Tapping in to the good use of cocoa (*Theobroma cacao*) pod husks: towards finding alternative sources of nutrients for animals in Nigeria.

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Abstract

Cocoa-pods husk (CPH) is one of the by-products of cocoa processing. The enormous quantities of these materials are produced annually on the farm after crop harvesting and can serve as raw materials to food processing industry which could be converted to animal feeds. This present research is designed to determine the proximate, antinutrient and mineral contents of selected three different species (which are Criollo, Forastero, Trinitario) of fermented and unfermented cocoa pod husks (CPH). The cocoa pod husks were washed, sundried and later milled into powder. A portion of each sample powder was subjected to fermentation and analyses were carried out. The results obtained showed significant (p<0.05) differences in proximate, antinutrient and mineral compositions in the fermented and unfermented forms of treated CPH. The fermented Trinitaro, Foraste and Criollo species of cocoa pod husk recorded higher crude protein content (8.52%, 12.32% and 12.90%) compared to the (4.83%, 4.35% and 5.21%) of unfermented forms respectively. A significant increase was observed in the ash (11.74%), fat (12.07%) and moisture (9.42%) content of the 3 species of fermented while there was a decrease in their crude fibre (21.50%) contents but significant increase was observed with their carbohydrate (48.81%) contents. There was a significant decrease in the level of sodium and iron content in all the selected fermented husks while a significant increase was recorded with the potassium, calcium, magnesium and manganese contents in all the samples with fermentation. The decrease in antinutritional contents (tannin, oxalate and phytate) was recorded in the 3 fermented cocoa pod husks. The flavonoid content was also significantly increased in the fermented species. It could be suggested that fermentation improves the condition of cocoa pod husks for them to be considered as animal feed ingredients in the nearest future.

Keywords: Cocoa pod husk, Phytate, Flavonoid, Proximate, Anti nutrient.

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Introduction

The exploitation of agro by-products and farm wastes as alternative feed ingredient for poultry and livestock feeding trials has been the current trend in animal production. In many developing countries, a largely untapped potentials of these agro-wastes are yet to be discovered. Some of these by-products include wheat and rice offals, maize bran and brewers dried grains; which can replace conventional feed resources in animal diets without deleterious effects [1]. One of the agricultural by-products that have been utilized in animal feeding trials with optimum results is cocoa pod husk meal [2]. Reports have shown that each ton of dry cocoa (beans) represents ten (10) tonnes of cocoa pod husk [3]. Presently, cocoa pod husks are causing environmental pollution problem in cocoa producing areas of the world. They serve as potential sources of disease transmission when used as mulch in cocoa farms. In West Africa for instance, a very small proportion is used for traditional soap making and as a fuel source. The use of CPH in animal feed production has not really been popular due to high content of antinutrients, theobromine and crude fibre which could pose threat to animal health [4]. However, these deleterious chemicals can be reduced if further processed for advantageous consumption and promote digestibility and are evidently promising to be advantageous to animal health if consumed at appropriate amounts [5]. Various processing methods have been adopted in the processing of cocoa pod husk for inclusion in livestock feed formulation among them include natural fermentation [6], alkali treatment [7] urea treatment [8], fungal treatment [9], enzyme (mannanase) treatment [10], microbial detheobromination [11] and hot-water treatment [12]. This study was therefore designed to determine the chemical status proximate, antinutrients and mineral compositions of fermented and unfermented husks of 3 different cocoa (Criollo, Forastero, Trinitario) species.

Materials and Methods

Collection of cocoa pods husk

Freshly broken composite cocoa pods (Criollo, Forastero, Trinitario) species were obtained from the fermentary units of the Cocoa Research Institute of Nigeria (CRIN) Owena-Ondo. Ondo. Nigeria. The broken pods were washed and sun-dried for 14 days and then milled into powder. The powder of each species was divided into two portions (fermented and unfermented cocoa pod husk powder). The portion to be fermented was thoroughly mixed with 60% deionized water, relative to its weight as ascertained in the *in vitro* natural fermentation study by ref. [13] and bagged in air-tight polythene bags; allowed to ferment anaerobically for 3 days under room temperature. Thereafter, the fermented bags were opened and contents shade dried for 5 days, before being packed, bagged and stored in a

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cool dry place. The unfermented powder was also bagged and stored in a cool dry place for use prior to analysis.

Chemical assay

Proximate composition analysis

The proximate analysis (carbohydrate, fats, proteins and ash) of the CPH samples were determined using AOAC methods. Carbohydrate was determined by difference method [100-(Pr otein+Fat+moisture+ash)]. The nitrogen value, which is the precursor for protein of a substance, was determined by micro Kjeldahl method. The nitrogen value was converted to protein by multiplying to a factor of 6.25. The moisture and ash were determined using weight difference method while dtermination of fat content of the samples was done using Soxhlet type of the direct solvent extraction method. The solvent used was petroleum ether (boiling range 40-60°C). All the proximate values were reported in percentage (%) by AOCS.

The mineral content determination

The minerals were analyzed from the solution obtained by first dry ashing. 2 g of each sample was placed in a crucible and placed in a muffle furnace at 550°C for 5 h to ash and then transferred into a desiccator to cool. The cooled ash was dissolved in 10% HCl and filtered then diluted to 50 ml volume in standard flask with deionised water. The solution was aspirated into the atomic absorption spectrophotometer to obtain the mineral contents. The minerals analyzed include; calcium, iron, potassium, zinc, magnesium and sodium, while phosphorous were determined colorimetricalry by the phosphovanadomolybdate method [14].

Analysis of antinutritional factors

Oxalate: The calcium oxalate content was determined using the method of ref. [15]. This involves the digestion of the sample, precipitation of the oxalate contained in the sample to remove ferrous ions on addition of ammonium hydroxide solution and then titration against permanganate solution (0.05 M KMnO₄) to a faint pink colour, which persisted for 30 seconds. Phytate: was determined using method [16]. Four gram sample was soaked in 100 ml of 2% hydrochloric acid for 3 h and then filtered. 5 ml of 0.3% ammonium thiocyanate solution was added to 25 ml of the filtrate. 53.5 ml of distilled water was also added to the mixture. This was then titrated against a standard iron (III) chloride solution until a brownish yellow colour persisted for 5 min. The phytate content was calculated from the iron determinations, using a 4:1 iron-phytate molecular ratio.

Tannins

To 0.5 ml of extract solution 1 ml of water and 1-2 drops of

ferric chloride solution was added. Blue color was observed for gallic tannins and green black for catecholic tannin [17].

Flavonoid

Four milliliters of extract solution was treated with 1.5 ml of 50% methanol solution. The solution was warmed and metal magnesium was added. To this solution, 5-6 drops of concentrated hydrochloric acid was added and red color was observed for flavonoids and Orange colour for flavones [18].

Statistical analysis

All the analyses were conducted in triplicate and subjected to statistical analysis using analysis of variance (ANOVA). Means were separated using Duncan's multiple range test.

Results and Discussion

The results of the proximate composition of Fermented and unfermented Trinitaro, Forestaro and Criollo cocoa pod husks are presented in Table 1. There was an observed increase in the values of the moisture content in the fermented cocoa pod husks ranging from 8.02-9.42% than the unfermented cocoa pod husks values ranging from 3.90-5.40%. The result of the moisture content is in agreement with what was reported by Igbabul et al. [19] in which the moisture content of fermented cocoyam flour samples were higher than the unfermented. The reduction of the moisture content in the cocoa pod husks might be attributed to the soft and porous texture of the samples after fermentation which results in maximum moisture loss [19]. The ash content of the unfermented to fermented husk samples ranged from 10.48-11.74% for Trinitaro, 9.49-10.51% for Forastero and 8.40-10.97% for Criollo. The ash content values observed in this study were higher than the ash contents reported earlier for unfermented and fermented breadfruit-cowpea blend flours [20]. The decrease in ash content of the unfermented portion can be attributed to possible leaching of soluble mineral elements into fermenting medium which could have resulted in breakdown of the food components into their absorbable forms [19]. The changes in the protein content from unfermented to fermented as observed in the selected cocoa pod husks samples showed an increase from 4.83-6.52% for Trinitaro, 4.35-5.32% for Forestaro and 3.21-6.92% for Criollo. This finding is lower than what was reported by Ozung et al., 2016 but is in agreement with the fact that cocoa pod husk has low nutrient content as a result of its low metabolizable energy [21]. The fat content showed a significant increase (p<0.05) with fermentation with values ranging from 7.18-12.07% for Trinitaro, 7.87-15.06% for Forastero and 7.72-13.47% for Criollo. This increase could be attributed to the extensive break down of large fat molecules

Table 1. Proximate composition of fermented and unfermented selected species of cocoa pod husks.

Proximate%	Trinitaro Unfermented Fermented		Fora	stero	Criollo Unfermented Fermented	
			Unfermente	d Fermented		
MC	3.90° ± 0.02	9.42 ^b ± 0.11	5.29° ± 0.21	8.34 ^d ± 0.05	5.40° ± 0.31	8.02 ^d ± 0.31
Ash	10.48 ^a ± 0.05	11.74 ^b ± 0.05	9.49° ± 0.01	10.51ª ± 0.03	$8.40^{d} \pm 0.06$	10.97ª ± 0.05
Fat	7.18ª ± 0.13	12.07 ^b ± 0.77	7.87 ^a ± 0.03	15.06° ± 0.05	7.72 ^a ± 0.01	13.47 ^d ± 0.01
Crude Fibre	21.50ª ± 0.34	8.72 ^b ± 0.13	20.32 ^a ± 0.05	5.43° ± 0.05	27.25 ^d ± 0.01	9.53° ± 0.03
Protein	4.83ª ± 0.67	6.52 ^b ± 0.33	4.35 ^a ± 0.03	5.32° ± 0.01	3.21 ^d ± 0.03	6.92 ^b ± 0.01
Carbohydrate	52.11 ^a ± 0.05	49.53 ^b ± 0.05	52.68 ± 0.03	48.34° ± 0.01	46.02 ^d ± 0.11	45.09 ± 0.78

Values are means ± standard deviation of triplicates. Values in the same row with different superscripts are significantly different at p<0.05. MC: Moisture content.

to simpler units due to the high activity of the lipolytic enzymes during fermentation [22]. On the other hand, the crude fibre and carbohydrate contents showed significant decrease (p<0.05) with fermentation. The reduction of the crude fibre contents ranged from 21.50-8.72% for Trinitaro, 20.32-5.43% for Forestaro and 27.25-9.53% for Criollo. The low fibre values recorded with fermentation could be due to the activities of microorganisms which partake in the bio-conversion of carbohydrates and lignocelluloses into protein [23]. Antinutrients also known as secondary metabolites in plants have been reported to be highly biologically active. Antinutrients in plants have been evolved to carry out a defense mechanism as one of its biological functions among others and to also reduce the maximum utilization of nutrients such as proteins, vitamins, and minerals thus preventing their optimal exploitation in a food by decreasing the nutritive value. However, these plant chemicals have been shown to be dangerous to health but research has exhibited the fact that it could be advantageous to human and animal health if consumed at appropriate amount [5]. The effect of fermentation on anti-nutritional properties (phytate, tannin and oxalate) of unfermented and fermented cocoa pod husks samples are presented in Table 2. The study showed that unfermented portion contained 1.83 mg/100 g, 0.89 mg/100 g and 0.59 mg/100 g concentration of phytate for Trinitaro, Forestaro and Criollo respectively while the fermented portion contained 0.35 mg/100 g, 0.17 mg/100 g and 0.47 mg/100 g for Trinitaro, Forestaro and Criollo respectively and these values were significantly different (p < 0.05). The same trend of values were also observed for the tannin and oxalate in all the experimented cocoa pod husks. The tannin and oxalate values obtained are lower than the value reported by Gbadamosi and Famuwagun [24] in the literature on the protein isolates of fermented Kariya seeds (underutilized oil-bearing seeds). The result of the phytate content followed the trend of Marfo et al. [25] on a decrease in phytate content with fermentation. The reduction of phytate content may be linked to the activity of the endogenous phytase enzyme and inherent microorganisms that are capable of hydrolyzing the phytic acid into inositol and orthophosphate [26]. The significant decrease in the concentration of the antinutrient could be suggested to take place as a result of fermentation of the cocoa pod husks.

According to Eka [27], fermentation also significantly (P<0.05) reduced antinutrients thus making the substrates less toxic and the minerals content available [27]. In addition, the reduction in antinutritional contents of the cocoa pod husks after fermentation can be suggested as one of the options of increasing the bioavailability of the rich nutrients present in the fermented products [20], which are within the documented acceptable concentration that animals can tolerate without recording any hazard. The quantity of flavonoids was determined and is represented in Table 2. The results showed that there was significant increase (P<0.05) in the content of the flavonoids for all the experimented cocoa pod husks. In the time past the influx of information on flavonoids and their relevant advantage in the life of man and animals continues to substantiate already established fact that flavonoids exert their antioxidant effects by neutralizing and chelating all forms of oxidizing radicals such as the superoxide and hydroxyl radicals. Flavonoids can also act as powerful chain breaking antioxidant due to their ability to donate the electron on their phenolic groups. All these potent activities underlies many of their actions in the body [28]. The results of the mineral composition of the unfermented and fermented cocoa pod husks are represented in Table 3. The findings revealed that some essential elements are present in the unfermented and fermented cocoa pod husks. There was a significant decrease in the level of sodium and iron content in the fermented husks while a significant increase was recorded with the potassium, calcium, magnesium and manganese contents with fermentation. The current study revealed that metals were accumulated to greater or lesser extents by all the three investigated cocoa pod husk species. They contained appropriate amounts of nutrients that were rich in potassium and calcium. The high concentration of potassium in plants will be very important for enzyme activation, starch formation and protein synthesis. Potassium also participates actively in the maintenance of the cardiac rhythm [29], which will be of importance in the production of animal feeds and to the man who will eventually consume the animals. The use of fermentation has in time past formed an integral part of food detoxification processes that is practiced worldwide. A wide variety of fermented foods are produced and packaged for consumption throughout the world. Fermentation is a simple

Table 2. Anti-nutritiona	l contents of fermented a	and unfermented of sele	ected species of cocoa pod husks.
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Content (mg/100 g)	Trinitaro Unfermented Fermented		Fore	staro	Criollo Unfermented Fermented	
			Unfermente	d Fermented		
Phytate	1.83ª ± 0.03	0.35 ^b ± 0.01	0.89° ± 0.21	0.17 ^d ± 0.11	0.59 ^e ± 0.01	$0.47^{f} \pm 0.03$
Tannins	$0.42^{a} \pm 0.06$	0.25 ^b ± 0.03	0.31° ± 0.06	0.13 ^d ± 0.06	0.67° ± 0.01	$0.09^{f} \pm 0.01$
Oxalate	2.95ª ± 0.30	1.25 ^b ± 0.11	1.77° ± 0.06	0.32 ^d ± 0.11	1.02 ^e ± 0.01	$0.24^{f} \pm 0.03$
Flavonoid	10.43ª ± 011	20.97 ^b ± 0.21	11.34° ± 0.21	14.66 [°] ± 0.11	20.36 ^b ± 0.06	28.07 ^d ± 0.21

Values are means ± standard deviation of triplicates. Values in the same row with different superscripts are significantly different at p<0.05.

Sample	Processing method	Na	K	Са	Mg	Fe	Mn
Trinitaro	Fermented	$9.67^{a} \pm 0.60$	14.30 ^b ± 0.11	14.00° ± 0.03	5.60 ^d ± 0.02	2.43° ± 0.03	$0.80^{f} \pm 0.03$
	Unfermented	16.90ª ± 0.01	10.32 ^b ± 0.02	12.32° ± 0.06	4.59 ^d ± 0.03	3.34° ± 0.01	$0.78^{f} \pm 0.03$
Forestaro	Fermented	3.20ª ± 0.11	15.21 ^b ± 0.03	18.00° ± 0.01	8.64 ^d ± 0.03	2.87° ± 0.11	$1.52^{f} \pm 0.01$
	Unfermented	$7.43^{a} \pm 0.06$	9.67 ^b ± 0.03	16.54° ± 0.02	6.45 ^d ± 0.11	4.03° ± 0.03	$1.00^{f} \pm 0.03$
Criollo	Fermented	$6.40^{a} \pm 0.03$	13.55 [♭] ± 0.01	32.00° ± 0.06	5.20 ^d ± 0.03	3.52° ± 0.06	$1.23^{f} \pm 0.06$
	Unfermented	7.32ª ± 0.01	8.87 ^b ± 0.09	15.73° ± 0.03	4.85 ^d ± 0.07	7.37° ± 0.03	1.00 ^f ± 0.03

Values are means ± standard deviation of triplicates. Values in the same row with different superscripts are significantly different at p<0.05.

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process that can be practiced both at an industrial and household scale. it is a known fact that they are presently adopted in underdeveloped countries for the detoxification of alternative food sources explained by Jansman et al. [30] to alleviate the condition of food scarcity that leads to the competition that exists between the production of animal feeds and limitation of human access to food consumption. Agro-product such as cocoa pod husk has continued to gain the interest of researchers towards converting them to valuable uses such as in production of animal feeds which can critically be a valuable resource for improving food security. Some researchers have been carried out by feeding pigs with fresh samples of cocoa pod husks with recorded outcome of results [20]. The present research has worked on the fermented samples of cocoa pod husks by determining the proximate, antinutrients and mineral contents. It was not only cocoa pod husk that had been considered for animal feeding. There are other by-products such as the bagasse and peels from beverages and juice industries, coffee pulp got from the coffee industry and husks from the cereal industries (Graminha et al. [31]) wheat and rice offals, maize bran and brewers dried grains; which can be included in conventional feed resources in animal diets without hazardous effects. It should not be forgotten that if cocoa pod husks are not treated, kept or disposed of properly they are likely to cause pollution to the environment which no doubt will become harmful to human health. Doing the needful on the long run could lead to increased public awareness on the benefits and potential hazards of agricultural wastes particularly in Nigeria. Nevertheless, the safety of these agro-wastes in the production of animal feeds should be verified by carrying out in vivo experiments and clinical studies. In all, these findings have revealed that the fermented cocoa pod husks could exhibit acceptable products in relation to improvement in the examined parameters which are proximate composition, antinutrient and mineral contents.

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Declaration of Conflict of Interests

The author declared no conflicts of interest with respect to the research, authorship and publication of this article.

Conclusion

The analyses of proximate composition, antinutrient and mineral contents of the cocoa pod husks of Trinitaro, Forestaro and Criollo species which revealed their corresponding concentrations present to be at acceptable values could be further looked into for future incorporation into animal feed production and at the same time promote the optimum utilisation of cocoa pod husks which are always hugely accumulated at the point of harvest in various farms. Judging by the findings of this present investigation and in line with earlier researches, it can be inferred that fermentation minimized the concerns posed by tannin, oxalate, the protein-binding action that could be exerted by phytate and significantly improved the proximate, antinutrient and mineral contents of the cocoa pod husks. In all, these could be tapped into to aid the production of profitable animal feed and also reduce food scarcity that is ravaging the developing countries especially Nigeria.

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