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# *Aspergillus niger* Degraded Shea Butter Cake and Leaves (*Vitellaria paradoxa* )

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## Abstract

The study investigated the proximate and phytonutrient composition of Shea butter cake and leaves treated with and without *Aspergillus niger* for a two week period. Shea butter leaves were collected from the University of Ilorin and its environs while the cake was collected from Shea butter oil factory within Ilorin metropolis. The treatment protocols were A (Untreated fresh Shea butter leaves), B (Fungus treated fresh Shea butter leaves), C (Untreated Dried Shea butter leaves), D (Fungus Treated dried Shea butter leaves), E (Untreated Shea butter cake), F (Fungus Treated Shea butter cake). Proximate composition (which was of six determinations for each sample) was determined according to the method of AOAC while phytonutrient was determined using Atomic Spectrophotometer and the Crude fibre by the method of Goering and Van-Soest. All data collected were subjected to 'T' test analysis and Analysis of Variance of a Completely Randomised design model. The results revealed a significant ($P<0.05$) dry mater content of the samples with high percentage recorded for Treatments C (93.4%) and E (92.8%). The highest Crude Protein content was observed in Treatments E (18.16%) and F (18.59%). The highest Crude fibre content was recorded for Treatments C and D while other Treatments were of similar values ($P>0.05$). It is interesting to note that Treatments E and F had highly significant ($P<0.05$) ether extract compared to other Treatments which were similar ($P>0.05$). A value of 16.15% was observed as Ash percentage for Treatment E compared to other Treatments. Highest Lignin percentage (59.70%) was found in Treatment C and the least of 31.4% for Treatment B. The ADF was 53.9% (A), 38.1% (B), 68.8% (C), 54.6% (D), 56.2% E and 46.9% (F). The cellulose content was B > F > A> D>E > C respectively. Contrarily, the tannin content was C >D >E >F >A > B. The mineral contents were significantly higher for the fungus treated samples compared with the untreated samples. This study concludes that Shea butter cake and leaves could be a good source of dry season feeding for livestock animals due to its rich nutrient contents

Keywords: Shea butter cake, leaves, chemical composition, fibre fractions *Aspergillus niger*, mineral content

## Introduction

Africa is a livestock-rich continent representing about one-third of the global livestock population (Macmillan, 2020), its full potential of supplying meat and milk to the teeming African population was hindered due to poor genetic constitution and nutrition. Additionally, high cost of feeding stuffs and poor weather condition constitute another major bottleneck in livestock industry in African. The poor weather condition makes the available forages and grasses to become fibrous easily while existing industrious agricultural wastes constitute nuisance to the environment. These wastes could be useful if treated by adding value to it. Example of such wastes are Shea butter cake and Leaves which are found to be rich in secondary cell wall with high cellulose and lignin content which cannot be adequately broken down by animals resulting in low consumption, low digestibility and poor feed efficiency (Longe and Ogedengbe, 1989).

Shea butter cake is a by-product of oil extraction from the kernel of Shea nut tree (*Vitellaria paradoxa)*. Provision of Shea butter cake will assist in the replacement of expensive conventional feedstuffs like Groundnut cake and Soya bean cake (Olorede and Longe, 1999). Shea butter cake has about 2620k.cal/kg Gross energy and about 17% Crude protein. But the problem with the use of this cake is its bitterness due to the presence of Saponin and Tannin (Belewu and Yahaya, 2008)

However, the rumen contains microbial population which could assist in the fermentation and break down of cellulose materials as well as protosan into simple sugar

Apart from rumen microbial fermentation, fibre fraction can be broken down through various methods like physical, chemical and microbial biotechnology. The physical method is either by grinding/ milling so as to increase the surface area while the chemical method was noted to results in chemical load (sodium load) if treated with Sodium hydroxide) ( Ardnt *et al.*, 1980). The biological method was found to be more effective than any other methods (Belewu *et al.*, 2015a, b). The method involves microbial inoculants and cellulolytic enzymes with easy and safer handling and application to its credits, the method is neither volatile nor corrosive which is aimed at breaking down cell wall to provide a wealth of readily available substrates (Dutton, 1987). Therefore, the thrust of this study was to evaluate the proximate composition and phytonutrient contents of *Aspergillus niger* treated and untreated Shea butter cake and leaves

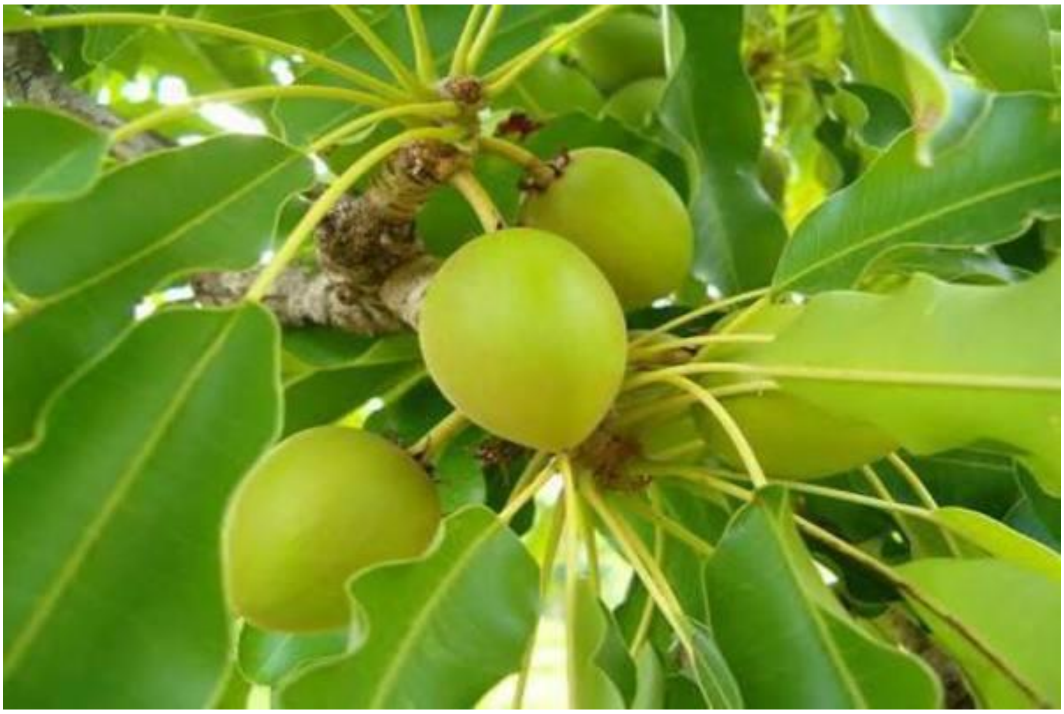

Fig 1 : Fresh shea butter leaves and fruits.

naturalhomes.org

Vitellaria Paradoxa seed ..(alamy.com)

**Materials and methods**

**Study area**

The study was conducted at the Microbial Biotechnology and Dairy Science Laboratory, University of Ilorin, Nigeria. University of Ilorin has Latitude and longitude of 8.4928°N, 4.5962°E respectively and a land mass of approximately 5,000 hectare. (https://www.distancesfrom.com).The temperature falls between 30 and 31$^{O}$C with relative humidity of 26%.

Samples Collection and sampling

Samples were collected from the University environment as well as from Ilorin metropolis.

### Study samples

The study samples were Fresh untreated Shea butter leaves (A); fungus treated fresh Shea butter leaves (B), untreated dried Shea butter leaves, (C), Fungus treated dried Shea butter leaves (D), untreated Shea butter cake (E) and Fungus (*Aspergillus niger)* treated Shea butter cake (F).

### Medium and *Aspergillus niger*

*Aspergillus niger* originally isolated from the soil sample collected from the Teaching and Research farm, University of Ilorin and maintained on Potato dextrose agar (200g), dextrose (20g) and agar (15g) at the Microbial Biotechnology Laboratory.

### Substrate Preparation

The Shea butter Leaves (Fresh and Dry) were collected from around the Department of Animal Production, University of Ilorin. The leaves were destalked, washed and drained and allowed to be airdried in the laboratory while the cake was collected from Shea butter oil factory within Ilorin metropolis Kwara State. The leaves were cut into smaller bits, sterilized in autoclave for 15 minutes at 121$^{O}$C and 15psi and later packed in ventilated containers ready for inoculation with the fungus while the untreated samples were sterilized but not inoculated with fungus

### Inoculation and incubation

One sixth (0.02kg) of the petri-dish content of *Aspergillus niger* was used to inoculate each of the 50g of the sterilized substrates. The inoculated samples were later incubated for 7 days at ambient temperature. Duplicate for each fungal culture was left for three weeks at ambient temperature until fungal growth had covered the substrate surface. The entire content was later dried at 70$^{0}$C in a forced –air laboratory dryer oven. The control samples were un-inoculated but received similar sterilization and incubation as described. Each experiment was repeated six times and the data presented are means of six determinations.

### Treatments Protocol

The substrates protocol include Untreated fresh Shea butter leaves (A); Treated Fresh Shea butter leaves (B); Untreated dried Shea butter leaves (C); Treated dried Shea butter leaves (D); Untreated Shea butter cake (E); Treated Shea butter cake (TF).

### Chemical analysis

Analysis of samples for dry matter, crude protein, crude fibre, ether extract and ash were done according to the methods of AOAC (2006) while the fibre fractions were determined by the methods of Goering and VanSoest (1963).

### Phytonutrients determination

Tannin percentage was determined by the method described by Broadhurst and Jones (1978). The saponin content of the samples was determined by double extraction gravimetric method described by Harborne (1973).

### Data analysis

All data collected were subjected to Student's 'T' test and Analysis of variance (ANOVA) of a Completely Randomised design model (Steel and Torie, 1990) while means were separated by Duncan (1955) Multiple Range test

## Results and Discussion

### Proximate composition

The proximate composition of the Dried untreated and fungus treated Shea butter leaves are presented in Table 1. The dry matter ranged from 87.30 to 93.40%. Treatment A had significantly higher dry matter content as compared with Treatments B (P<0.05). The Crude Protein was higher in Treatment B (5.25%) compared to Treatment A. The higher Protein content could be due probably to the addition of microbial protein during fermentation process. The Crude fibre content of Treatment B was lower than Treatment A due to the biodegradation of fibre of Treatment B by the Fungus. It has been reported that the fungus produced lignocellulolytic enzymes (Villena *et al.*, 2007). The ether extract content of Treatment B was higher than Treatment A which showed that the fungus is lipolytic in nature. The lignin, cellulose and the ADF contents were lower in Treatment B compared to Treatment A which means the fungus could have used the fibre content for their body growth due to the lignocellulolytic enzymes secreted by the fungus. Additionally, the tannin content was significantly decreased in Treatment B compared to Treatment A

Table 2 showed the Chemical composition of fungus treated and untreated Fresh Shea butter cake. The dry matter content ranged between 86.90 and 92.80%. The low dry matter content of Treatment C might be due to the microbial action. Hence, it is evidence from this study that Treatments C and E had higher Dry matter compared to other Treatments. The higher Dry matter of Treatment C could be due probably to the fact that it was already dried. The dry matter is mostly influenced by the weather and environmental factors. The findings in this study concur with other studies which reported that dry matter is influenced by weather and the environmental factors (DAIReXNETdairy-cattle.extension.org). Dry matter is a pointer of the amount of nutrients that are available to the animal in a particular feed. Livestock need to eat a certain amount of dry matter per day (measured in lbs or kg/day) to maintain good health and production.

The slight but significant percentage of Crude Protein could be due to the addition of microbial protein during fermentation process. This corroborates the report of Belewu *et al.* (2015a). The high protein content of Treatments E and F may be attributed to the production of protease by the fungus.

The lower contents of lignin, ADF and Tannin follow similar trend as reported for Fungus treated Dried Shea butter leaves.

With the exception of ether extract and ash content of fungus treated and untreated Shea butter cake, other parameters are significantly lower (P<0.05) for the Fungus treated samples compared with Untreated samples.

Subjecting the results to Analysis of Variance showed that Dry matter ranged from 82.40 to 93.40% whereas Treatment C had Dry matter content of 93.40% which was similar to Treatment E while Treatments A, B, C, D and F were similar due probably to the action of the fungus on the samples. The Crude Protein content of the Fungus treated samples was slightly higher than the untreated samples due to the addition of the microbial protein and various enzymes secreted (protease, cellulose, xylanase, amylase and phytase) during fermentation process.

The higher ether extract of Treatments E and F was expected as it contained cake. The results of this study agreed with the reports of Yahaya and Belewu (2007) which reported that Shea butter cake contain slightly higher ether extract content.

The observed range of ash content in Treatments E and F of this study indicated that the sample is a good source of mineral since ash is a measure of the mineral content of food item (Oladipo and Bankole, 2013).

Conversely, the fibre content was lower for the fungus treated samples compared to the untreated samples. The enzymes secreted (Cellulase, xylanase) by the fungus could have assisted in the degradation of the fibre content and used for their growth. The fibre fractions followed similar trend as the crude fibre content. The assertion supported the report of Jacquline and Visser (1996). The fibre fractions analysed in this study are known to be vital to livestock health hence, fibre in the diet of livestock is advantageous since it helps in enhancing digestion

The result also confirmed that *Aspergillus niger* could be useful in reducing phytonutrient in samples as the result of the Tannin and Saponin content was significantly lower in fungus treated samples.

Furthermore, the results indicated that Treatment F is richer in most of the nutrients than other Treatments. The results corroborate the findings of other studies which compared the composition of Shea butter cake (Yahaya and Belewu, 2007). According to these studies the proximate composition differed based on the various parts of the plant used (fresh and dried leaves as well as the cake),

**Mineral content**

Calcium is needed as a component of diet as it is essential for the full activity of many enzymes (Nitric oxide synthase, protein phosphatase, adenylase kinase) and also vital for the maintenance of optimal bone formation and development (Jacbson *et al.*, 2017). It is also important for proper growth and development of bone, teeth and muscles (Jacbson *et al.*, 2017). The Calcium content reported herein showed that the samples are rich source of Calcium.

Magnesium content which acts as co-factor of pyruvate dehydrogenase and transform pyruvate into Acetyl-CoA which is used in the Citric acid cycle to carry out cellular respiration for the release of energy. The magnesium content in all the samples showed that all the samples used for this study are good candidate for this element.

Phosphorus is vital as it form the structure of teeth and bone as well as cell membrane. This element also acts as a co-factor for various enzymes as well as activates Vitamin B complex. The Phosphorus content reported herein agreed with the reported values of Abdul-Mumeen *et al*. (2013).

Iron is vital in metabolic reaction and regulation of cell growth and differentiation. It is an essential trace element needed for haemoglobin formation, normal function of central nervous system as well as oxidation of carbohydrate, protein and fats and as a catalase-enzyme that catalysis the conversion of hydrogen peroxide to water and oxygen (Shaffer, 2017).

**Conclusion**

The finding of this study reaffirm that Shea butter leaves and cake can provide vital nutrients needed for normal body function and maintenance of livestock mostly during the dry season. The proximate composition reported herein would be useful to livestock farmers in utilizing Shea butter leaves and cake based on their nutritional values

Table 1: Comparative Chemical Composition of Untreated Dried Shea butter leaves and *Aspergillus niger* Treated Dry Shea butter leaves

| Parameters (%) | Untreated Dried Shea butter leaves (A) | *Aspergillus niger* Treated Dried Shea butter leaves (B) | Significant Levels (P<0.05) |
|---|---|---|---|
| Dry matter | 93.40 | 87.30 | A vs B* |
| Crude Protein | 3.94 | 5.25 | A vs B* |
| Crude fibre | 36.3 | 34.95 | A vs B* |
| Ether extract | 4.95 | 5.70 | A vs B |
| Ash | 8.60 | 7.65 | A vs B |
| Lignin | 59.70 | 46.40 | A vs B* |
| ADF | 68.80 | 54.40 | A vs B* |
| NFE | 32.29 | 38.83 | A vs B |
| Cellulose | 4.10 | 4.40 | A vs B |
| Tannin (g/kg) | 0.083 | 0.067 | A vs B* |
| Saponin (g/kg) | 4.50 | 2.34 | A vs B |

Table 2: Comparative Chemical Composition of Untreated Fresh Shea butter leaves and *Aspergillus niger* Treated Fresh Shea butter leaves

| Parameters (%) | Untreated Fresh Shea butter leaves (C) | *Aspergillus niger* Treated Fresh Shea butter leaves (D) | Significant Levels (P<0.05) |
|---|---|---|---|
| Dry matter | 89.10 | 82.40 | C vs D* |
| Crude Protein | 9.63 | 10.50 | C vs D |
| Crude fibre | 25.35 | 22.40 | C vs D* |
| Ether extract | 4.65 | 7.60 | C vs D* |
| Ash | 5.10 | 3.50 | C vs D* |
| Lignin | 47.10 | 31.40 | C vs D* |
| ADF | 53.90 | 38.10 | C vs D* |
| NFE | 47.88 | 48.63 | C vs D |
| Cellulose | 4.80 | 5.40 | C vs D |
| Tannin (ppm) | 0.054. | 0.042 | C vs D* |
| Saponin | 2.10 | 1.25 | Cvs D* |

Table 3: Comparative Chemical Composition of Untreated Shea Butter cake and *Aspergillus niger* Treated Shea butter cake

| Parameters | Untreated Shea butter cake (E) | *Aspergillus niger* Treated Shea butter cake (F) | Significant Levels (P<0.05) |
|---|---|---|---|
| Dry matter | 92.80 | 86.90 | E vs F* |
| Crude Protein | 18.16 | 18.59 | E vs F* |
| Crude fibre | 26.85 | 9.20 | E vs F* |
| Ether extract | 24.80 | 24.95 | E vs F |
| Ash | 10.15 | 10.00 | E vs F |
| Lignin | 44.70 | 34.70 | E vs F* |
| ADF | 56.20 | 46.00 | E vs F* |
| NFE | 10.05 | 23.19 | E vs F* |
| Cellulose | 4.40 | 5.10 | E vs F* |
| Tannin (g/kg) | 0.064 | 0.058 | E vs F* |
| Saponin (g/kg) | 3.2 | 1.90 | E vs F* |

Table 4: Comparative Proximate Composition of Untreated Dried Shea Butter Leaves, Fresh Shea Butter Leaves and Shea Butter cake and Fungus Treated Dried Shea butter Leaves, Fresh Shea Butter and Shea Butter cake

| Parameters (%) | Untreated Fresh Shea butter leaves (A) | *Aspergillus niger* Treated Fresh Shea butter leaves (B) | Untreated Dried Shea butter leaves (C) | *Aspergillus niger* Treated Dried Shea butter leaves (D) | Untreated Shea butter cake (E) | *Aspergillus niger* Treated Shea butter cake (F) | Significant Levels (P<0.05) |
|---|---|---|---|---|---|---|---|
| Dry matter | 89.10$^{b}$ | 82.40$^{b}$ | 93.40$^{a}$ | 87.30$^{b}$ | 92.80$^{a}$ | 86.90$^{b}$ | 3.53 |
| Crude Protein | 9.63$^{b}$ | 10.50$^{b}$ | 3.94$^{d}$ | 5.25$^{c}$ | 18.16$^{a}$ | 18.59$^{a}$ | 1.50 |
| Crude fibre | 25.35b | 22.40c | 36.30a | 34.95a | 26.85b | 9.20d | 2.34 |
| Ether extract | 4.65$^{d}$ | 7.60$^{b}$ | 4.95$^{d}$ | 5.70$^{c}$ | 24.80$^{a}$ | 24.95$^{a}$ | 1.70 |
| Ash | 5.10$^{d}$ | 3.50$^{e}$ | 8.60$^{c}$ | 7.65$^{c}$ | 16.15$^{a}$ | 10.00$^{b}$ | 0.90 |
| Lignin | 47.10$^{b}$ | 31.40$^{e}$ | 59.70$^{a}$ | 46.40$^{b}$ | 44.70$^{c}$ | 34.70$^{d}$ | 1.79 |
| ADF | 53.90$^{b}$ | 38.10$^{d}$ | 68.80$^{a}$ | 54.60$^{b}$ | 56.20$^{b}$ | 46.00$^{c}$ | 3.10 |
| NFE | 47.88$^{a}$ | 48.63$^{a}$ | 38.29$^{b}$ | 38.83$^{b}$ | 10.05$^{d}$ | 23.19c | 2.45 |
| Cellulose | 4.80 | 5.40 | 4.10 | 4.40 | 4.40 | 5.10 | 0.14 |
| Tannin (ppm) | 0.083$^{c}$ | 0.067$^{d}$ | 0.054$^{a}$ | 0.042$^{b}$ | 0.064$^{b}$ | 0.058$^{c}$ | 0.0056 |
| Calcium | 0.25$^{e}$ | 0.35$^{d}$ | 0.45$^{c}$ | 0.65$^{a}$ | 0.56$^{b}$ | 0.70$^{a}$ | 0.020 |
| Magnesium | 0.13$^{c}$ | 0.15$^{c}$ | 0.25$^{b}$ | 0.55$^{a}$ | 0.15$^{c}$ | 0.25$^{b}$ | 0.015 |
| Phosphorus | 0.20$^{d}$ | 0.35$^{c}$ | 0.40$^{b}$ | 0.58$^{a}$ | 0.21$^{d}$ | 0.45$^{b}$ | 0.024 |
| Iron | 1.96$^{b}$ | 2.00$^{b}$ | 2.65$^{a}$ | 2.80$^{a}$ | 1.86$^{b}$ | 2.50$^{a}$ | 0.034 |
| Saponin (g/kg) | 4.50$^{a}$ | 2.34$^{c}$ | 2.10$^{c}$ | 1.25$^{d}$ | 3.20$^{b}$ | 1.90$^{e}$ | 1.04 |

Means with similar superscripts are not significantly different from each other (P>0.05)

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