Proximate, Mineral and Amino acids compositions of Arachis hypogaea L. cake application in fish feed formulation

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Abstract

The proximate, mineral and amino acids constituents of groundnut (*Arachis hypogaea* L.) cake were determined. The results showed that the groundnut cake had moisture content of 10.40 ± 0.435, crude protein 9.470±435, lipids concentration 16.040±0.264, carbohydrate 53.016±0.166, crude fibre 3.583 ±.2185 and ash content 10.72± 0.384. Minerals in mg/100g in parenthesis for Na (189.216±0.240), K (176.880±0.288), Mg (53.133±0.272), Ca (366.276±0.754), Fe (34.146±0.961), and P (139.706±0.260) were quite adequate. Thirteen amino acids were recorded; nine were essential amino acids and four non-essential amino acids. Tryptophan (0.433±0.233), lysine (3.26±0.23) and valine (2.88±0.23) yielded approximately 50% of the required standard while methionine (0.926±0.240) and threonine (1.479±0.103) produced $^1/_3$ of the optimum requirement. Aspartate (1.080±0.152), a non-essential amino acid was limiting. *A. hypogaea* was found to be rich in Phenylalanine (4.30. ± 0.219), histidine (2.170±0.208), arginine (4.953±0.233), leucine (6.770±0.230), serine (2.356±0.202), glutamic acid (12.886±0.260) and alanine (2.056±0.145). Consequently, groundnut cake can thus be considered as a good source of protein with high nutritional value rather than just being seen as a waste product following lipid extraction.

Key words: *Arachis hypogaea* cake, proximate, mineral, amino acids, feed formulation

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Introduction

Arachis hypogaea, is a legume crop grown mainly for its edible seeds (USDA 2016). It is widely grown in the tropics and subtropics and a native to a region in eastern South America (Weiss, 1983) being important to both small and large commercial producers. *A. hypogaea* grows as an annual crop principally for its edible oil and protein rich kernels seeds, borne in pods which develop and mature below the soil surface. The groundnuts, is a herbaceous plant of which there are varieties, common in the united states, grow up to 30-46 cm high and do not spread. Runner varieties, the most common in the West Africa are shorter and run along the ground for 30-60 cm (Asiedu, 1992). It is classified as both a grain legume and, because of its high oil content, an oil crop (Hymowitz, 1990).

Nutrition plays a very important role in sustaining aquaculture development because the health of a fish, its growth as well as reproduction depends on the sufficient supply of nutrient in quality and quantity without really considering the kind of culture system being practiced. Adequate supply of fish feed must be guaranteed in other to meet the nutritional requirements of the fish being cultured which in turn increases aquaculture production. (Hassan 2001).An increase in the cost of ingredients especially protein based for the production of high quality feed is a major problem for the development of aquacultural industry in Africa and Nigeria especially. Ekanem *et al* (2011) attributed this to the condition of monopolizing trash fish and using it as a major protein source during fish feed formulation. Interestingly, trash fish is facing much competition from humans and livestock (Ekanem *et al* 2010). Several fish nutritionists are working hard to eliminate the problems by looking for cheaper sources of feed ingredients

especially protein that will help maximize fish production and growth which in turn increases profit margin in aquaculture over a short period of time, (Ekanem *et al.*, 2010).

This study analyses the proximate, mineral and amino acids composition of ground nut cake, the end product of lipid extraction from groundnut (*Arachis hypogaea* L.) and its applicability in the formulation of fish feed.

MATERIALS AND METHODS

Collection and preparation of food items

Groundnut cake

Arachis hypogaea L,(Groundnut cake) was purchased from the local market, smashed and ground into pieces using blender (mortar) and put in an air tight container until needed for analysis.

Laboratory analysis of samples.

Analytical studies of the samples were carried out using the facilities of the Analytical Laboratory in Benin, Edo State Nigeria.

Proximate analysis

The moisture content, ash, crude protein, crude fiber and fat were determined using the following A.O.A.C., (2016).

Moisture content determination

The moisture content of the samples was determined by first weighing out 0.53g each sample the samples. of was weighed using a Metlar PM 2000 model of electric balance at 0.00g initial reading. After weighing, samples were oven dried at 110^0c using a laboratory oven model (DHG-9101.1SA). Samples were dried for 7 to 8 hours after which they were then brought out and allowed to cool in desiccators and then reweighed (A.O.A.C., 2012).

Thus, Moisture (%) = $\frac{\text{Weight of wet sample – weight of dried sample}}{} \times 100$

Weight of wet sample

Ash content determination

0.3grams of the sample was transferred into a pre-weighed porcelain crucible and weighed. The crucible was then placed in a muffle furnace for 6 h at 600 °C to burn off all organic matter. The inorganic material does not volatilize at that temperature and is called ash. The furnace was allowed to cool below 200 °C and maintained at this temperature for 20 min. Then the crucible was placed in a desiccators with a stopper top, allowed to cool and then reweighed to measure the ash content (A.O.A.C., 2012).

$$\text{Ash (\%)} = \frac{\text{Weight of Ash}}{\text{Weight of sample}} \times 100$$

Crude fiber:

20- 50ml of 1.25% H_2S0_4 was introduced into 0.3grams of the sample in a beaker and was boiled at specific temperature of 60 ^{0}C for 20-30 minutes, after which the hot solutions was filtered and the residue collected. This was done to remove the acid content in the sample. After the residue was collected, 10ml sodium hydroxide was added to the sample and heated again at 40 ^{0}C for 5mins and filtration was done with a filter paper.

$$\text{Crude fiber (\%)} = \frac{\text{Weight of fibre}}{\text{Weight of sample}} \times 100$$

Crude protein determination

The protein content of the sample was determined using Kjeldahl method which is the standard method for determining protein and other nitrogen containing compounds. 0.03grams of the individual samples were used. Nitric acid was applied in the digestion of the samples for decomposition and conversion of nitrogen to ammonium sulphate. The sample was heated with a

heating mantle device with capacity 2000 ML at random temperature until the color of the sample changed at 70 ^{0}C. The solution was cooled and concentrated sodium hydroxide was added to make the solution alkaline and distilled into a weak acid (boric acid) containing methyl red indicator until the solution turned from red to green. Following distillation, the ammonia was trapped as ammonium borate and quantified by titrating with a strong standard hydrochloric acid until the solutions turned from green to wine to measure the nitrogenous content. The absorbent of the samples was determined using a spectrophotometer. The amount of crude protein was calculated by multiplying the nitrogen content in percent by 6.25 (A.O.A.C., 2012).

Lipid extracts determinations

0.03grams of the sample was put in a conical flask and sealed in order to avoid evaporations. The sample was heated at 50 ^{0}C for about 2 minutes and then filtered immediately into another conical flask using a filter paper. After filtration was completed, the liquid from the filtered sample was heated again using an electric balance (Metlar 2000). The heated sample was weighed upon cooling using Metlar 2000.

The lipid content was calculated using

$$\text{Lipid (\%)} = \frac{\text{Weight of fat/oil}}{\text{Weight of sample}} \times 100$$

Carbohydrate content determination

The total carbohydrate was determined by subtracting the sum of the percentages of moisture, ash, crude lipid, crude protein, and crude fibre content from 100%.

Determination of mineral composition

The digestion of the ash of the sample using perchloric and nitric acid was carried out in other to determine the concentration of potassium and sodium. Readings were taken in the digital flame photometer/spectronic 20 (Bonire *et al*, 1990). Varidomolybdate chlorimetric method was used in the determination of phosphorous. The concentration of magnesium, iron and calcium were determined spectrophotometrically with buck 200 atomic absorption spectrophometer (Buck Scientific Nouwalk) (Essien *et al*, 1992) and absorption compared with absorption of standards of these minerals.

Determinations of Amino acids.

The amino acid profile was determined using high performance liquid chromatography. High-performance liquid chromatography or high-pressure liquid chromatography (HPLC) is a chromatographic method that is always used to separate the individual amino acids and identify compounds that are present in the different samples that can be dissolved in a liquid in trace concentrations as low as parts per trillion. The new method by Ishida *et al* (1981) earlier described by Mohanty *et al* (2012), adapted by Mohanty *et al* (2014) wherein the non-switching flow method that added sodium hypochlorite to all the amino acids for high efficiency and reliability was adopted. Briefly, muscle protein was hydrolyzed with 6N hydrochloric acid at 110°C under anaerobic condition for 24 h. It simply involve the neutralization of hydrolyzed samples with 6N NaOH and derivatized using a kit (AccQ-Flour Reagent, WAT) 052880, Waters). Then the derivatized samples were injected in high performance liquid chromatography (HPLC) (1525, Waters) equipped with a C_{18} RP column and a fluorescence detector (2475, Waters). The amino acids were identified and quantified by comparing with the retention times and peak areas of standards (WAT088122, Waters). During tryptophan analysis, the prepared sample was digested with 5% (w/v) NaOH for 24 h and neutralized to pH 7.0 with 6N HCl and tryptophan content measured spectrophotometrically at 530 nm (Sastry & Tammuru 1985).

Results and Discussions

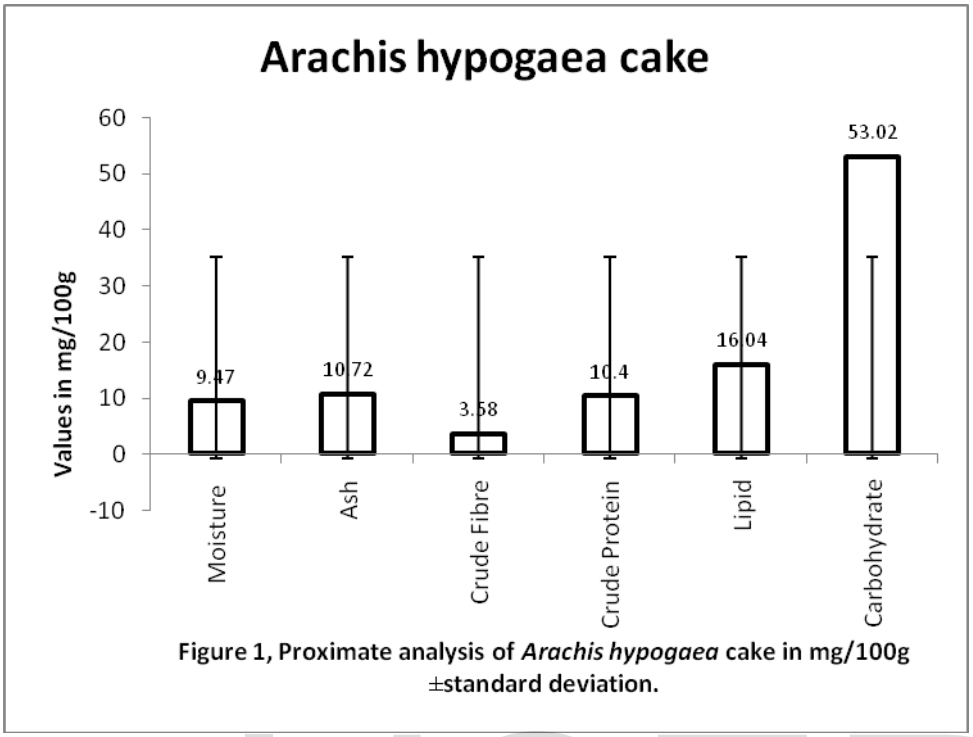

Figure 1, Proximate analysis of *Arachis hypogaea* cake in mg/100g ±standard deviation.

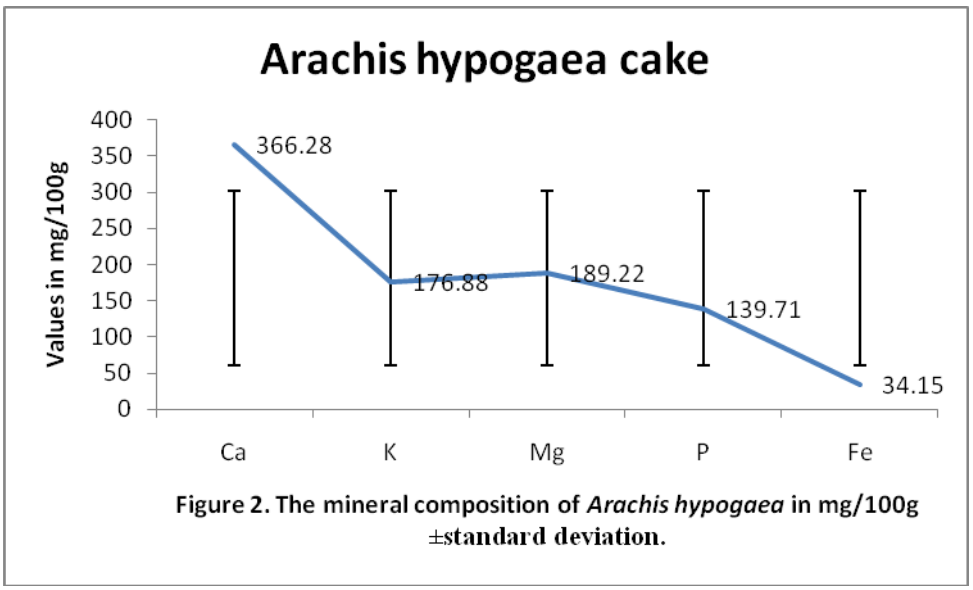

Figure 2. The mineral composition of *Arachis hypogaea* in mg/100g ±standard deviation.

TABLE 1

Amino Acids	*Arachis hypogaea.*	Fish meal IAFMM (1970)	FAO reference (1957)
Phenylalanine	4.30. ± 0.219[a]	3.91	4.55
Lysine	3.26 ±0.231[a]	7.77	6.85
Histidine	2.170±0.208[a]	2.45	1.76
Methionine	0.926±0.240[a]	2.86	3.58
Arginine	4.953±0.233[a]	5.84	4.58
Leucine	6.770±0.230[a]	7.50	4.20
Threonine	1.479±0.103[a]	4.26	4.55
Valine	2.880±0.230[a]	5.41	6.85
Tryptophan	0.433±0.233[a]	1.15	2.28
Alanine	2.056±0.145[a]	6.25	2.67
Glutamic acid	12.886±0.260[a]	12.77	17.56
Serine	2.356±0.202[a]	3.82	5.34

Aspartate	1.080±0.152[a]	9.10	8.79

Figure 1 presents the proximate composition of *Arachis hypogaea* from Cross River state. The moisture content of the plant was high with 9.47± 435, and fibre with 3.583 ±0.219. The concentration of ash was 10.72± 0.384 and protein was also high with 10.40 ± 0.435. The Lipid extract had the highest concentration value in the analysis with 16.040±0.264 and carbohydrate with 53.016±0.166. Protein and lipid contents are in agreement with the findings of Antasie *et al*., (2009). The authors reported high values of lipids and protein for *A. hypogaea.* This result is also in conformity with the result of Nelson and Carlos (1995). It is interesting to know that the concentration of Lipids/ fat plays a vital role in fish diet in promoting fat solubility and vitamin absorption and it is a high energy nutrient and more importantly it does not add to the bulk of the diet. The crude protein concentration in this study is in agreement with the findings of Umoh (1998). Meanwhile the crude protein values for *A. hypogaea* revealed that groundnut cake is rich in protein. This result is in agreement with the result recorded by Boyd, (1974) for Corn grit. The concentration of fibre was low and it is in conformity with the findings of Antasie *et al*., (2009).

The result of the present study shows the ability of groundnut in maintaining the internal expansion in other to allow the muscle especially that of the digestive system to relax and contract to enable the movement of food. Such role is physiologically played by fiber. It is difficult for man to digest crude fiber, meanwhile this fiber plays a useful and significant role in facilitating easy digestion and also not allowing the body to store carcinogen. High crude fiber content has some negative effects resulting to the limitation of its utilization especially when used in fish feed formulation. Diets low in crude fibre is undesirable as it could cause constipation and that such diets have been associated with diseases of colon like piles, appendicitis and cancer Antasie *et al.,* (2009). The knowledge of the concentration of ash

provides a measure of the total amount of minerals contained within an ingredient (Oyeyede 2005) and this also shows the level of mineral absorption facilitated by the root system of the plant. Boyd (1974) mentioned that the chemical compositions of many water plants at maturity stage are in varying concentrations in different locations. This fact was proven in this study as they were varying concentrations of the proximate indices in *Arachis hypogaea*. The carbohydrate level was high and it is in agreement with the findings of Osabor *et al.*, (2008).

Figure 2 represents the mineral element composition of *Arachis hypogaea*. The predominant elements in this study were calcium, potassium, sodium and phosphorus. The values of calcium recorded in the present study are much higher than the result of Atasie *et al*, (2009). The concentration of Na, Ma, P and Fe in this study was considerably higher than the findings of Atasie *et al.*, (2009). These finding was in conformity with the result of Achinewhu, (1983) and NRC (1978). The mineral composition of *A. hypogaea* ranged from 34.6 mg/100g for iron to 365.1 mg/100g for calcium. The plant had very high concentration of calcium, potassium, sodium and phosphorus. Magnesium and iron occurred in considerable amount. Calcium and Phosphorous are considered the most important major element. Calcium as an important micro element is responsible for the activation of several important enzymes and plays important role in the clotting of blood, bone formation and concentration of muscles. When deficient in fish diet result in deficiency signs such as growth retardation, poor appetite, poor bone formation and development and eventual death. Whereas when in high dose result in negative and stimulatory effects on the specific physiological processes. They result for potassium was expected because potassium is one of the most prominent mineral in Nigerian Agricultural products. In the present study, the values of potassium were less than what was recorded by Atasie *et al.*, (2009). The high presence of potassium is for enhancement of good growth. Calcium (Ca), Phosphorus (P)

and magnesium (Mg) are essential elements needed by fish for growth and other physiological functions of the body. They must be available in water and in the supplementary/artificial diets at optimum levels to enhance the well being of the fish. When deficient in fish diet result in deficiency signs such as growth retardation, poor appetite, poor bone formation and development and eventual death, whereas, when in high dose, negative and stimulatory effects on the specific physiological processes take place.

Table 1 is the amino acids composition of *Arachis hypogaea.* Thirteen amino acids were recorded comprising nine essential amino acids and four non-essential amino acids. Tryptophan, lysine, and valine, yielded approximately 50% of the required standard while methionine and threonine $^{1}/_{3}$ of the optimum requirement. Glutamic acid was very high in concentration and other amino acids were found in considerable amount. The amino acid profile obtained for *A. hypogaea* (Ground nut cake) revealed that Aspartic acid a non-essential amino acid was limiting. Lysine is extensively required for optimal growth and development and its deficiency usually cause immunodeficiency. Consequently, such diet has to be augmented if *A. hypogaea cake is to serves as a mono diet.* It is known that the quality of protein is determined by the quality of the component amino acid, the relative proportion of each of the amino acids and their availability as well as digestibility (Taylor and Robbin, 1968). This implies that the essential amino acids studied in this project cannot be synthesized by the animal therefore it should be provided and incorporated into the fish feed. Some of the amino acid are not required just for building blocks but for other metabolic functions.

Amino acids of *Arachis hypogaea* were compared with the amino acid profile of fish meal I.A.F.M.M (1970) and FAO (1957) recommended standard for plant protein. Dietary protein plays a role in provision of substrates needed for the synthesis of body proteins and other

metabolic important nitrogen-containing indispensable amino acids (AAs) in food proteins and is usually the main determinant of nutritional quality of protein (Young and Pellett 1984). Furthermore, amino acids are linked with health issues and amino acid deficiencies lead to a number of diseases. Hence, knowledge of the amino acid composition of foods is the starting points for establishing their potential nutritive value which may also allow evaluate changes in the nutritive value that may emanate in the cause of preparing, processing, and storing foods (Williams 2005). Apart from the limiting amino acids recorded in this study *A.hypogaea* has a good amino acid profile and it is a good source of plant protein.

Conclusion

The compositional analysis of *Arachis hypogaea* has revealed that this plant contained high level of lipids, protein, ash and carbohydrate with low level of moisture and fibre. The high protein level of the defatted groundnut makes it good as cake for consumption and also useful as an essential ingredient for feed formulation. When considering the shelf life of this plant, the low level of moisture is an advantage. The high ash content is indication of the concentration of minerals. Elemental abundance speeds up metabolic process and improves growth and development (Akanni, 2005). This work has enabled us to verify and draw a conclusion that *A. hypogaea* from Cross River State is a good source of protein with a quality amino acid structure. It contains major mineral elements which can be used as supplement in diets of fish and other animals. It is found to be a very important ingredient that can serve as an alternative to the very expensive ones when formulating fish feed. Also in the growing economy groundnut cake is readily available and cost effective thereby reducing the high cost of purchasing fish feed ingredient.

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