117 mm, respectively (Asaba Metrological Station 2011).

Collection and preparation of G. arborea leaves. Fresh *G. arborea* leaves (GAL) were collected from Delta State University, Asaba Campus. The samples were authenticated at the Herbarium unit of the Department of Forestry and Wildlife, Delta State University, Asaba Campus. To ensure uniformity, all samples were collected from within the same location in the University premises.

Processing techniques of G. arborea leaves samples. 250 g of *G. arborea* leaves each were weighed and processed as fresh, air dried; sun dried and boiled dried samples. <u>Fresh</u>. The fresh green samples were collected, crushed in a mortar (Pyrex CR), packed in a cellophane bag and stored in freezer for subsequent analysis.

<u>Air-dried</u>. Leaves samples meant for air-drying (i.e. indoor drying) were collected and spread in a well-ventilated laboratory at the Animal Science Department Laboratory, Asaba Campus at a mean room temperature of 28.5°C for 48 hrs.

<u>Sun-dried</u>. Leave samples for sun drying (i.e. outdoor drying) were collected and spread on a special drying platform at the University premises Asaba Campus at a mean temperature of 33.2°C for 48 hrs.

<u>Boil-dried</u>. Samples for boiled and dried was boiled for 3 minutes and sun dried for two days (48 hrs). The air dried, sundried and boiled dried processed samples of *G. arborea* leaves were ground using hammer mill (Arthur Thomas Co. USA) to a mesh size of 2 mm. The milled samples were packed in labeled envelopes and kept in a cool and dry shelf for subsequent analysis.

Determination of the nutritional profiles of G. arborea leaves samples. The milled samples of processed *G. arborea* leaves were used for the proximate analysis according to standard procedures (AOAC 2000). For each parameter determined, analyses were done in triplicates. The crude fibre, acid detergent fibre, and neutral detergent fibre were determined by the methods of Van Soest & Robertson (1985). The mineral contents were analyzed from the ashed samples, calcium, sodium and potassium components were determined by flame emission spectrophotometry method using Jenway digital flame photometer and phosphorus was estimated by colourimetric method using spectronic 20D. Iron, magnesium, zinc, and copper contents were determined by Atomic Absorption Spectrophotometric (AAS) method.

Determination of the anti-nutritional factors (phytochemicals) in G. arborea leaves samples. Tannin, Alkaloid, oxalate and flavonoids content was determined by the method of Makkar et al (2007). While total saponin content was determined using a spectrophotometric method of Hiai et al (1976). The method of Smith et al (1995) was used to estimate the steroid content.

Experimental design and analysis. The sets of data collected for nutritional profiles (NP) and ANF of the various processing techniques of *G. arborea* leaves were subjected

to Analysis of Variance, and significant means were separated by Duncan's Multiple Range Test at 5% level of probability using the procedures of SAS (2000).

Results and Discussion. The result of the proximate composition of *G. arborea* leaves (GAL) with different processing techniques is shown in Table 1. The values shows that there were significant differences (P<0.05) in the proximate composition of GAL processed using different techniques such as fresh, sun-dried, air-dried, and boiled-dried. Values for dry matter content (%) varied from 91.5% in boiled dried, 92.54% in fresh, 93.60% in air-dried to 93.67% in sun-dried GAL.

Ash content of GAL showed significantly different mean values across different processing techniques. Values recorded were 2.73% (sun-dried), 3.03% (air-dried), 3.73% (boiled-dried) and 6.00% for fresh sample. Crude fibre values varied from 7.31% (air-dried), 8.35% (sun-dried), and 10.86% (boiled-dried) to 14.18% for fresh. Crude protein shows significantly different mean values across different processing techniques with 18.05% (boiled-dried), 19.08% (sun-dried), 19.79% (air-dried) and 20.38% for fresh.

Ether extract content also showed significant difference (P<0.05) with sun-dried (4.73%), air-dried (5.53%), boiled-dried (8.41%), and (13.77%) for fresh.

Significant differences existed in Nitrogen Free Extract (NFE) (P<0.05) across the different processing techniques; the values were 42.62% for fresh, boiled-dried 55.25%, sun-dried 58.74% and 61.18% for air-dried techniques.

While values recorded for acid detergent fibres were 15.38% for boiled-dried, 17.82% sun-dried, 24.38% air-dried and 31.48% for fresh.

Neutral detergent fibre varied significantly (P<0.05) across the different treatments with fresh recording higher value. Values recorded for boiled-dried, sun dried and air dried were 34.28%, 43.91% and 50.74% respectively.

Table 1
Proximate and cell wall composition of processed *Gmelina arborea* leaves using different processing techniques

	Processing technique				
Nutrient	Fresh	Sun-dried	Air-dried	Boiled-dried	SEM
	T1	Т3	T4	<i>T5</i>	
Dry matter (%)	92.54 ^b	93.67 ^a	93.60 ^a	91.50 ^c	0.20
Ash (%)	6.00 ^a	2.73 ^c	3.03 ^b	3.78 ^b	0.20
Crude Fibre (%)	14.18 ^a	8.35 ^c	7.31 ^c	10.86 ^b	0.42
Crude Protein (%)	20.95^{a}	19.08 ^b	19.79 ^b	18.05 ^c	0.33
Ether Extract (%)	13.77 ^a	4.73 ^c	5.55 ^c	8.41 ^b	0.23
NFE (%)	42.62 ^c	58.74 ^b	61.18 ^a	55.25 ^b	1.06
ADF (%)	31.48 ^a	17.82 ^c	24.38 ^b	15.38 ^c	0.23
NDF (%)	61.45 ^a	43.91 ^c	50.74 ^b	34.28 ^c	0.20
Hemicellulose	-29.97 ^a	-26.09 ^b	-26.36 ^b	-18.90 ^c	0.03

abc - Mean on same row with different superscripts are significantly different (P<0.05), NFE - Nitrogen Free Extract, ADF - Acid Detergent Fiber, NDF - Neutral Detergent Fiber, SEM - Standard error of mean.

Mineral content of processed G. arborea leaves using different processing techniques. The result of the percentage mineral content of GAL with different processed techniques is shown in Table 2. GAL processed as the fresh recorded a higher value for phosphorous 0.390 (mg/L), sodium (162.034 mg/L), copper (4.409 mg/L), iron (52.830 mg/L) and calcium (0.018 mg/L), than the sun-dried, air-dried and the boiled-dried treatments, while in the air-dried treatment, potassium (2.856 mg/L) and magnesium (0.260 mg/L) recorded a higher (P<0.05) value than all other treatments. In the boiled-dried treatment, zinc (0.230 mg/L) content was higher than for other treatments. However, apart from zinc, the boiled-dried treated GAL recorded the least value for any other mineral analyzed.

Table 2 Mineral content of *Gmelina arborea* leaves processed using different processing techniques

	Processing technique				
Minerals (mg/L)	Fresh	Sun-dried	Air-dried	Boiled-dried	SEM
	T1	Т3	T4	T5	
Phosphorous	0.390^{a}	0.360 ^b	0.360 ^b	0.360 ^b	0.005
Zinc	0.0250^{c}	0.0220^{c}	0.100 ^b	0.230^{a}	0.002
Potassium	2.819^{a}	1.224 ^b	2.856 ^a	0.817 ^c	0.271
Sodium	162.034 ^a	98.004 ^c	158.600 ^b	84.602 ^d	10.508
Magnesium	0.028 ^d	0.140 ^b	0.260^{a}	0.078 ^c	0.037
Copper	4.409^{a}	3.562 ^b	4.022 ^a	2.032 ^c	0.272
Iron	52.830 ^a	32.081 ^c	44.620 ^b	19.798 ^d	3.779
Calcium	0.018^{a}	0.014 ^c	0.017 ^b	0.014 ^c	0.001

abcd - Means on same row with different superscripts differ significantly (P<0.05), SEM - Standard error of mean.

The fresh leaves had DM value of 92.54%. These DM values are within the range (80–93%) reported for *G. arborea* leaves based diets in earlier investigations (Adamu et al 2013) but higher than those reported by Babayemi (2009) (55.5%) and Okagbare et al (2004) with 40.87. The differences may be due to the effects of the different processing techniques adopted.

The ash and crude fibre values were relatively higher for the fresh and the boileddried treatments. The mineral content of the fresh G. arborea leaves in this study, as shown in Table 2, was higher than of those reported by Adamu et al (2013) but similar to other browse plants reported by Osuntokun & Olajubu (2014), which makes the plants in study rich in minerals and could serve as a complete fodder for livestock. The CP values obtained with the various processing techniques in this study, is comparable with previously reported values (Adamu et al 2013; Okagbare et al 2014). The crude protein value of G. arborea leaves obtained in this study are similar to crude protein values reported by Babayemi (2009) for other browse plants such as Albizia lebbeck (22.3%), Gliricidia sepium (24.7%), Leucaena leucocephala (23.8%), Parkia biglobosa (17.9%), Enterolobium cyclocarpum (19.0%) and Vernonia amygdalina (29.1%). The crude protein value in this study also compares favorably with the crude protein values of some other browse plant such as Bamubusa vulgaris (22.38%), Mangifera indica (15.13%) and Newbouldia laevis (15.57%) that have been evaluated and integrated into ruminant feeding (Ikhimioya 2005; Osuntokun & Olajubu 2014). The crude protein of G. arborea leaves in this study is also higher than those reported by Babayemi (2009) and Osuntokun & Olajubu (2014).

The crude protein values obtained in this study (18.05–20.95%) are far above 7% recommended value for tropical livestock by Minson (1990), below which there will be a deficiency in performance. The CP value in the fresh sample was slightly higher than the concentration observed for the boiled-dried treatment. The apparent lower CP value recorded for the boiled-dried *G. arborea* leaves diet may be attributed to the influence of heat treatment which perhaps may have resulted in denaturation of some protein components thereby given lower CP values. Toasting as a processing method has been shown to lower the CP content of pigeon pea relative to raw seed (Ahamefule & Udo 2010). The high crude protein content recorded for *G. arborea* leaves in this study may be considered as an important factor in the utilization of *G. arborea* leaves in ruminants' diet because feed intake by ruminants is increased by the increase in crude protein content of diets (Alderman 1980). It is quite obvious that the browse plant under study (*G. arborea*) has a high potential compared to other available browse plants.

The ether extract (EE), nitrogen free extract (NFE), acid detergent fibre (ADF) and neutral detergent fiber (NDF) values of the experimental diet containing fresh *G. arborea* leaves are within the range of what has been reported by other researches on the same specie (Babayemi 2009; Okagbare et al 2005; Adamu et al 2013). But the values

obtained for the other processing techniques were rather low probably due to the various processing techniques adopted. However the *G. arborea* leaves used in this study has a fairly high level of nutritional composition and this suggests that they are potential sustainable feed resources that could be used in ruminant feeding for optimum performance. These values recorded in the present study are closely related to the reports of Onabanjo & Onwuka (1998), Okagbare et al (2004), Adamu et al (2013) and Okagbare et al (2014). Also, they are in accordance with the nutritional requirements recommended for ruminants (Idahor 2006). However, the lower values observe for the crude protein variations observed in this study could be attributed largely to the processing methods adopted (FAO 2000; Mecha & Adegbola 2004). Also, the observed disparities in the processing methods evaluated in this study could be largely due to the effect of the heating techniques which lead to the denaturation of some nutrients thereby enhancing the concentration of others (Idahor 2006). These findings provide an impetus for adoption of these processing techniques for improvement for ruminant feeding.

Anti-nutritional components of GAL processed with different techniques. The results of the anti-nutritional content of *G. arborea* leaves, using different processing techniques are presented in Table 3. There were significant (P <0.05) differences in the anti-nutrient concentrations among the different processing techniques of *G. arborea* leaves. The value observed for the anti-nutritional factors present in the fresh *G. arborea* leaves were higher (P<0.05) for tannin (4.40 g/L), alkaloid (6.74 g/L), saponin (1.51 g/L), oxalate (17.07 g/L), flavoniod (9.64 g/L) and steroid (43.47 g/L) than for the sundried, air-dried and the boiled-dried processed leaves. The sundried and the air-dried processed *G. arborea* leaves recorded lower values of ANFs than the fresh *G. arborea* leaves, while the boiled dried processed leaves recorded the lowest value of ANFs (P<0.05) for tannin, alkaloid, saponin, oxalate, flavoniod and steroid recording 1.64 g/L, 0.01 g/L, 0.01 g/L, 10.61 g/L, 3.11 g/L and 0.01 g/L respectively.

Table 3
Anti-nutritional components of *Gmelina arborea* leaves subjected to different processing techniques

Anti-Nutrient	Fresh T1	Sun-dried T3	Air-dried T4	Boiled-dried T5	SEM
Tannin (g/L)	4.40 ^a	2.44 ^b	2.37 ^b	1.64 ^c	2.31
Alkaloid (g/L)	6.74 ^a	1.32 ^b	1.05 ^b	0.01 ^c	0.35
Saponin (g/L)	1.51 ^a	1.32 ^b	1.05 ^c	0.01 ^c	0.28
Oxalate (g/L)	17.08 ^a	14.44 ^b	15.52 ^b	10.61 ^c	2.86
Flavonoid (g/L)	9.64 ^a	4.50 ^b	3.89 ^b	3.11 ^c	2.43
Steroid (g/L)	43.47 ^a	40.27 ^b	41.47 ^b	0.00^{c}	18.39

abc - Mean on same row with different superscripts are significantly different (P<0.05), SEM - Standard error of mean

The tannin concentrations in the present study were fairly higher in the fresh samples and lower in the processed treatments. They are higher than 0.62% detected in sesbania (Reed et al 1988) and 2.05% found in Gliricidia (Ahn et al 1989). But they were however lower than the range of 3-4% in Leucaena reported by D'mello (2000).

The tannin concentration in this study is similar to the tannin of some selected tropical plants sucs as *Ficus poacellie, Lannea coromandelica* and *Hibiscus tiliaceus* (Osuntokun & Olajubu 2014). Idahor (2006) reported that tannin concentrations greater than 4% depressed feed intake in ruminants. Barry (1989) demonstrated with *Lotus pedunculatus* that the ideal concentration of condensed tannins in forage legume is between 2-4% of the diet dry matter at which level they bind with dietary proteins during mastication and appear to protect the protein from microbial attacks in the rumen. This is in agreement with the tannin concentration found in the present study. According to Barry (1989), tannins become highly detrimental at higher level (5-9%) as it reduce

digestibility of fibre in the rumen and inhibiting the activity of bacteria (Chesson et al 1982) and anaerobic fungi (Wahid 1990). Conversely, tannins in feed may have detrimental effects on rumen function when the basal diet is low in protein. However, there is now a possibility of utilizing these ANF to nutritional advantages in ruminant nutrition (Idahor 2006). This is evident in an investigation described by Barry & McNabb (1999) that 2-4% tannin in a diet protected protein from rumen degradation and increased absorption of essential amino acids. Other positive effect of tannin in animal feeding as reported by Essien et al (1983) includes: increased efficiency of protein utilization, reduction of parasite burden, reduction of proteolysis during ensilage, bloat prevention, increases quality of animal products and defaunate rumen.

The saponin concentration determined in this study were fairly high in the chopped treatment which probably led to the depression of growth rate, inhibition of small muscle activities and reduction in nutrient absorption by the experimental animals in the chopped treatment. This is in agreement with the earlier report of Tadele (2015) which states that saponin can affect animal performance and metabolism through erythrocyte haemolysis, reduction of blood and liver cholesterol, depression of growth rate, bloat in ruminant animals, inhibition of smooth muscle activity and nutrient absorption. They also reported that oxalic acid on the other hand bind calcium and form calcium oxalate which adversely affects the absorption and utilization of calcium in the animal body. The relatively poor performance of animals fed fresh diet may be attributed to their high oxalate concentration.

Nevertheless, the anti-nutrients determined in this study appeared to be within the tolerable levels for ruminant nutrition (Idahor 2006). More so, the concentration of saponin, flavonoid, alkaloid, oxalate of G. arborea leaves determined in this study are similar to the concentration of ANF and are within the range of Gliricidia sepium, Leucaena leucocephala, Albizia lebbeck, Desmodium sp reported by Idahor (2006) but are lower than the anti-nutrients of selected browse plants such as Ficus sp, G. sepium, L. leucocephala, G. arborea, A. lebbeck, Desmodium sp, Sesbania sp, Desmanthus sp, Calliandra sp and Prosopis sp reported by Osuntokun & Olanjubu (2014). More significantly, it was observed that sun drying, air drying and boil drying reduced the ANF concentrations in G. arborea leaves. This observation agreed with the report of D'Mello (2000) and Idahor (2006). More so, Osuntokun & Olanjubu (2014), stated that boiling, simmering and blanching causes significant reduction in the level of cyanide content of Moringa oleifera leaves. While Mada et al (2012), further stated that boiling also reduces oxalate content of Arachis hypogaea which is in agreement with the reduction in the oxalate concentration of the processed dietary treatment in this present study. Superiority of sun-dried, air-dried and boiled-dried (respectively) over fresh treatment as observed in this study explains the need for browse processing preferably into hay form before utilization in ruminant nutrition (Idahor 2006; Ahamefule & Udo 2010). The decline in the anti-nutrients concentrations and the observed concomitant improvement in the nutritional values are probably due to the processing technique of G. arborea leaves and heat labile nature of the ANF. Comparative study of the effect of processing on the ANF of G. arborea leaves showed that boiled-dried was better than sun dried, which was in turn better than air-dried technique. These findings are in agreement with the report of Idahor (2006) and Ahamefule & Udo (2010), although boiled-dried method was not studied in their findings, which proved to be the best method in the present

Conclusions. In all the processing methods analyzed in this study, the boiled-dried method appears to be the best in terms of nutrient composition and detoxification of antinutritional factors. However, since *G. arborea* is a drought resistance plant, the availability of its leaves at the peak of the dry season is an advantage and can be used as supplement for ruminant feeding all year round.

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