



Effect of Storage on Mycology, Functional properties and Sensory attributes of Tiger nut Flour

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Abstract: Tiger nut (*Cyperus esculentus* L.) samples were collected from two different markets (*Ojo* and *Bodija*) within Ibadan metropolis. The nut were processed into flours, stored using two different storage method (nylon and plastic) for a period of four weeks, and the following parameters were monitored fungi constituent, proximate composition, functional properties, mineral constituent, aflatoxin constituent and the sensory evaluation during processing into snack using different ratio of wheat flour. The predominate fungi isolated after four weeks of storage were; *Aspergillus niger*, *A. fumigatus*, *A. flavus* and *Penicillin citrinum*. The aflatoxin level in the stored samples ranges from 4.3 – 7.2 ng/g, proximate composition values (%) for *Bodija* and *Ojo* samples were: Moisture content (8.20 and 8.07), crude protein (14.05 and 13.90) and carbohydrate (23.97 and 30.14). After four weeks of storage, the functional properties of *Ojo* tiger nut flour stored in nylon (OTN) had the highest water absorption capacity of 2.98% while the lowest was the *Bodija* tiger nut flour stored in nylon (BTN) at 1.97%, OTN had the highest oil absorption capacity at 2.57% while *Bodija* sample stored in plastic (BTP) had the lowest capacity at 1.99%. The swelling capacity (SC) of BTP was the highest at 7.35%, OTN and BTN had the lowest SC at 7.20%. There was no significant difference within the functional properties of the tiger nut flours and wheat flour. The minerals assessed show that Mg had the highest value then followed by Na, Fe, Zn and Cu respectively. Among the formulated blends, sensory evaluation shows that substitution of wheat flour with 60% tiger nut and 40% wheat flour was the most acceptable blend for puff-puff snack produced.

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Key words: Tiger nut; functional property; Puff-puff; Aflatoxin; Proximate

1. Introduction

Lifestyle diseases such as cardiovascular diseases, cancer, osteoporosis and respiratory diseases account for 59% of the 56.5 million deaths annually and 45.9% of the global burden of disease (WHO, 1998). This brought about the rise in the study of effect of diet on health and the influence of specific food components on health, hence the emergence of designer foods. Designer foods are processed foods, supplemented with food ingredients that are rich in natural disease preventing substances (Manjula and Suneetha, 2011). Designing of food is based on technical perspective such as product formulation, novel processing and modification of raw materials (Manjula and Suneetha, 2011). Development of product formulation with functional foods which may be defined as food with bio-active compounds that confer health benefits, beyond their traditional nutrients regardless of any or no modification, may enhance product. Functional foods can improve health and well-being, beyond the provision of basic

nutritional requirements (Chukwuma *et al.*, 2010). Tiger nut (*Cyperus esculentus*) is one among so many functional foods that may be used in food designing.

Tiger nut is known majorly for its milk which is a very nutritive energetic drink and said to contain aphrodisiac properties, also promotes urine production and menstruation (Adejuyitan, 2011). In addition, tiger nut has been demonstrated to contain higher essential amino acids than those proposed in the protein standard by the FAO/WHO (1985). The tiger nut oil also has high monounsaturated fatty acids, similar as those of olive, avocado and hazelnut oil (Ezeh *et al.*, 2014). These monounsaturated oils contain high unsaponifiable matter, phospholipids and other bioactive compounds such as tocopherols, phytosterols and polyphenol (Sanchez- Zapata *et al.*, 2012). The nutritional profile and unique functional properties have made tiger nut a unique food (Ekeanyanw and Ononogbu, 2010) with edible oil (Muhammed *et al.*,

2011; Lasekan and Abdulkarim, 2012), and a source of animal feed (Sanchez- Zapata *et al.*, 2012). Tiger nut has been known for the value of the health benefit, presences of high soluble glucose and oleic content and other energy giving nutrient such as fats, starch, protein and sugars, also rich in other mineral content such as potassium, phosphorus, magnesium, iron and calcium which are essential for bones and tissue formation and also rich in vitamin C and E (Mason, 2009).

Tiger nut has both nutritional and therapeutic advantage and could serve as good alternative to cassava in the confectionary industry (Ade-Omowaye *et al.*, 2008). Tiger nut powder may be an option in the enrichment of bakery products to enhance the nutritional properties of product (Zahra and Ahmed, 2014; Aguilar *et al.*, 2015).

This work studies the effect of storage on the mycological, functional properties of tiger nut flour and sensory acceptability of tiger nut flour.

2. Materials and methods

Sample collection

Tiger nut were purchased from two local markets within Ibadan metropolis (*Ojoo* and *Bodija*). The samples were labeled and transported immediately to the Biology laboratory of the Polytechnic, Ibadan.

Production of tiger nut flour

Tubers were hand-picked to remove the spoilt, tiger nut, debris and stones before washing in sterile water to remove dirt. About 300g of each of the fresh tiger nut tubers were dried in an oven at about 120°C for 30 minutes. After drying, the tubers were milled using an electric grinder and were sieved using a 70mm mesh sieve to remove shafts. The fine flour obtained was packaged separately into a plastic container and a polythene bag stored for a period of four weeks while further analyses were carried out.

Isolation, identification and characterization of fungal growth in stored tiger nut flour

Different fungal isolates were transferred from Potatoes Dextrose Agar (PDA) incubated at 30°C for 5 days into PDA slants for further analysis. The isolate was purified by sub-culturing and grouped with respect to their morphological characteristics. A small portion of the mycelium of fungal culture was picked, placed and evenly spread on a slide, a drop of lactophenol blue was added, before covering with a cover slip and gently apply a little pressure to eliminate air bubbles. Fungi isolates were identified with reference to Atlas. The preparation was mounted and observed using a light microscope × 1000 (Watanbe, 2002).

Aflatoxin analyses

The analyses were carried out according to Joanna *et al.* (2001), using Enzyme linked immune sorbent assay (ELISA).

Determination of the proximate composition of Tiger nut flour

Moisture content, crude fiber, crude fat, ash and carbohydrates contents of samples were determined using AOAC (2010) method. Crude protein was determined by Kjeldahl method as described by Kirk and Sawyer (1991).

Determination of functional properties of the stored Tiger nut flour

Water absorption capacity was determined by the modified method of Abbey and Ibeh (1988), Oil absorption capacity was carried out according to the method of Sathe and Salunkhe (1981) and the swelling power capacity of the tiger nut flour was carried out using method of Leach *et al.* (1959).

Mineral analysis of the tiger nut flour

The mineral analysis on copper, magnesium, zinc, sodium and iron was carried out according to Paul *et al.* (2014). The tiger nut flour (5g) was weighed into the beaker and 10 ml each of nitric acid and hydrochloric acid was added and the solution was shaken on an electric shaker for an hour at 5000rpm, the solution was then filtered and made up to 100ml with de-ionized water. The filtrate was taken for Atomic Spectrophotometer analysis. Flame Photometer was used to analysed Sodium content (Chen *et al.*, 2005).

Flour blend preparation

Four blends of flours were prepared by substituting tiger nut flour for wheat flour in the percentage proportion of 100:0, 20:80, 30:70, 40:60 and 50:50 % respectively.

The recipe used was as described using modified method of Flo Chinyere (2018). Tiger nut-wheat Flour of 250g, 2 teaspoons of yeast (the one used in baking bread), teaspoon ground nutmeg, ½ cup granulated sugar (or to your taste), 3 pinches of salt, lukewarm water (for mixing) vegetable oil (for frying).

Add the flour in a bowl, mix in the sugar, nutmeg, yeast powder and salt with a spatula, add warm water gradually and mix together until ingredients blend together. Cover with aluminum foil to avoid air and keep mixture in a warm place to rise for about 45mins, after 45 mins the batter should have double its size and air bubbles seen. Pour vegetable oil in a deep pan over medium heat and heat up. Scoop the mixture with your measuring spoon into the hot oil and deep fry. Once the underside of the puff -puff has turned golden brown, flip the ball so that the topside will be fried as well. When both sides are golden brown, take out the puff- puff balls and place in a sieve to drain.

Sensory Evaluation

The sensory properties of tiger nut milk were evaluated according to Garanato *et al.* (2012), using a seven-point hedonic scale. The panel was made up of 20 persons drawn from among the student of the Polytechnic Ibadan. The panel was instructed to evaluate each sample independently for taste, colour, aroma, texture and acceptability. Carryover effect was avoided by asking panelist to rinse their mouth with water after each sample.

Data analysis

The experimental data was analyzed using Analysis of variance (ANOVA). The level of significance was $P \leq 0.05$. The data were analyzed using SPSS version 15.0, 2007.

3. Results and Discussion

The predominate fungi isolated after four weeks of storage were identified as; *Aspergillus niger*, *A. fumigatus*, *A. flavus* and *Penicillium citrinum*. The fungal load via viable spore count was estimated to be 5×10^4 after storing for 2 weeks and 8×10^4 after 4 weeks in *Bodija* tiger nut flour stored in plastic (BTP), 1.1×10^4 and 1.3×10^4 in *Bodija* tiger nut flour stored in nylon (BTN) respectively. The *Ojoo* tiger nut flour stored in plastic (OTP) has estimated viable spore count of 2.0×10^4 cfu/ml at 2 weeks and 4.0×10^4 after 4 weeks. *Ojoo* tiger nut flour stored in nylon (OTN) had viable spore counts of 8.0×10^4 and 9×10^4 after 2 and 4 weeks of storage respectively. The results indicated that all were within maximum acceptable limit as recommended by the International Commission on Microbiological Specifications for Food (ICSMF) of less than 10^5 . According to Shamsuddeen and Aminu (2016) a lower fungal population with a mean value of 1.8×10^3 cfu/g was isolated from dry tiger nut in Kaduna stored for over a year, probably due to surface area of the tiger nut flour and moisture content. However, treatment is of utmost important as ineffectual antimicrobial treatment of plant-based product at various point of production may lead to consumption of pathogen in the final product (Sagoo *et al.*, 2001).

Aflatoxin levels in stored tiger nut flour

After four weeks of storage, *Bodija* tiger nut stored in Nylon (BTN) had the highest level of contamination at 7.2 ng/g, *Bodija* tiger nut plastic (BTP) was 4.5ng/g, *Ojoo* tiger nut Nylon (OTN) 5.6 ng/g, and the lowest, *Ojoo* tiger nut plastic (OTP) 4.3ng/g (fig 1). Although none of the aflatoxin level was above the permissible standard of 15ng/g (*Codex Alimentarius*, EC 2001) was found in all the samples. This indicated that further storage of sample, may increase the aflatoxin levels, and then become a health risk. Also, the increasing values of the aflatoxin show that the storage conditions favor the production of fungal toxins. Shamsuddeen and Aminu (2016)

recorded a varying aflatoxin concentration of 0.2 μ g/kg to 23.0 μ g/kg from samples from different markets in Kaduna.

Proximate analysis of Tiger nut flour

The moisture content of dried tiger nuts from *Bodija* (TBJ) was estimated to be 8.46 %, crude protein 15.05 %, ash 2.29%, crude fiber 18.50 %, crude fat 36.19%, carbohydrate 23.97 % (Table 1). While dried tiger nuts from *Ojoo* (TOJ) yield moisture content of 8.07 %, crude protein 13.90 %, ash 1.79 %, crude fiber 18.90 %, crude fat 28.09 %, carbohydrate 30.14 %, with the exception of moisture content, the result is comparable to the work of Oladele and Aina (2007) and Suleiman *et al.* (2018) except for moisture content of 42.40% obtained from fresh tiger nuts. The result is also in agreement with the work of Sánchez - Zapata *et al.* (2012) who observed that the moisture content was lower than reported for true tubers such; yam and sweet potatoes. This result imply that the moisture content might be dependent on the variety and the dryness of tiger nuts while other properties are consistent, besides flours with moisture content above 14% are not stable (Iwe *et al.*, 2016). Codina-Torrella *et al.* (2015) suggested that nutritional content of tiger nuts may vary depending on type, geographical location and the harvesting period.

The Mineral Analysis (table 2) show that Magnesium is the highest occurring mineral while copper is the least among the minerals analysed. In a decreasing order they occurred as; magnesium, sodium, iron, zinc and copper respectively. The trend is in accordance with the work of Oladele and Aina (2007). Mineral nutrient profile is an integral in the use of tiger nut in food design. As the iron content of tiger nuts could contribute to prevention of anemia, an essential element of hemoglobin which is used in respiration, immune function and cognitive and psychomotor development (Ai *et al.*, 2012; Suleiman *et al.*, 2018; Pivinia *et al.*, 2019). The element zinc is vital for many metabolic activities, in growth, reproduction, glucose metabolism, immunity and neurological functions (Wang *et al.*, 2018). Magnesium is an essential cofactor vital in food metabolism, fatty acids and protein synthesis, transmission of nerve impulses, provides bone strengthen and heart functions (Al Alawi *et al.*, 2018; Suleiman *et al.*, 2018), while copper facilitates iron metabolism.

The functional properties are characteristics that measure the applicability of food materials in various food products (Adebowale *et al.*, 2012). The sample BTP had higher water absorption capacity at 2.17% than BTN at 2.00 % while OTP had lower water absorption of 2.46% compared to OTN at 2.97% (table 2.3). After storage for two weeks, OTN had the highest water absorption capacity. At four weeks of

storage, BTP maintained a higher water absorption capacity of 2.18% to that of BTN at 1.97% while OTN water absorption capacity was higher at 2.98% to OTP 2.46%. In comparison to the water absorption capacity of wheat flour (WF) there was no significant difference between the tiger nuts bought neither from *Bodija* and *Ojoo* market nor with the wheat flour after storage at both two weeks and four weeks. At two weeks of storage, OTN had the highest oil absorption capacity and BTP had the lowest capacity at 1.98%. After four weeks of storage, OTN had the highest oil absorption capacity at 2.57% while BTP had the lowest capacity at 1.99%. When stored for two weeks, BTP swell better than the rest at 7.34% while OTN had the least SC at 7.16%. The swelling capacity (SC) of *Bodija* tiger nut stored in plastic (BTP) was the highest at 7.20%. OTN and BTN had the lowest SC at 7.20%. Overall, there was no significant difference between the tiger nut flour and wheat flour functional properties. However, this work is limited by not evaluating dispensability and pasting properties of the tiger nut flour in comparison to wheat flour.

The sensory values of tiger nut and wheat flour puff-puff are shown in Table 4. The puff-puff tiger nut blend was well accepted. There was no significant change in colour and aroma fig. 2 (a-e) but a

significant difference was observed in taste, texture and acceptability. The result indicated that among the formulated blends, the 100% puff-puff texture was not good fig 2d and was least accepted, but 40% tiger nut and 60% wheat flour fig. 2c had the most acceptable sensory attributes.

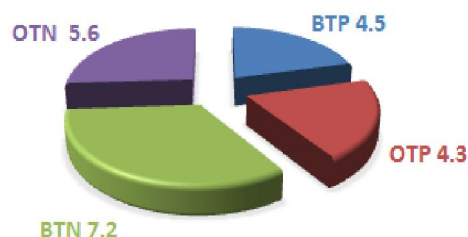


Fig 1: Aflatoxin level in stored tigernut flour (ng/g)

BTP-*Bodija* tiger nut plastic, BTN - *Bodija* tiger nut Nylon, OTP - *Ojoo* tiger nut plastic, OTN - *Ojoo* tiger nut Nylon.

Table 1: Proximate Composition of *Bodija* and *Ojoo* tiger nut samples

Samples	Moisture content%	Crude protein%	Crude fat%	Crude fibre%	Ash%	CHO%
TBJ	8.20 ^a ±0.01	14.05 ^a ±0.01	36.19 ^a ±0.01	18.50 ^a ±0.01	2.29 ^a ±0.01	23.97 ^a ±0.0
TOJ	8.07 ^a ±0.1	13.90 ^a ±0.1	28.01 ^a ±0.01	18.09 ^a ±0.01	1.79 ^a ±0.01	30.14 ^a ±0.01

TBJ –Tiger nut from *Bodija* market and TOJ- Tiger nut from *Ojoo* market

Values are means of triplicate determination with standard deviation.

Table 2: Mineral Analysis of the tigernut flour stored for 4 weeks in different packages.

Samples	Cu (mg/L)	Mg (mg/L)	Zn (mg/L)	Fe (mg/L)	Na (mg/L)
BTP	0.083 ^a ±0.01	5.60 ^a ±0.044	0.26 ^a ±0.001	2.24 ^a ±0.010	3.02 ^a ±0.166
OTN	0.088 ^a ±1.00	6.44 ^b ±0.142	0.26 ^a ±0.001	4.99 ^b ±0.450	3.20 ^a ±0.219
BTN	0.085 ^a ±0.01	6.74 ^b ±0.049	0.30 ^a ±0.010	2.39 ^a ±0.008	2.13 ^b ±0.419
OTP	0.086 ^a ±1.00	7.95 ^c ±0.90	0.30 ^a ±0.002	3.03 ^b ±0.0071	4.10 ^c ±0.252

BTP-*Bodija* tiger nut plastic, BTN - *Bodija* tiger nut Nylon, OTP - *Ojoo* tiger nut plastic, OTN - *Ojoo* tiger nut Nylon

Values are means of triplicate determination with standard deviation.

Table 3: Effect of storage on functional properties of tiger nut flour

Functional properties (%)	Tiger nut flour storage system									
	WEEK TWO					WEEK FOUR				
	BTP	BTN	OTP	OTN	WF	BTP	BTN	OTP	OTN	WF
WAC	2.17 ^a ±0.1	2.00 ^a ±0.1	2.46 ^a ±0.0	2.97 ^a ±0.1		2.18 ^a ±0.0	1.97 ^a ±0.0	2.46 ^a ±0.0	2.98 ^a ±0.0	2.12 ^a ±0.0
OAC	1.98 ^b ±0.1	2.03 ^b ±0.1	2.33 ^b ±0.1	2.62 ^b ±0.1		1.99 ^b ±1.0	2.51 ^b ±0.0	2.33 ^b ±0.0	2.57 ^b ±0.1	2.02 ^b ±0.0
SC	7.34 ^c ±0.1	7.30 ^c ±0.1	7.30 ^c ±0.1	7.16 ^c ±0.1		7.35 ^c ±0.0	7.20 ^c ±0.0	7.33 ^c ±0.0	7.20 ^c ±0.0	7.13 ^c ±0.0

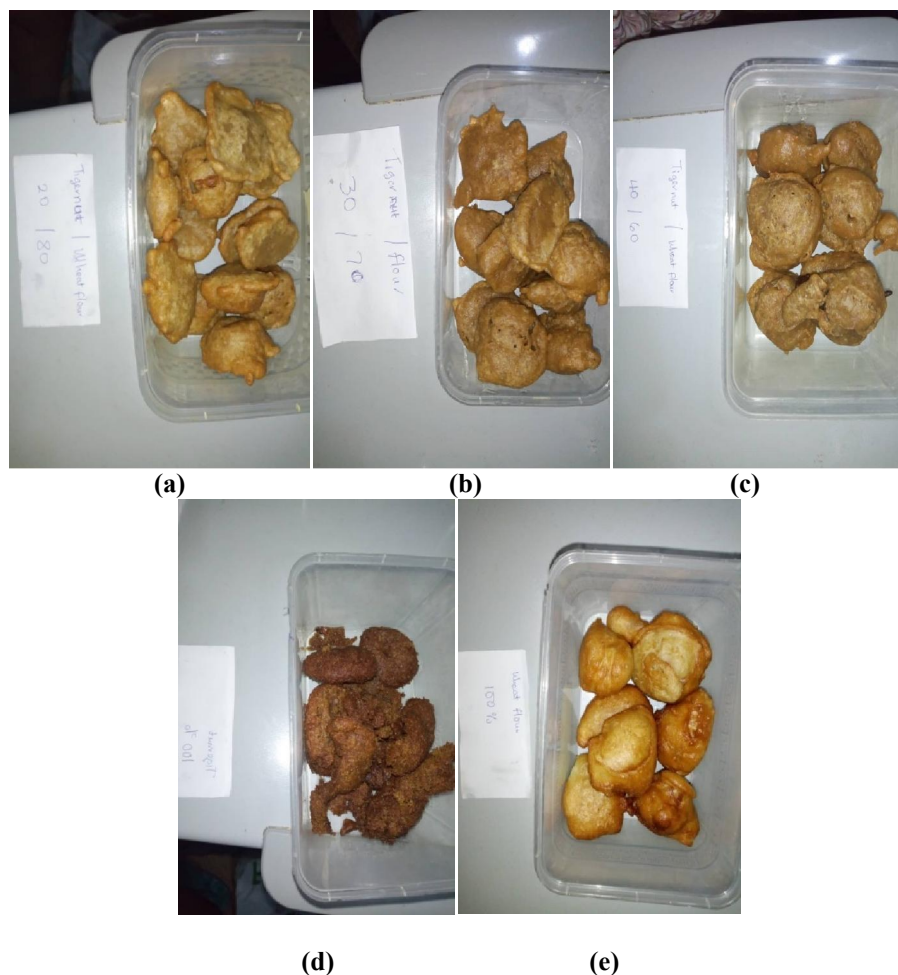
Key: WAC -Water Absorption Capacity, OAC, Oil absorption capacity, SC, Swelling capacity, BTP-*Bodija* tiger nut plastic, BTN-*Bodija* tiger nut Nylon, OTP-*Ojoo* tiger nut plastic, OTN – *Ojoo* tiger nut Nylon, WF- Wheat Flour

Values are means of triplicate determination with standard deviation.

Table 4: Sensory evaluation of formulated blends of wheat and Tiger nut flour stored for 4 weeks as Puff-Puff snack

Ration of W.F and T.F %	Taste	Colour	Aroma	Texture	Acceptability
20:80	60.0 ^a ±1.00	60.5±0.00	50.0 ^a ±1.00	46.0 ^a ±0.00	50.1 ^a ±0.00
30: 70	65.0 ^a ±1.00	62.0 ^a ±1.00	50.0 ^a ±1.00	48.0 ^a ±0.00	54.0 ^a ±1.00
40: 60	85.0 ^b ±2.00	60.0 ^a ±1.00	55.0 ^a ±1.00	81.0 ^b ±2.00	71.0 ^b ±2.00
50:50	76.0 ^c ±0.00	65.0 ^a ±1.00	52.0 ^a ±1.00	64.0 ^c ±2.00	60.0 ^c ±0.00
100:0	55.2±1.00	35.0±1.00	50.0 ^a ±1.00	26.0 ^d ±0.00	20.1 ^d ±0.00

p value = >0.05

**Fig 2: Tiger nut puff puff with the ratio of tiger nut flour to the wheat flour**

(a) Tiger nut to wheat flour ratio 20:80 (b) Tiger nut to wheat flour Ratio: 30:70 (c) Tiger nut to wheat flour ratio: 40:60 (d) Tiger nut to wheat flour ratio: 100:0 (e) Tiger nut to wheat flour ratio: 0:100

Conclusion

Tiger nut flour can be an alternative to wheat flour or an additive, in the confectionary industry. It can easily be stored for short periods; however, effectual antimicrobial treatment during prolonged storage is essential. The fiber and carbohydrate are high, hence offering a boost of energy. The array of nutrients could significantly contribute to the human metabolic processes and body refreshment. It has a unique taste and can easily blend in foods. Tiger nut flour is perfect in food designing and its potentials are endless.

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