

Effect of Treatment Methods on the Nutritive Quality of Elephant-Ear Seeds (*Enterolobium Cyclocarpum* Jacq Griseb) as Feed for Ruminant Production

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ABSTRACT

The study was carried out to evaluate the nutritive quality of *Enterolobium cyclocarpum* seeds. Matured pods of *E. cyclocarpum* were handpicked and seeds were manually separated. Seeds were either toasted, boiled or untreated (raw). The experiment was laid out in a completely randomized design consisting of three treatment groups replicated four times with a total seed of 0.5 kg per replicate. The result of the chemical analysis showed that there were significant differences ($P < 0.05$) for all the parameters investigated. The crude protein content was significantly highest ($P < 0.05$) (24.9%) in boiled *E. cyclocarpum* seeds and least (22.4%) in the raw seeds. Untreated (raw) seeds recorded highest ($P < 0.05$) contents for all the secondary metabolites (saponin, tannin, oxalate and trypsin) investigated in this study while boiled seeds recorded lowest. Treatment methods had a significant ($P < 0.05$) effect on the *in vitro* gas production of *E. cyclocarpum* seeds with the boiled seeds having highest gas production (72 ml/200 mg DM at 48 hours of incubation). The study showed that boiling of *E. cyclocarpum* seeds improved its chemical composition and gas production, suggesting that moist heat treatment is preferable when making use of seeds in ruminant diets.

Keywords: Anti-nutritional factors, chemical composition, *Enterolobium cyclocarpum* seeds, gas production, processing

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INTRODUCTION

Ruminants face problems on the availability of high quality feeds throughout the year

and negatively affects their performance and productivity. Supplementation of concentrates in the diet of animals can achieve high productivity. However, conventional seed sources such as groundnuts and soybean are scarce and expensive. There is a need to find alternative sources of protein to enhance the nutritional value of poor quality roughage feed provided to ruminants in the dry season, which is both cheap and easily available (Sarnklong et al., 2010).

Forage seeds have been reported to be high in crude protein and hold promise as a feed for ruminants (Babayemi & Bamikole, 2006). Recent trials revealed that some of the indigenous multipurpose tree species especially *E. cyclocarpum* are evergreen and produce seeds that have a very high protein content (Arigbede et al., 2008). The tree is a legume tree which belongs to the family Mimosadeae (Idowu et al., 2013) and has been used in intensive feed garden. As a leguminous multipurpose plant, it has the potential of fixing atmospheric nitrogen into the soil and can also be exploited for feeding ruminants. Studies have shown that the seeds produce higher volatile fatty acids on degradation by rumen microbial organisms (Babayemi et al., 2004). Arigbede et al. (2008) further reported that these seeds are edible and contain substantial nutrients to support high productivity in animals. Although, the seeds are very rich in protein, they are however, constrained by high content of anti-nutritional factors, arising from secondary metabolism in plants.

Anti-nutritional factors can become detrimental to animals if their concentration is high, lowering feed intake and reducing rumen microbial activity and growth (Soetan & Oyewole, 2009). Hence, there is need to reduce or minimise the effects of these compounds in animal feed materials by treating the seeds. Most of the toxic and anti-nutrient effects of phytochemicals compounds in plants could be reduced by several processing methods such as soaking, germination, boiling, autoclaving, fermentation, genetic manipulation and other processing methods (Soetan, 2008). *In vitro* gas production is inexpensive and a not time-consuming method for evaluating nutritive value of feeds for ruminants (Makkar, 2002). Although gas production is a nutritional wasteful product, it provides a useful basis from which metabolizable energy, organic matter digestibility and short chain fatty acids may be predicted (Oloche et al., 2013).

Browse plants seeds are very rich in nutrients, but there is insufficient information on their nutritive value and utilization for ruminants following processing. The objective of this research is to determine the effect of processing *E. cyclocarpum* seeds on their nutritional quality and use in ruminants' diet.

MATERIALS AND METHODS

Experimental Site

The experiment was carried out at the College of Animal Science and Livestock

Production farm, Federal University of Agriculture, Abeokuta, Nigeria. The area lies within the savannah agro-ecological zone of Western Nigeria (Latitude 7° 13 49.46 N, longitude 3° 26 11.98 E, average annual rainfall of 1037 mm). Temperatures are fairly uniform with daytime values of 28–30°C during the rainy season and 30–34°C during the dry season with the lowest night temperature of around 24°C during the harmattan period between December and February.

Sourcing, Collection and Processing of Test Ingredients

Matured pods of *E. cyclocarpum* which were planted in the multi-purpose trees garden of the research farm in 2004 were handpicked during the dry season of 2015, after falling off from the tree stands. The collected pods were sun-cured for three days and dehulled to obtain the seeds. The seeds were then divided into three parts and treated by boiling 500 g of seeds in 1 litre of water on hot plate (Stuart, heat stir, CB162, United Kingdom) with four replicates (100°C for 60 minutes), toasting of 500 g seeds per replicates on hot plate (Stuart, heat stir, CB162, United Kingdom) (170°C for 15 minutes) (Ogunsakin, 2014) and untreated (raw seeds) (control). In the boiling method, water was used as the heat medium and toasted seeds were directly heated.

Experimental Design

The experiment was laid out in a completely randomized design consisting of three

treatment groups replicated four times with a total seed of 0.5 kg per replicate.

Chemical Composition Analyses

Samples of different treatments of *E. cyclocarpum* seeds were oven dried at 65°C until constant weight was obtained and ground to pass through 1 mm sieve using laboratory hammer mill (Model DFZH-Bühler, Uzwil, Switzerland) and analysed for proximate composition (dry matter (DM), crude protein (CP), ether extract (EE) and ash contents) according to the method of AOAC (2010). Fibre fractions analysis (acid detergent fibre (ADF), neutral detergent fibre (NDF) and acid detergent lignin (ADL)) were determined as described by Van Soest et al. (1991). Calcium and Phosphorus contents were determined by atomic absorption spectrophotometry (Fritz & Schenk, 1979). Anti-nutritional factors such as tannins was determined according to the procedures of Jaffe (2003), saponin was determined according to the procedures of Obdoni and Ochuko (2001), oxalate according to Munro (2000) while trypsin was determined as describe by Prokopet and Unlenbruck (2002) cited by Steve and Babatvnde (2013). The *in vitro* gas production was determined following the procedure of Menke and Steingass (1988). Upon recording the final gas volume at the end of incubation (48 h), the lower end of the syringe was connected to the lower end of a pipette containing 4.0 ml of NaOH (10 M). The NaOH (10M) was then introduced from the latter into the incubated contents,

thereby avoiding gas escape. Mixing of the contents with the NaOH solution allowed for the absorption of CO₂, with the gas volume remaining in the syringe considered to be CH₄ (Anele et al., 2011).

Organic matter digestibility (OMD) were estimated as $OMD = 14.88 + 0.889 GV + 0.45 CP + 0.651 \text{ ash}$ (Menke and Steingass, 1988). Short-chain fatty acids (SCFA) were estimated as $SCFA = 0.0239 GV - 0.0601$ (Getachew et al., 2000). Metabolizable energy (ME) were calculated as $ME = 2.20 + 0.1357 GV + 0.0057 CP + 0.0002859 EE2$ (Menke and Steingass, 1988). Total gas volume (GV) was expressed as ml/200mgDM, CP and ash as percentage DM, ME as MJ/kgDM and SCFA as $\mu\text{mol/g DM}$.

Statistical Analysis

The variance of the data was analysed and significant treatment means separated using Duncan's Multiple Range Test using SAS (2009) Package.

RESULTS AND DISCUSSION

Chemical composition of different treatments of *E. cyclocarpum* seeds are presented in Table 1. Treatment methods significantly ($P < 0.05$) influenced the chemical composition of *E. cyclocarpum* seeds. Boiled seeds had the highest value of 94.0% for dry matter content while least value of 86.0% was for raw seeds. The crude protein content was highest ($P < 0.05$)

Table 1
Effect of treatment methods on the chemical composition of *Enterolobium cyclocarpum* seeds

Parameters	Untreated (raw)	Boiled	Toasted	SEM
Dry matter (%)	86.0 ^c	94.0 ^a	92.0 ^b	1.24
Crude protein (%)	22.4 ^c	24.9 ^a	22.8 ^b	0.40
Ash (%)	4.5 ^a	3.9 ^c	4.2 ^b	0.10
Ether extracts (%)	15.0 ^a	10.3 ^c	11.7 ^b	0.77
Neutral detergent fibre (%)	33.0 ^c	41.0 ^a	34.0 ^b	1.36
Acid detergent fibre (%)	12.0 ^a	9.0 ^c	11.0 ^b	0.67
Acid detergent lignin (%)	6.0 ^a	3.0 ^c	4.0 ^b	0.51
Calcium (g kg ⁻¹)	6.6 ^b	6.6 ^b	8.6 ^a	0.33
Phosphorus (g kg ⁻¹)	6.0 ^b	7.2 ^a	5.8 ^b	0.28
Saponin (%)	3.1 ^a	2.0 ^c	2.5 ^b	0.17
Tannin (mg kg ⁻¹)	3.2 ^a	2.0 ^c	3.0 ^b	0.18
Oxalate (mg kg ⁻¹)	12.8 ^a	10.6 ^c	12.1 ^b	0.33
Trypsin (g kg ⁻¹)	0.4 ^a	0.1 ^c	0.2 ^b	0.04

^{a, b, c, d}: Means within the same row with different superscripts are significantly different ($p < 0.05$) according to Duncan Multiple Range Test
SEM = Standard Error of Mean

(24.9%) in boiled and least (22.4%) in raw seeds. The highest CP content in boiled seeds in this study could have been due to reduction in anti-nutritional factors as reported by Deshpande et al. (2000). The crude protein content of *E. cyclocarpum* seeds in this study was higher than the value reported by Babayemi (2006) for some seeds. The variation could be due to varying times when seeds were collected and their handling. The range of CP of seeds in this study would provide adequate nitrogen requirement by rumen microorganism to maximally digest the main components of dietary fibre leading to the production of volatile fatty acid (Lamidi & Ogunkunle, 2016).

The values obtained for neutral detergent fibre of processed seeds in this study ranged from 33.0 to 41.0% in raw and boiled seeds respectively while acid detergent fibre ranged from 9.0 to 12.0% in boiled and untreated (raw) seeds respectively. Neutral detergent fibre contents of *E. cyclocarpum* seeds in this study followed the same trend as recorded for the crude protein contents. The appreciable NDF value in boiled seeds (41.0%) from this study implies higher dry matter intake for animals (Bamikole et al., 2004). Acid detergent lignin content was significantly ($P < 0.05$) highest (6.0%) in untreated (raw) seeds compared with other treatment methods. Lower ADL in the treated seeds in this study could have been due to the effects of heat treatment according to report of Singh and Harvey (2009).

Calcium content of the processed seeds was highest (8.6 g kg⁻¹) in toasted

seeds while the contents in the raw and boiled seeds were similar. The contents of phosphorus were highest (7.2 g kg⁻¹) in boiled and lowest (5.8 g kg⁻¹) in toasted seeds. This observation is similar to the work of Sotolu & Faturoti (2008) where higher mineral contents in *Leucaena leucocephala* seeds that were either toasted or put in hot water was reported. The higher mineral contents of treated seeds in this study seems not explainable as one can only speculate on the probable cause. The NRC (2001) recommends that calcium and phosphorus rates should be at least 6.5 g kg⁻¹ and 4.0 g kg⁻¹ respectively, of the total ration DM for productive cows. Indeed, for the seed processing method in this study, calcium and phosphorus were found to be higher than recommended values.

Values for all secondary metabolites (saponin, tannin, oxalate and trypsin) observed in this study followed a similar trend with the untreated (raw) *E. cyclocarpum* seeds recording the highest contents while the boiled seeds scored the lowest. Treated seeds proved to reduce the secondary metabolites of *E. cyclocarpum*. In this study, boiled seeds gave the least tannin, saponin, oxalate and trypsin contents. This may be attributed to moist heat compared to toasting which is dry heat application. Treatment of seeds, toasting and boiling, have been reported to decrease the anti-nutritional factors of browse seeds (Wiryan, 1997).

The chemical composition of the treated seeds revealed that they have the potential to fill the gap in ruminant nutrition, especially crude protein content particularly in the

dry season. Supplementing browse plants seeds with grasses could solve the problem of feeding ruminant livestock during this season.

Table 2 presents the effects of treatment on the *in vitro* gas production of *E. cyclocarpum* seeds. Treatment methods had significant ($P < 0.05$) effect on the *in vitro* gas production of *E. cyclocarpum* seeds. In all the treatments, the volume of gas produced consistently increased ($P < 0.05$) from the beginning of 3 hours with increasing hour of the *in vitro* gas incubation. Boiled seeds recorded the highest value while toasted recorded the least for all the incubation periods. Gas production basically results in the fermentation of carbohydrate into volatile fatty acids (Getachew et al., 1999). The amount of gas released when feeds are incubated *in vitro* has been reported to be closely related to digestibility of feed for ruminants (Mebrahtu & Tenaye, 1997).

Therefore, the highest gas production observed for boiled seeds suggests a higher digestibility of the seeds than the other treatments. This could be a reflection of a higher proportion of carbohydrate available for fermentation. The high gas production in the present study for all the seeds incubated could be the result of high crude protein contents (Babayemi et al., 2009). The higher CP and other nutrients content in the seeds are essential for growth of rumen micro-organisms that degrade feedstuff prior to gastric and intestinal digestion by the host animal (Reed et al., 1990). Moreover, gas production has been reported to be positively related to microbial protein synthesis (Hillman et al., 1993). This is nutritionally significant to the ruminants because this serve as an indicator to how dry matter is being degraded in the rumen of ruminants.

Table 2
Effect of treatment methods on the *in vitro* gas production (ml/200 mg DM) of *Enterolobium cyclocarpum* seeds

Treatments	Incubation period (hours)					
	3	6	12	24	36	48
Untreated (raw)	14.0 ^a	24.0 ^b	38.0 ^b	46.0 ^b	50.0 ^b	50.0 ^b
Boiled	16.0 ^a	30.0 ^a	54.0 ^a	70.0 ^a	70.0 ^a	72.0 ^a
Toasted	6.0 ^b	14.0 ^c	24.0 ^c	34.0 ^c	34.0 ^c	42.0 ^c
SEM	1.63	2.40	4.37	5.32	5.24	4.63

^{a, b, c, ...} Means within the same column with different superscripts are significantly different ($P < 0.05$) according to Duncan Multiple Range Test
SEM = Standard Error of Mean

Table 3 shows the effects of treatment on the post incubation parameters of *E. cyclocarpum* seeds. The parameters

investigated followed similar trend in all the processed seeds. Boiled seeds recorded significantly ($P < 0.05$) highest values for

metabolizable energy (ME), organic matter digestibility (OMD) and short chain fatty acid (SCFA), followed by raw seeds and the least values were in toasted seeds. The SCFA value is an indication that the energy content in feeds will be readily utilized after digestion. The high metabolizable

energy, organic matter digestibility and short chain fatty acid that were reported in this study for the differently processed *E. cyclocarpum* seeds, could translate to higher dry matter intake in ruminants for an improved performance.

Table 3

Effect of treatment methods on the post incubation parameters of *Enterolobium cyclocarpum* seeds

Treatments	Organic matter digestibility (%)	Metabolizable energy (MJ/kgDM)	Short chain fatty acid ($\mu\text{mol}/200 \text{ mg DM}$)
Untreated (raw)	68.8 ^b	8.6 ^b	1.0 ^b
Boiled	81.9 ^a	10.5 ^a	1.4 ^a
Toasted	58.1 ^c	7.0 ^c	0.8 ^c
SEM	5.1	1.5	0.1

^{a, b, c,...} Means within the same column with different superscripts are significantly different ($P < 0.05$) according to Duncan Multiple Range Test

SEM = Standard Error of Mean

The amount of methane (CH_4) in the present study ranged from 24 $\mu\text{mL}/200 \text{ mg DM}$ in raw to 29 $\mu\text{mL}/200 \text{ mg DM}$ in boiled *E. cyclocarpum* seeds (Figure 1). Highest anti-nutritional factors in the raw *E. cyclocarpum* seeds could have been responsible for its lowest methane production especially for saponin which had the potential to slightly

depress methanogenesis because of its capacity to suppress protozoa, the main butyrate producers in the rumen (Babayemi et al., 2004). Hess et al. (2004) also reported that tannin suppressed methane production from *Calliandra calothyrsus*, a tropical legume.

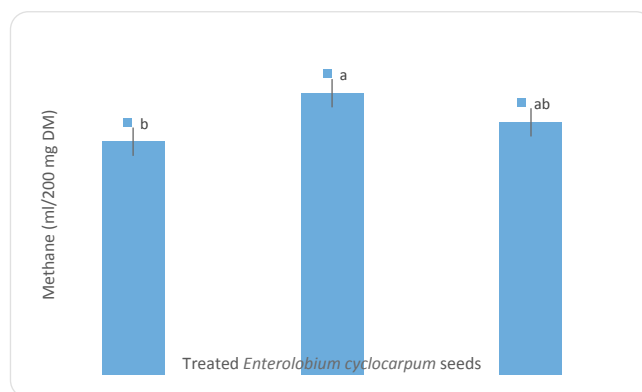


Figure 1. Effects of treatment methods on the methane (ml/200 mg DM) production of *Enterolobium cyclocarpum* seeds following incubation period of 48 h

CONCLUSION

This study found that treating *E. cyclocarpum* seeds by boiling improved its chemical composition and *in vitro* gas production, thus suggesting moist heat treatment of the seeds if they are to be incorporated into animal feeds. Livestock farmers are therefore advised to propagate the tree, since the seeds are rich in nutrients.

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