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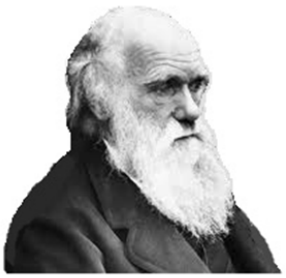

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Comparative study of the properties of yellow and brown *Cyperus esculentus* L.

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ABSTRACT

The present research accounts for the physicochemical and phytochemical characteristics of yellow and brown *Cyperus esculentus*, which were subjected to standard chemical and biochemical analysis. The results obtained from the analysis of yellow tiger nut showed the following phytochemical properties: anthocyanin 0.82 ± 0.02 µg/ml, oxalate 1.43 ± 0.05 µg/ml, tannin 12.22 ± 0.10 µg/ml, rutin 39.19 ± 0.29 µg/ml, phenol 10.94 ± 0.05 µg/ml, lunamarine 38.99 ± 0.07 µg/ml, saponin 44.67 ± 0.15 µg/ml, ribalinidine 1.35 ± 0.03 µg/ml, phytate 0.33 ± 0.01 µg/ml, catechin 48.29 ± 0.04 µg/ml, and kaempferol 38.59 ± 0.02 µg/ml. The brown tiger nut showed the presence of the phytochemicals with values as: anthocyanin 0.01 ± 0.00 µg/ml, oxalate 2.66 ± 0.02 µg/ml, tannin 12.67 ± 0.04 µg/ml, rutin 43.99 ± 0.05 µg/ml, phenol 11.02 ± 0.10 µg/ml, lunamarine 39.66 ± 0.03 µg/ml, saponin 47.79 ± .06 µg/ml, ribalinidine 1.21 ± 0.04 µg/ml, phytate 0.28 ± 0.03 µg/ml, catechin 46.77 ± 0.05 µg/ml, and kaempferol 38.34 ± 0.15 µg/ml. The observed level of mineral elements in the tiger nut is in the increasing order of zinc > sodium > iron > copper > calcium > magnesium > potassium for yellow tiger nut, and for brown tiger nut the order is zinc > sodium > iron > copper > calcium > magnesium > potassium. The values of physical and chemical properties recorded for yellow tiger nut is in the increasing order of moisture > ash content > crude protein > crude fiber > crude fat > carbohydrate, the increasing order for brown tiger nut shows ash content > moisture > crude protein > crude fiber > crude fat > carbohydrate. This indicates that tiger nuts contain elevated carbohydrate levels, crude fat and protein. From the data obtained, the high potassium to low sodium ratio of the two species of tiger nuts consequently might be imperative in diet recommendations for patients with high blood pressure (high BP) and edema as well. The investigated tiger nut varieties are rich sources of the phytochemicals, oil and contain moderate amounts of protein. They are also rich sources of fiber and carbohydrates. The phytochemical constituents of the tiger nuts are important and could be of high commercial significance

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in both, research institutes and pharmaceuticals companies for manufacturing of new drugs and for therapeutic applications.

***Keywords*:** Mineral content, nutritive value, phytochemicals, medicinal value, yellow and brown tiger nuts

1. INTRODUCTION

Cyperus esculentus (tiger nut), popularly known as "akiausa" in Igbo language, can also be called yellow nut sedge, earth almond or tiger nut sedge. It is a crop of the sedge family which is found in most parts of the world [1] (Mishra et al. 2016). This plant is instinctive in most of the Western Hemisphere as well as Middle East, Southern Europe, Africa, and the Indian Subcontinent. It has turned out to be naturalized in many other places, including Ukraine, China, New South Wales, Hawaii, New Guinea, Java, and various oceanic Islands [2] (Bamgbose *et al*., 2003). *Cyperus esculentus* can be found in the wild, as a weed. There is indication for its cultivation in Egypt since the sixth millennium BC, and for several centuries in Southern Europe. Tiger nut is broadly used for animal (feed) and human consumption. In Spain, these nuts are mainly used to make a milk-like beverage called horchata de chufa [2] (Bamgbose *et al*., 2003). This beverage (tiger nut milk or orgeat in English-speaking countries) is a nonalcoholic stimulating drink of dairy form and is usually taken in summertime [3] (Adejuyitan, 2011). It has been reported that grainy sandy soil and mild temperatures are special for the cultivation and growth of the nut [4] (Belewu and Belewu, 2007). When hydrated, it has slightly harder nut texture, but with a rather more deep and concentrated taste. The cultivation time is within April and November. Being cultivated through protraction irrigation, tiger nut has to be properly dried before storage [5] (Temple *et al*., 1990).

The dehydrating procedure ensures longer shelf life, preventing rot or any other bacterial infection securing their quality and nutritional level [6] (Sanchez-Zapata *et al*., 2012). This process makes tiger nut skin to be wrinkled, a factor which prevents its acceptance by some people. They are quite hard and are generally soaked in water before they can be eaten, thus making them much softer and giving them a better taste [7] (Oladele *et al*., 2011). It is known with different names in Nigeria according to different languages as "Ofio" in Yoruba, "Akiausa" in Igbo, and "Aya" in Hausa where the different varieties are grown [8] (Nwaoguikpe, 2008). Among these, the yellow variety is preferable over other species because of its essential properties such as the large size, attractive yellow color and fleshiness in nature [9] (Ejoh *et al.*, 2006). It has high milk yield, contains lower fat and high protein levels and less anti-nutritional factors, especially polyphenol. Recently, there is awareness in the increased usage and consumption of tiger nut [2, 9] (Bamgbose *et al*., 2003; Ejoh *et al*., 2006).

Cyperus esculentus is part of the *Cyperaceae* family. The elevated level of sucrose and starch as well as the high arginine content that stimulates insulin production makes tiger nut good for consumption by diabetic patients [4] (Belewu and Belewu, 2007). It can also be cooked, dried, prepared in powdered form, and may be used in confectionary to make biscuits with a delicious nut-like flavour. Mixing the ground nuts with vanilla, sugar, cream and the cinnamon, makes it a refreshing beverage. The roasted nuts are substitute for coffee [10] (Bamishaiye and Bamishaiye, 2011). *Cyperus esculentus* have long been documented for their beneficial importance as they are rich in fiber, protein and natural sugars, minerals (phosphorus,

potassium), including high levels of vitamins C and E [4, 9] (Belewu and Belewu, 2007; Ejoh *et al.*, 2006).

The aim of this research work is to determine the physicochemical and phytochemical characteristics of yellow and brown tiger nuts. The study work compared the physicochemical and phytochemical characteristics of the investigated tiger nuts. The need for this study lies in the importance of tiger nuts as it has many uses; the nuts are edible, with a slightly sweet, nutty flavour, compared to the more bitter-tasting nut of the related *Cyperus rotundus* (purple nut sedge). The tiger nut milk compared to other soft drinks is not just a refreshing drink, also a very healthy source of nutrients [11] (Ekeke and Shode, 1990). Its content of vitamin E also works together with cholesterol because it has antioxidant effect over fats, which is ideal for coronary heart disease [12] (Yemm and Cocking, 1994).

Cyperus esculentus has been reported to help in the prevention of thrombosis, heart related diseases, as well as assist in circulation of blood, which could help in the prevention and treatment of bacterial and urinary tract infections, which reduces the risk of colon cancer [13] (Chukwuma *et al.*, 2010). The rich mineral content of tiger nut milk makes its intake good for the supply the essential nutrients needed for body growth and development. Its calorific value (100 cal/100 g) makes it a very good energy drink. A very significant point is that it does not contain lactose or gluten; therefore it is very good in reproduction and in menstruation [14] (Belewu and Abodunrin, 2006). The importance of tiger nut in terms of its nutrient content therefore calls for the analysis of the different varieties of the nut to ascertain their phytochemical and physicochemical composition.

2. MATERIALS AND METHOD

Materials and reagents used for this analysis include the tiger nuts shown in **Figure 1a** and **b**, clean water, electric weighing balance, ethanol, Soxhlet extractor, Whatman filter paper, 5% $FeCl_3$, NH_3, Na_2CO_3, volumetric flask, HCl, tetraoxosulphate (vi) acid (H_2SO_4), n-hexane, Fehling's solutions A and B, chloroform, picric acid, acetic anhydride, aqueous Ferric chloride solution distilled water.

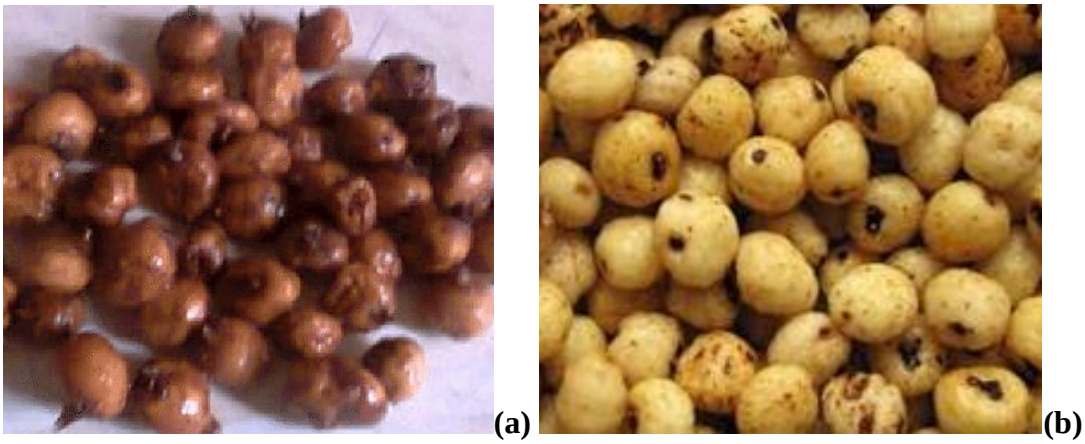

Figure 1. The pictures of: **(a)** brown tiger nut, and **(b)** yellow tiger nut

2. 1. Source and Preparation of the sample

Two different samples of *Cyperus esculentus* seed (tiger nuts) were bought from a market in Owerri, Imo State. The tiger nuts were identified and authenticated at the Department of Crop science and Biotechnology, Imo State University, Owerri, and then taken to the laboratory for standard analysis. The *Cyperus esculentus* species (yellow tiger nut and brown tiger nut) were thoroughly washed with clean water separately accordingly, based on how they were collected and sun dried. They were then grounded using electric blender (Kenwood BL450 series) and were stored in a well labeled air-tight container for analysis [15] (Ihenetu *et al.* 2019).

2. 2. Method of Extraction

250 g of the tiger nut sample were pulverized in a blender, followed by four consecutive extraction of the oil using n-hexane (1:3 w/v) of high purity at room temperature. Separation of the miscella was accomplished by the help of a Whatman No. 1 filter paper. Then, n-haxane was removed from the miscella under vacuum, at 50 °C. The obtained oil was dried over anhydrous sodium sulphate and was then analyzed, as described by Duru *et al*. [16] (2014).

2. 3. Extraction of phytochemicals

1 g of sample was weighed with Nanbei NBT-A200 and transferred into a test tube to which 15 ml ethanol and 10 ml potassium hydroxide (50% m/v) and introduced into it. The content of the test tube was allowed to stand on a water bath for 60 minutes at 60 °C. At completion of reaction, the products were separated with a separating funnel. The tube was washed successfully with 20 ml of ethanol, 10 ml of cold water, 10 ml of hot water and 3 ml of hexane, which was all transferred to the funnel. This extract were combined and washed with 10 ml of 10% v/v ethanol aqueous solution three times. The solution was dried with anhydrous sodium sulfate and the solvent was evaporated. 1000 µl of pyridine was used to solubilize the sample of which 200 µl was taken for analysis. The extraction and analysis for the phytochemicals were according to the methods described by previous publications [17-19] (Duru *et al*., 2015; Ikpa *et al*., 2016; Ibe *et al*., 2019a)

2. 4. Physical parameters

Refractive index of extracted oils was calculated according to AOAC 1990 using Abbe refractometer (Model 1230 - Percent Brix, Gingerbread Company, USA) at 25 °C [20]. The method, according to Lee *et al*., [21] (2004), was applied in colour determination, in which the absorbance at 420 nm of chloroform (5% w/v) solutions of the oil was determined using a spectrophotometer (Spectronic 20D, Newer Milton Roy, USA) [22] (Adel *et al*., 2015). Calcium, sodium, potassium, magnesium, iron, zinc, and copper were determined using Atomic Absorption Spectrophotometer (AAnalyst 400, Perkins Elmer, Houston), according to Ibe *et al*., (2017) [23].

2. 5. Chemical content

Standard methods of fats and oil analysis were adopted for the determination of saponification value, peroxide value, iodine value, acid value and unsaponifiable matter, as described in a previous publication [24] (IUPAC 1981). The value obtained by subtraction of

acid value from saponification value gave the ester value. All determinations were conducted in triplicate to ensure precision and accuracy of the results.

3. RESULTS AND DISCUSSION

Result of the phytochemical screening of the tiger nuts is presented in **Table 1**. The result revealed the presence of some phytochemicals which are considered as important medicinal chemical constituents. Anthocyanin, oxalate, tannin, rutin, phenol, lunamarine, saponin, ribalinidine, phytate, catechin, and kaempferol were present at varying concentrations, as shown in Table 1. The result of the phytochemical examination of the samples revealed that the two samples are rich in rutin, tannin, phenol, lunamarine, saponin, catechin, and kaempferol.

Table 1. Result of phytochemical screening of tiger nut yellow and tiger nut brown.

Component	Tiger nut yellow (µg/ml)	Tiger nut brown (µg/ml)
anthocyanin	0.82 ± 0.02	0.01 ± 0.00
oxalate	1.43 ± 0.05	2.66 ± 0.02
tannin	12.22 ± 0.10	12.67 ± 0.04
rutin	39.19 ± 0.29	43.99 ± 0.05
phenol	10.94 ± 0.05	11.02 ± 0.10
lunamarine	38.99 ± 0.07	39.66 ± 0.03
saponin	44.67 ± 0.15	47.79 ± 0.06
ribalinidine	1.35 ± 0.03	1.21 ± 0.04
Phytate	0.33 ± 0.01	0.28 ± 0.03
Catechin	48.29 ± 0.04	46.77 ± 0.05
Kaempferol	38.59 ± 0.02	38.34 ± 0.15
total	236.82 ± 0.05	277.43 ± 0.04

n = 3

The results obtained from Table 1, show that the yellow tiger nut is rich in anthocyanin, phytate, catechin and kaempferol while tiger nut brown has higher values of other phytochemical parameters that were assessed. Brown tiger nut gave the highest content of phytochemicals, with a total of (277.43 ± 0.04 µg/ml) as against the yellow tiger nut (236.82 ± 0.05 µg/ml). From the results obtained in Table 1, catechin gave the highest concentration

(48.29 ± 0.04 µg/ml), while phytate gave the least concentration (0.33 ± 0.01 µg/ml). Phytochemical analysis of plants is important as the constituents of such plants could be useful in the discovery and manufacture of new drugs [16] (Duru *et al.*, 2014). It was observed that saponin had the highest concentration (47.79 ± 0.06 µg/ml) in brown tiger nut, while the least concentration (0.01 ± 0.00 µg/ml) recorded was in anthocyanin.

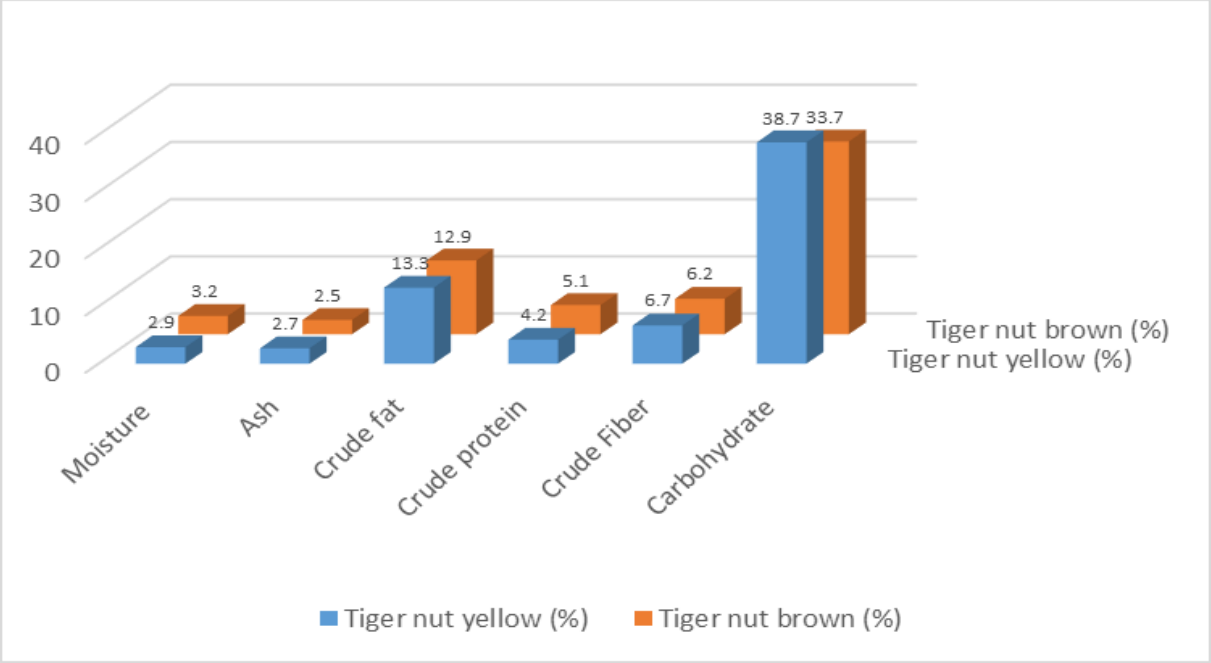

Figure 2. Chemical composition of the tiger nuts

The inference made from the physicochemical composition of the different varieties of tiger nuts displayed in **Figure 2** shows the different components in an increasing order for yellow tiger nut as moisture > ash content > crude protein > crude fiber > crude fat > carbohydrate; the increasing order for brown tiger nut indicates that ash content > moisture > crude protein > crude fiber > crude fat > carbohydrate. This suggests that tiger nuts contain high levels of carbohydrate, crude fat and protein. This is in agreement with the result obtained by Umuerie *et al.*, (1997) [25]. The present study indicates that the crude fat content of the two species of tiger nuts is higher than that of other starchy fruits seeds (Ade-Omowaye *et al.*, 2008) [26]. Tiger nuts can serve as a spring of plant protein in its bio-availability form. Tiger nuts may perhaps supply up to 18% of protein to adult's regular protein need and invariably more than 24% to a children's regular protein and carbohydrate requirements. The two species of tiger nut's ash value was comparable to values reported by Suleiman *et al.* (2008) [27]. However, values reported by some researchers are significantly higher than the values reported in the present study (Ejoh *et al.*, 2006) [9].

The mineral content of the two tiger nut samples is presented in **Table 2**. The study recorded calcium concentration of 59.36±0.19 mg/100 g in yellow tiger nut, and 45.69±1.12 mg/100 g in brown tiger nut. Calcium could enhance resistance of tissues that makes it possible for stem of plants to be upright. Calcium is vital in the development and proper functioning of

the bone and teeth (Ibe *et al.*, 2019a) [19]. The observed level of sodium in yellow tiger nut is 26.41±1.12 mg/100 g, while brown tiger nut showed 22.89±0.096 mg/100 g. It was observed that elevated potassium concentration of 200.11±2.03 and 212.32±1.96 mg/100 g, respectively for yellow tiger nut and brown tiger nut were recorded in the study. Magnesium levels were 114.74±0.65 mg/100 g for yellow tiger nut and 116.39±0.96 mg/100 g brown tiger nut. Iron concentrations of 26.58±0.78 and 27.87±0.87 mg/100 g were recorded, respectively for yellow tiger nut and brown tiger nut in this study.

Table 2. Mineral composition of the Tiger nut yellow (T.Y) and brown (T.B).

Phytochemicals	**Yellow tiger nut (mg/ 100 g)**	**Brown tiger nut (mg/ 100 g)**
Calcium	59.36 ± 0.19	45.69 ± 1.12
Sodium	26.41 ± 1.12	22.89 ± 0.96
Potassium	200.11 ± 2.03	212.32 ± 1.96
Magnesium	114.74 ± 0.65	116.39 ± 0.96
Iron	26.58 ± 0.78	27.87 ± 0.87
Zinc	6.22 ± 1.33	3.65 ± 0.26
Copper	41.35 ± 2.31	36.56 ± 1.58

The zinc levels were 6.22±1.33 and 3.65±0.26 mg/100 g for yellow tiger nut and brown tiger nut, respectively. It was also observed that copper values were 41.35±35 mg/100 g yellow tiger nut and 36.56±1.58 mg/100 g brown tiger nut. The mineral elements are in the increasing order of zinc > sodium > iron > copper > calcium > magnesium > potassium for yellow tiger nut, and for brown tiger nut the order is zinc > sodium > iron > copper > calcium > magnesium > potassium. Tiger nuts may be eaten as snacks by young, old, pregnant and lactating mothers because of their high nutrients content. These nutrients could expressively help the body in most metabolic processes as well as refreshing the body. Consumption of tiger nut may be very good for growing children due to its nutrient content. It can be converted to flour, can be used to produce tiger nut juice, and can be mixed with other high nutritious seed plants for protein and energy drinks. High concentration of calcium, potassium and magnesium were recorded in the samples. Magnesium also offers bone strength, nerve, aids enzyme and heart functions (Ibe *et al.*, 2019a) [19]. The high magnesium and zinc content of tiger nut indicates that it could probably supply the needed magnesium and zinc for children and adults. From the data obtained, the high potassium to low sodium ratio of the two species of tiger nuts consequently, might be probably imperative in diet recommendations for patients with high blood pressure (high BP) and edema as well. Tiger nuts contain shielding nutrients because they can probably supply adequate zinc, copper and iron. Zinc is a significant part of many hormones and more than 100 different enzymes. Zinc plays major role in several metabolic reactions and might

probably play a vital role in alcohol metabolism, immunity, reproduction and sexual development. Zn has been known for antioxidant activities as well as protection against rapid aging in humans (Ibe *et al.*, 2020) [28]. Cu is considered as one of the essential element needed in trace amount, it takes part in some important biological processes such as in the hematological, nervous, cardiovascular, immune and reproduction systems (Cerone *et al.*, 2000) [29]. Also Cu plays a significant role in proper functioning of some proteins (Hefnawy and Elkhaiat, 2015) [30]. It is known to be present in many enzymes, helps in the metabolism of iron, as well as facilitates the transmission of electrical signals in human body (Ashish *et al.*, 2013) [31]. It should be noted that elevated concentration of Cu could be very toxic to humans (Saravu *et al.*, 2007; Ibe *et al.*, 2018) [32, 33].

Iron is an important element that is required for the developmental processes of most living organisms. It also plays an essential role in the metabolic processes of living organisms (Ibe *et al.*, 2019a; Valko *et al.*, 2005) [19, 34]. The observed iron levels in the samples is an indication that anemia may be prevented with the help of iron present in tiger nuts. Iron is the functional constituent of hemoglobin as well as other key compounds used in respiration, immune function, and cognitive development, especially for children and adolescence (Ibe *et al.*, 2019b) [35]. Elevated levels of iron in human blood could be toxic with the tendency of cancer risk (Sane *et al.*, 2018) [36]. The observed levels of mineral elements in the analyzed tiger nuts could be attributed to so many factors such as the atmospheric deposition (Opara *et al.*, 2016; Ibe *et al.*, 2016; Ibe *et al.*, 2017) [23, 37, 38] improper disposal of metallic and electronic wastes in the environment (Ibe *et al.*, 2018; Ibe and Ibeachu, 2020) [33, 39], roof and road offs (Ibe and Ibe, 2017) [40], as well as nature of the soil on which the crop was planted (Rahman *et al.*, 2012; Enyoh and Isiuku, 2020) [42, 41].

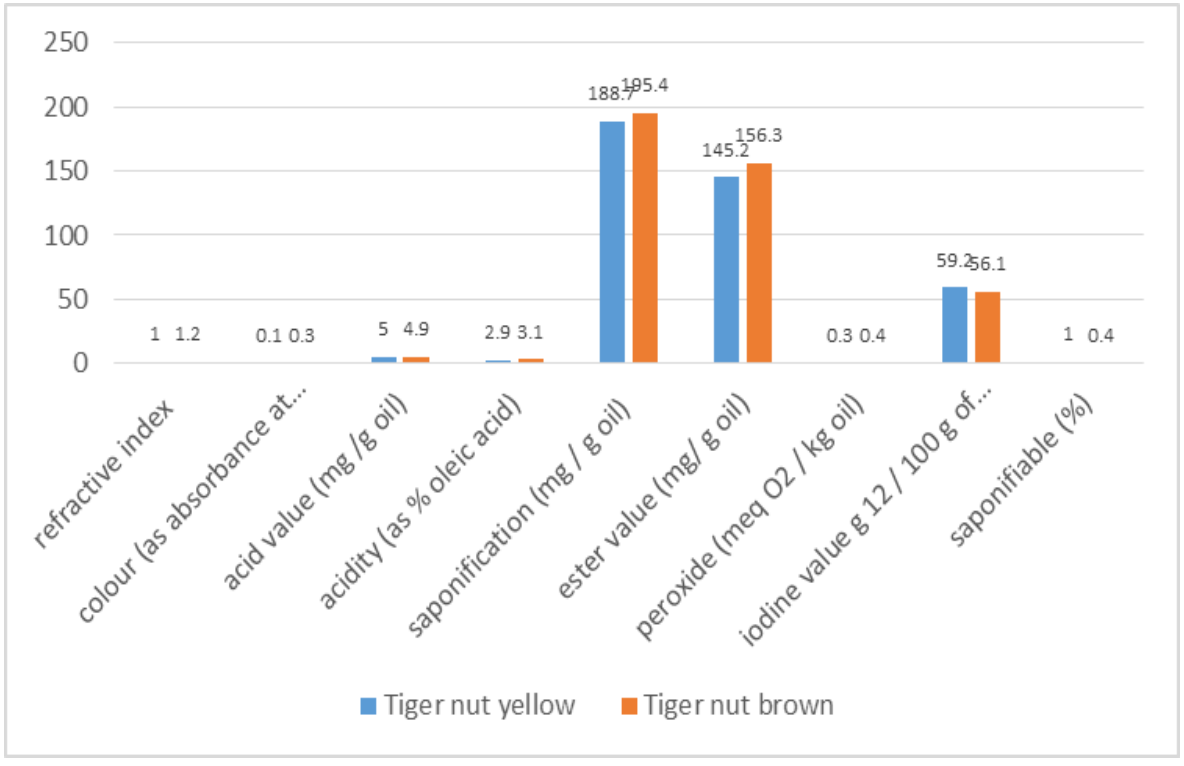

Figure 3. The properties of tiger nut oils

The properties of the extracted tiger nut oils are presented in **Figure 3**. The refractive index usually presents the structural properties which encompass the average molecular mass and degree of unsaturation of the fatty acids in fats and oils. The characteristics of the extracted oil from tiger nut are comparable to values of most conventional seed oils, such as soya been and cotton seed oils, peanut and olive oils (Alemayhu *et al*., 2019; Gunstone *et al*., 1986; Ejoh *et al*., 2006) [9. 43, 44]. Therefore, tiger nut oils could be produced in commercial quantities and used for the different purposes, most conventional oils are put to such as for cream and soap production as well as for nutrition.

This implies that tiger nut oil could be used in place of most conventional oils (Omode *et al*., 1995) [45]. The different refractive index values for the ground yellow tiger nut and brown tiger nut, given in Figure 2, would tend to suggest that the two samples showed similar level of fatty acid (FA) chain length and extent of unsaturation. Similarly, the related saponification values of 188.1 and 195.4 for the ground tiger nut seeds is an indication of comparable average chain lengths for the FAs in all the samples. Undeniably, the iodine values for the two ground tiger nut seeds 59.2 and 56.1 are in agreement with the gas chromatographic estimation of the extent of unsaturation in seed oils [1]. Other parameters checked, as seen in Figure 2, are quite judicious as they compare very well with Codex recommended values for virgin olive oil (CODEX Standard, 2013) [46].

4. CONCLUSIONS

The research work which was carried out on yellow and brown tiger nuts revealed the phytochemical constituents in the samples. The phytochemicals that were recorded in the samples include anthocyanin, oxalate, tannin, rutin, phenol, lunamarine, saponin, ribalinidine, phytate, catechin, and kaempferol. Some level of differences were noticed during studies concerning the phytochemical constituents of the samples.

This difference in phytochemical properties might be due to a change in location and genetic variation as a result of cross pollination. The genetic makeup due to cross pollination of the two varieties of the tiger nuts investigated may have caused variation in the results of their phytochemical screening as well as other properties of the sampled tiger nut. Phytochemical analysis of plant materials is important as the constituents of such could be useful in the discovery and manufacture of new drugs. Tiger nuts are rich source of fibre, carbohydrate, oil, and moderate level of protein. The observed levels of mineral elements are an indication that tiger nut could be a source trace of minerals needed in the body for proper functioning. The edible and stable oil obtained from the nut is believed to be superior oil that compares favorably with olive oil.

The result of the study justifies the use of tiger nut oil in different food products. Thus tiger nuts cultivation should be developed into commercial quantity for its use in food and industrial products.

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References

[1] Mishra S., Tripathi A., Tripathi D.K. and. Chauhan D.K. (2016) Role of sedges (Cyperaceae) in wetlands, environmental cleaning and as food material: Possibilities and future perspectives (chapter 18); In Plant-Environment Interaction: Responses and Approaches to Mitigate Stress, 1st Ed. M. M. Azooz and P. Ahmad. (Ed.), John Wiley & Sons, Ltd USA, pp 327 – 338.

[2] Bamgbose A. M., Eruvbetine D., and Dada W. (2003). Utilization of tigernut (Cyperus esculentus) in the diets from cockerel starters. *Bioresour. Technol*. 89, 245-248

[3] Adejuyitan J.A. (2011). Tiger nut processing: its food uses and health benefits. *American Journal of Food Technology,* 6(3), 197-201

[4] Belewu M.A. and Belewu K.Y. (2007). Comparative physiochemical evaluation of tigernut, soybean and coconut milk sources. *Int. J. Agric. Biol*. 5, 785-787

[5] Temple V.J., Ojobe T.O. and Kapu M.M. (1998). Chemical analysis of tigernut (*Cyperus esculentus*). *J. Sci. Agric.* 50, 261-263

[6] Sanchez-Zapata, E., Fernandez-Lopez, J. and Angelperez-Alvarez J. (2012). Tiger nut (Cyperus esculentus) commercialization, health aspects, composition, properties and food applications. *CRFSFS*, 11, 366-377

[7] Oladele K. A., Osundahunsi F. O. and Adebowale A. Y. (2009) Influence of Processing Techniques on the Nutrients and anti-nutrients of Tigernut (Cyperus esculentus L.). *World J. Dairy and Food Sci*. 2: 88-93

[8] Nwaoguikpe R. N. (2008). Effect of the extracts of African Yam Beans (Sphenostylis stenocarpa), Hyacinth beans (Lablab purpureus) and Bambara groundnut (Voandzeia subterrenea) on hemoglobin polymerization and the Fe^{2+}/ Fe^{3+} ratio of sickle cell blood. *Niger*. J. *Biochem. Mol. Biol*. 23(2), 46-50

[9] Ejoh R.A., Djomdi and Ndjouenkeu R. (2006). Characteristics of tiger nut (Cyperus esculentus) tubers and their performance in the production of a milky drink. *Journal of Food Processing and Preservatio*n, 30, 145-163

[10] Bamishaiye E.I. and Bamishaiye O.M. (2011). Tiger Nut: As a plant, its derivatives and benefits. *African Journal of Agriculture, Food, Nutrition and Development,* 11(5), 5157-5170

[11] Ekeke G.I. and Shode F.O. (1990). Phenylalanine is the predominant antisickling agent in Cajanus cajan seed extract. *Planta Medica*, 56, 41-43

[12] Yemm, E.W. and Cocking, E.C. (1994). The determination of amino acids with Ninhydrin. *Journal of American Analyst*, 80, 209-213

[13] Chukwuma E.R., Obiama N., and Christopher O.I. (2010). The phytochemical composition and some Biochemical effect of Nigerian Tiger nut (Cyperus esculentus. L) tuber. *Pakistan Journal of Nutrition*, 9(7), 709-715

[14] [14] Belewu M.A. and Abodurin A.O. (2006). Preparation of Kuunu from unexploited rich food source-tigernut (Cyperus esculentus). *Pakistan Journal of Nutrition* 7, 109-111

[15] Ihenetu S.C., Enyoh C.E. Inyamah P.C., and Enyoh E.C. (2019). Physicochemical properties, phytochemicals and fat soluble vitamins of seed oil extracts from Sesamum indicum L. *International Journal of Chemical and Biological Sciences*, 1 (4), 8-12

[16] Duru C.E., Duru I.A., Ikpa C.B.C., and Ibe F.C. (2014). Chemical and spectra studies of the alleged killer seed of Berlinia grandiflora, *IOSR - Journal of Applied Chemistry*, 7(1) Ver 11, 14-14

[17] Duru C.E., Duru I.A., Ibe F.C., Achinihu I.O., and Ukiwe L. (2015). Functional group analysis and antibacterial studies of column chromatography eluates from the fruit of Garcinia kola. *IOSR Journal of Applied Chemistry*, 8(9), Ver I, 35-38

[18] Ikpa C.B.C., Ibe F.C., and Ikpa C. (2016). Isolation, chemical composition, characterization and anti-bacterial activity of acridine diglycoside from Moringa Olifera. *International Journal of Pharmacology, Phytochemistry and Ethnomedicine*, 2, 30-36

[19] Ibe F.C., Ibe B.O., and Enyoh C.E. (2019a) Trace metal, FTIR and phytochemical analysis of Viscum album leaves harvested from Pentaclethra macrophylla. *World News of Natural Sciences* 25, 61-71

[20] Association of Official Analytical Chemists (AOAC) (1990). Official methods of analysis.Association of Official Chemists, 15th Edn. Washington, DC. pp. 10-30.

[21] Lee Y.C., Oh S.W., Chang J., and Kim I.H. (2004). Chemical composition and oxidative stability of safflower oil prepared from safflower seed roasted with different temperatures. *Food Chemistry*, 84, 1-6

[22] Adel A.A.M., Awad A.M., Mohamed H.H., and Iryna S. (2015). Chemical composition, physicochemical properties and fatty acid profile of Tiger Nut (Cyperus esculentus L) seed oil as affected by different preparation methods. *International Food Research Journal*, 22(5), 1931-1938

[23] Ibe F.C., Isiuku B.O., Enyoh C.E. (2017) Trace metals analysis of soil and edible plant leaves from abandoned municipal waste dumpsite in Owerri, Imo State, Nigeria. *World News of Natural Sciences*, 13, 27-42

[24] IUPAC (1981) International Union of Pure and Applied Chemistry Applied Chemistry Division Commission on Oils, Fats and Derivatives Standard Methods for the Analysis of Oils, Fats and derivatives 6th Edition 1st Supplement: Part 3 SECTION II: OILS AND FATS Prepared for publication by C. PAQUOT Pure & Appi. Chem., Vol. 53, Pergamon Press Ltd Britain, pp. 783-794.

[25] Umerie S.C., Okafor E.P. and Uka A.S. (1997) *Bioresource Technology*, 6, 171

[26] Ade-Omowaye B.I.O, Akinwande B.A., Bolarinwa I.F., and Adebiyi A.O. (2008) Evaluation of tigernut (Cyperus esculentus) - wheat composite flour and bread. *Afr. J. Food Sci*. 2, 87-91

[27] Mohammed S., Suleiman J., Eniola O., Jamila A., Omale O.C., Abbah D., and Ocholi E. (2017). Proximate composition, mineral and some vitamin contents of tiger nut (Cyperus esculentus). *Journal of Clinical Investigation*, 8(4), 161-165

[28] Ibe, F.C., Enyoh, C.E., Opara, A.I. *et al.* Evaluation of pollution status of groundwater resources of parts of Owerri metropolis and environs, Southeastern Nigeria, using health risk and contamination models. *Int J Energ Water Res* 4, 357–374 (2020). https://doi.org/10.1007/s42108-020-00071-8

[29] Cerone S. I., Sansinanea A.S., Streitenberger S.A., Garcia M.C., and Auza N.J., (2000). Cytochrome oxidase, Cu, Zn-superoxide dismutase, and ceruloplasmin activities in copper-deficient bovines. *Biological Trace Element Research*, 73, 269-278

[30] Hefnawy A.E. and Elkhaiat H.M. (2015). The importance of copper and the effects of its deficiency and toxicity in animal health. *International Journal of Livestock Research*, 5 (12), 1-20.

[31] Ashish B., Neeti K., and Himanshu K. (2013). Review Paper Copper Toxicity: A Comprehensive Study. *Research Journal of Recent Sciences*, 2, 58-67

[32] Saravu K., Jose J., Bhat M.N., Jimmy B., Shastry B.A. (2007) Acute ingestion of copper sulphate: A review on its clinical manifestations and management. *Indian J. Crit. Care Med.* 11, 74-80

[33] Ibe F.C., Opara A.I., Ibe B.O., Adindu B.C., Ichu B. C. (2018) Environmental and Health implications of trace metal concentrations in street dust around some electronic repair workshops in Owerri, Southeastern Nigeria. *Environ. Monit. Assess*. 190 (696), 1-14

[34] Valko M., Morris H., Cronin M.T.D. (2005) Metals, toxicity, and oxidative stress. *Curr. Med. Chem*. 12(10), 1161-1208

[35] Ibe F.C., Ibe B.O., Nzenwa P.O., and Enedoh M.C. (2019b). Phytochemical, FTIR and Elemental Studies of African Mistlotoe (*Viscum album*) Leaves on *Cola nitida* from South-Eastern Nigeria. *World Scientific News*, 132, 84-97

[36] Sane M.R., Malukani K., Kulkarni R., Varun A. (2018). Fatal iron toxicity in an adult: Clinical Profile and Review. *Indian J. Crit. Care Med.* 22(11), 801-803

[37] Opara A.I., Ibe F.C., Njoku P.C., Alinnor J.I., Enenebeaku C.K. (2016). Geospatial and geostatistical analyses of particulate matter (PM_{10}) concentrations in Imo State, Nigeria, *International Letters of Natural Sciences* 57, 89-107

[38] Ibe F.C., Njoku P.C., Alinnor J.I., and Opara A.I. (2016). Evaluation of ambient air quality in parts of Imo state, Nigeria, Research. *Journal of Chemical Sciences*, 6 (1), 41-52

[39] Ibe F.C. and Ibeachu B.E. (2020) Metallic contaminant levels of borehole water sources within metal scrap dumpsites in Aboh Mbaise, Imo State, Nigeria. *World Scientific News*, 144, 226-242

[40] Ibe F.C. and Ibe B.O. (2016). Roof runoff water as source of pollution: a case study of some selected roofs in Orlu metropolis, Imo State, Nigeria. *International Letters of Natural Sciences*, 50, 53-61

[41] Rahman S.H., Khanam D., Adyel T.M., Islam M.S., Ahsan M.A., and Akbor M.A. (2012) Assessment of heavy metal contamination of agricultural soil around Dhaka

Export Processing Zone (DEPZ), Bangladesh: Implication of seasonal variation and indices. *Appl. Sci*. 2, 584-601

[42] Christian Ebere Enyoh & Beniah Obinna Isiuku (2020) Characterisation of some soils from flood basin in Amakohia, Owerri, Nigeria. *International Journal of Environmental Analytical Chemistry*, DOI: 10.1080/03067319.2020.1773455

[43] Alemayhu A., Admassu S., and Tesfaye B. (2019) Shelf-life prediction of edible cotton, peanut and soybean seed oils using an empirical model based on standard quality tests. *Cogent Food and Agriculture*, 5(1622482) 1018. https://doi.org/10.1080/23311932.2019.1622482

[44] Gunstone F.D., Harwood J.L., and Padley F.B. (1986). The Lipid Handbook. Chapman & Hall, New York, NY.

[45] Omode A.A., Fatoki O.S., and Olaogun K.A. (1995). Physicochemical properties of some underexploited and nonconventional oilseeds. *J. Agr. Food Chem*. 43, 2850-2853

[46] CODEX Standard 33-1981, Standard for Olive Oils and Olive Pomace Oils. Codex Alimentarius. International food Standard (2013) pp. 1-9