

JOURNAL LA LIFESCI

VOL. 02, ISSUE 04 (032-053), 2021 DOI: 10.37899/journallalifesci.v2i4.421

Ochratoxins' Effects on the Functional Properties and Nutritional Compositions of Grains

Chinaza Godswill Awuchi^{1,2}, Clifford Ifeanyi Owuamanam¹, Chika C. Ogueke¹

¹Department of Food Science & Technology, The Federal University of Technology Owerri, Imo State, Nigeria ²School of Natural and Applied Sciences, Kampala International University, Kampala, Uganda



*Corresponding Author: Chinaza Godswill Awuchi Email: awuchi.chinaza@kiu.ac.ug

Article Info Article history: Received 3 September 2021 Received in revised form 22 October 2021 Accepted 27 October 2021

Keywords: Total Ochratoxins (A, B, C) Grains Proximate Composition Functional Properties Mycotoxins

Abstract Overall, the effects of total ochratoxins on the nutritional content and functional characteristics of grain flours were investigated in this research. The milling process was used for the grains cowpea, sorghum, maize, peanut, rice, millet, and acha. They were able to identify the proximate composition, functional characteristics, and total Ochratoxin levels in the samples. In this study, the researchers looked at the impact of the Ochratoxins on the nutritional and functional characteristics of the grains. The amounts of ochratoxin in grain flours varied from 0.09 to 54.41 g/kg, with some samples of rice showing no ochratoxin at all. The majority of Total Ochratoxin levels detected in the grains were higher than the WHO/EU/FAO permitted limit of 5.00 g/kg, which was exceeded. Groundnut and cowpea are both rich in protein and fiber, with a considerable amount of each. Groundnut had the greatest average fat level, accounting for 41.84 percent of total fat. It was discovered that the grains had an ash level ranging from 0.73 to 3.61 percent. The presence of ochratoxins had a substantial effect on the grain's carbohydrate, protein, and fat compositions, respectively. Their presence had a significant impact on crude fiber, ash, and functional characteristics, but had a minimal impact on the moisture content of the grain.

Introduction

Ochratoxin is a mycotoxin that exists in three secondary forms: A, B, and C. Ochratoxin is a mycotoxin that exists in three secondary forms. Ochratoxin B is the non-chlorinated arrangement of Ochratoxin A, while Ochratoxin C is an ethyl ester of Ochratoxin A. Ochratoxin A is a toxin that is toxic to humans and animals. Penicillium and Aspergillus species are responsible for the production of each and every one of them (Bayman & Baker, 2006). Occratoxin A has been shown in earlier studies to be a carcinogen and nephrotoxin in human urinary tract cancers, despite the fact that research in people has been restricted by unexplained reasons (Mateo et al., 2007; Awuchi et al., 2021a, 2020a,b). Aspergillus ochraceus and certain Penicillium species, particularly P. verrucosum and P. carbonarius, are responsible for the production of ochratoxins, which are a group of mycotoxins generated by several species of molds. Ochratoxin A is the most common and important mycotoxin in this category, whereas ochratoxin B and C are also insignificant.

In commodities such as grains, legumes, coffee, dried fruit, and wine, it is well known that ochratoxin A is present in high concentrations. Due to its prevalence in the flesh of certain animals and its potential as a cancer-causing agent in humans, it has piqued the attention of researchers. As a result, ochratoxins have the potential to infect meat and meat products. It is possible that exposure to ochratoxin via the food may induce acute toxicity in the mammalian kidneys to occur. Carriers of alleles with phenylketonuria were protected from natural abortive

treatment induced by ochratoxin exposure, which offered a heterozygous advantage to alleles despite the possibility that both parents would suffer from significant psychological delays in the event of legacy.

Grains were observed to be small and hard seeds harvested for human or animal ingestion, with or without attached hulls or fruit layers. Cereals such as wheat, maize and rye, and legumes such as cowpeas and soybeans are the two main types of commercial grain crops. The ubiquity of grains as a rich source of food informed the use of the word "grain" to describe other substances comparable to a single "grain seed" in volume or mass. Once harvested, dry grains last longer than other staple foods, like starchy fruits and tubers. This durability has made grains more suitable for industrial agriculture and processing, as they can be harvested mechanically, easily transported, stored in silos for more extended periods and milled for flour or used for oil.

The term "Mycotoxins" is used to refer to the toxic secondary intermediate compounds produced by fungal microorganisms, molds. Production and proliferation of mycotoxins before, during, and after harvesting is one of the major challenges plaguing grain industry. Ochratoxin, fumonisins, aflatoxin, citrinine, ergot alkaloids, zearalenone, and patulin are some of these mycotoxins. The term "mycotoxin" is typically reserved for toxic metabolic compounds produced only by crop- colonizing molds (Turner et al., 2009). A typical mold species may excrete various mycotoxins; various species may excrete the same or similar mycotoxin (Robbins et al., 2000; Chinaza et al., 2019). Ochratoxin- contaminated feed impact the economy of the poultry industry significantly. Chickens, ducklings, and turkeys are more vulnerable to these carcinogenic and toxic substances. Lesser egg production, a decrease in weight gains, reduced feed conversion, as well as poor quality of eggshell are common clinical symptoms of avian ochratoxicosis (Niemiec & Borzemska, 1994). In swine farms, economic damage occurs and may be attributed to nephropathy and overhead for carcass disposal. Toxicity does not seem to cause problems in cattle breeds, mainly because the protozoa in the rumen degrade Ochratoxin A (Battacone et al., 2010). However, contaminating milk is a likelihood, although rare.

Ochratoxin has been made known to be weakly mutagenic, indisputably by stimulating oxidative deoxyribonucleic acid (DNA) mutilation and causing cancer to human beings and some animals like aflatoxin B1 (Awuchi et al., 2020b; Chinaza et al., 2021a,b). Confirmation by the use of mice and rat models in experiments showed sufficient evidence of the carcinogenicity of ochratoxin A. The carcinogenicity of total ochratoxin was tested using mice and rats and was verified orally in the laboratory (Awuchi et al., 2021a). It to some extent amplified the incidence of hepatocellular carcinomas in mice. Also, the presence of ochratoxins resulted in renal adenomas and carcinomas in male mice and rats. Little histology data are available in humans, and thus no connection has been fully reported between ochratoxin A and renal cell carcinoma. However, the report has it that in Balkan patients with widespread nephropathy, the occurrence of urothelial urinary cancers is unusually high, particularly in the upper urinary tract. Although ochratoxin has nevertheless been suggested to play a leading role in decreasing antioxidant defense mechanisms, the molecular mechanisms of ochratoxin A carcinogenicity have been debated owing to the conflicting information in the literature (Cavin et al., 2007).

The purpose of this study was to investigate the impact of ochratoxins on the functional characteristics and nutritional components of grains. It clearly shown how the creation of these toxins impacts the nutrients present in grains, as well as the behavior of grain flour before and after food processing, in a scientifically valid manner. It also provided statistical information

on the impact of total ochratoxins on the nutritional compositions and functional characteristics of grains, as well as quantified the levels of total ochratoxins in these grains, as well as the proximate and functional properties of these grains. A significant physiologically dynamic component of plant-based foods is ochratoxins, which can have significant effects on the nutritional composition and functional properties of foods, particularly grains. Ochratoxins are found in high concentrations in grains and are particularly prevalent in cereals. The consumption of foods that contain it may have serious implications for the health of those who eat them

Methods

Source of Raw Materials

The various grains used in this research, including as cowpea, groundnut, sorghum, millet, maize, acha, and rice, were procured at Owerri Municipal, Owerri North, and Owerri West markets, where Ekeonunwa, Relief, and Ama-Hausa markets serve as the primary local suppliers (Owerri West).

Sample Collection and Preparation

In Owerri, between November 2016 and July 2017, three representative samples of each of the grains were gathered (chosen) at random from various marketplaces in each of the local government districts. A physical examination of the samples was performed, and they were crushed into flour using an Art's-Way portable roller mill (PRM30: USA) in accordance with AOAC guidelines (2000).

Quantitative Determination of Total Ochratoxin

The ELISA (kit) technique was utilized in this study. Extraction of ten gram (10g) of the test material using a mixture of dichloromethane and ethanol was performed (1:3). It was necessary to dilute the extract with phosphate buffer solution (PBS), which had a pH range of 7–7.5. The diluent from the extract was utilized in the analysis. Before using the kit reagents, it was necessary to bring them up to room temperature to ensure proper mixing. The antibody was initially coated onto the titer wells that will be used. Once this was done, 0.2ml of the extract diluent was placed in the indicated antibody coated well, and the toxin standard was placed in the marked toxin coated well. While the wells were rinsed three times with the phosphate buffer solution and rotated and tapped to dry, they were allowed to stand for 30 minutes at room temperature while they were dried. Later, 0.1 milliliters of the toxin conjugate were added to each well and left to stand for 10 minutes before being used. It was then necessary to add the reaction stop reagent and measure the absorbance at 450nm. The amount of ochratoxin was estimated using the following formula:

Ochratoxin (
$$\mu$$
g/1000g) = $\frac{1000}{W} \times \frac{au}{as} \times \frac{c}{va} \times vf \times D$

The vf = total volume of extract diluent, Va = the volume of extract diluent analyzed, au = absorbance of the sample extracted, as = absorbance of standard penicillic acid, c = concentration of the standard, and D = Dilution factor where applicable.

Analyses of Proximate Composition

Determination of % Moisture Content

The amount of moisture in the air was determined using a technique specified by the Association of Official Analytical Chemists (AOAC) (AOAC, 2005). Approximately 5g of the flour sample was placed in a petri dish with a specified weight and dehydrated in the oven at

105oC for approximately 4 hours before being used. The samples were weighed after they had been chilled in a desiccator. The moisture content was estimated using the following formula:

% moisture content = $\frac{\text{change in weight}}{\text{The initial weight of the sample prior to drying}} \times 100$

Determination of % Ash

The American Ornithological Association's 2005 approach was followed. In a muffle furnace at 550oC, about 5g of each sample was placed in duplicate crucibles, and the samples were burned until light gray ash was visible and the samples reached a consistent weight, at which point they were removed. The samples were chilled in desiccators to minimize moisture absorption and weighed to determine the amount of ash present.

Percentage ash was calculated using the formula: $\frac{W2 - W1}{W} \times 100$

Where; W= Dry weight of food sample, $W_1=$ weight of crucible, $W_2=$ weight of crucible and ash

Determination of Crude Protein

The protein content was determined using the micro-Kjeldahl technique published by the American Agricultural Chemistry Association (AOAC) (2005), which includes wet digestion, distillation, and titration. To determine the amount of protein present in the sample, it was placed in a boiling tube containing 25ml concentrated sulfuric acid and one catalyst tablet containing 5g H2SO4, 0.15g titanium dioxide, and allowed to boil for 15 minutes (TiO2). In order for digestion to take place, the tubes were heated at a low temperature. A solution of 100mL distilled water, 10mL 40% NaOH, and 5mL sodium thiosulfate was used to dilute the digestive enzymes (Na2S2O3). Anti-bumping agent was applied, and the sample was diluted with 10ml of Boric acid to get the desired result (H3BO3). The NH4 content of the distillate was measured by titrating it with 0.1N standard HCl in a 254ml burette and measuring the resulting concentration. A blank was created by removing the sample from the mix. The quantity of crude protein was determined by multiplying the protein value by a conversion factor (6.25), and the result was represented as the amount of crude protein.

% crude protein =
$$\%N2 \frac{100 \times N 14 \times Vf T}{W \times 1000 \times Va} \times 6.25$$

Where W = weight of sample analyzed, N = concentration of H_2SO_4 titrant, V_f = Total volume of digest, V_a = Volume of digest distilled, T = titre value – blank

Determination of % Fat Content

The amount of fat in the samples was determined using the methods developed by the AOAC (2005) and Awuchi et al (2021c). A ten-gram (10g) sample of the substance was weighed using a chemical balance and placed in a filter paper envelope for storage. It was then placed in an extraction thimble that had been cleaned and dehydrated in an oven, and then allowed to cool in desiccators before being weighed. The flask was filled with about 25ml of petroleum ether solvent, and the fat was extracted. Following the extraction, the solvent was removed from the sample by dehydrating it in the oven for several hours. The container and its contents were placed in desiccators and weighed after they had been cooled. The percentage of fat content was determined using the following formula:

$$\frac{\text{The weight of the extracted}}{\text{Weight of sample}} \times 100$$

Determination of % Crude Fibre

The amount of fiber in the sample was determined using the AOAC technique (2005). Each sample was defatted to a weight of five grams (5.0g) (during fat analysis). In a 200mL volume of 1.25 percent H2SO4 solution, the defatted sample was cooked for 30 minutes at a low temperature under reflux. Later, the samples were cleaned with several amounts of hot (boiling) water while being carefully transported quantitatively back to the flasks, where 200ml of 1.25 percent NaOH solution was carefully poured to each of them using a two-fold muslin towel to catch the particles. Again, the samples were exposed to 30 minutes of boiling and then rinsed with hot water; after that, they were carefully transferred to weighted porcelain crucibles and desiccated in an oven at 1050C for 3 hours, as was the case before. Once the samples had been allowed to cool in desiccators, they were reweighed (W2) and then put in a muffle furnace where they were burned at 5500 degrees Celsius for 2 hours, until they were reduced to ash. They were chilled in desiccators one more before being reweighed. The crude fiber content of each sample was determined gravimetrically using the following formula:

% crude fiber
$$= \frac{W2 - W3}{Weight of sample} \times 100$$

Where, W2 = the weight of crucible + the sample after washing and drying in the oven, W3 = the weight of crucible + the sample ash

Determination of % Carbohydrate

The carbohydrate composition was calculated by difference as the nitrogen-free extractive (NFE). The nitrogen-free extractive was calculated as % NFE = 100 - % (a + b + c + d + e

Where; a = protein, b = fat, c = fibre, d = ash, and e = moisture

Functional Properties Determination

The technique developed by Onwuka (2005) was utilized to determine the functional characteristics of the materials. Water and oil absorption capabilities, bulk density, swelling index, forming capacity, and emulsion capacity are some of the functional characteristics that have been determined.

Emulsion Capacity

Onwuka's technique of evaluating emulsion capacity was used to determine its capacity (2005). In a Kenwood blender, two grams of the flour sample was blended with 25 mL of distilled water at room temperature for 30 seconds until smooth (BL 330 series). Once the mixture had been well dispersed, 25ml of vegetable oil was gently added, and the blending process was repeated for another 30 seconds. After that, 15 mL of the flour sample was centrifuged at 1600 rpm for 5 minutes to extract the protein. Following that, the amount of oil that separated from the sample after centrifugation was directly measured from the table. Emulsion capacity was defined as the quantity of oil emulsified and retained per gram of sample divided by the amount of oil emulsified and retained per gram of sample

Emulsion capacity = $X/Y \times 100$

Where,

X = height of the emulsified layer, and Y = height of the whole solution in the centrifuge tube.

Swelling Index

Using the flour samples as an example, the swelling index was calculated as the ratio of the swelling volume to the ordinary volume of a unit weight of the flour sample (Awuchi et al., 2019). A gram of the material was weighed into a measuring cylinder that was clean and dry. Before adding 5ml of distilled water to the sample, it was measured how much space the sample had taken up in the container. Following that, the volume was measured and re-recorded after it had been left to stand for an hour without interruption. The following equation was used to calculate the sample's swelling ability index:

Swelling index
$$=$$
 $\frac{\text{the volume occupied by the sample after swelling}}{\text{the volume occupied by sample before swelling}}$

Water and Oil Absorption Capacity

A gram of the material was weighed into a clean conical graduated centrifuge tube and gently mixed with 10 ml of distilled water/oil using a warring mixer for 30 seconds until the sample was completely blended. After that, the sample was allowed to stand for 30 minutes at room temperature. Following that, it was centrifuged for 30 minutes at 5000 rpm. A straight reading from the graduated centrifuge tube was used to determine the amount of free water – the supernatant – or oil remaining after centrifugation. In order to convert the amount of absorbed water (wa) to weight (in grams), we multiplied it by the density of oil (0.894 g/ml) and water (1 g/ml). Each gram of flour sample had its oil and water absorption capabilities measured in milliliters of oil/water absorbed per gram of flour sample.

Absorbed water = total water - free water.

Foaming Capacity (FC)

In a Kenwood blender, 2g of the flour sample was blended with 100 mL of distilled water to create the final product. The suspension was thrashed for 5 minutes at 1600 rpm in an ace homogenizer (NSEIAM-6) with acetone. After 30 seconds, the mixture was placed into a graduated cylinder with a capacity of 250ml, and the volume recorded. The foaming capacity was expressed as a percentage increase in bulk by applying the following mathematical formula:

%Foam capacity
$$= \frac{\text{volume after whipping} - \text{volume before whipping}}{\text{Volume before whipping}} \times 100$$

Bulk Density

It was decided to utilize the gravimetric technique. A ten-gram sample was placed in a graduated 10-ml measuring cylinder after it had been weighed. After that, the cylinder's foot was repeatedly tapped on a hard pad on a laboratory bench until a steady volume was recorded. It was observed that the volume was very dense. The bulk density of the sample was calculated as the ratio of the sample weight to the volume occupied by the sample after it was tapped.

Bulk density
$$\left(\frac{g}{ml}\right) = \frac{\text{weight of sample}}{\text{volume of sample}}$$

Statistical Analyses

The statistical software used for the data analyses and refining include SPSS and Excel. The statistical analyses carried out were ANOVA, Fisher's LSD, Pearson's correlation, and Regression.

Results and Discussion

As shown in the tables below, the results demonstrate that the nutrient content and functional characteristics of the grains vary in accordance with the amount of ochratoxin in the sample.

Ochratoxins

The results of total ochratoxin levels are shown in Table 1. The levels of total ochratoxin in the grain flours ranged from 0.09 to 54.41 μ g/kg, most of which are beyond the WHO/EU permissible limit.

Grains		Ochratoxin (µg/kg)	Grains		Ochratoxin (µg/kg)	Grains		Ochratoxin (µg/kg)
Rice	Α	0.00 ± 0.00 ^c	Maize	Α	$8.20 \pm 0.02^{\text{ b}}$	Groundnut	Α	54.58 ± 0.40^{a}
	В	0.14 ± 0.04 ^b		В	1.66 ± 0.04 ^c		В	18.20 ± 0.06 ^c
	C	1.09 ± 0.12^{a}		C	13.86 ± 0.02^{a}		C	20.95 ± 0.13 ^b
	LSD	0.06		LSD	0.02		LSD	0.20
	Total	0.41 ± 0.52		Total	7.91 ± 5.29		Total	31.24 ± 17.54
Cowpea	Α	0.83 ± 0.07 ^b	Millet	Α	4.07 ± 0.06^{a}			
	В	11.57 ± 0.06^{a}		В	0.97 ± 0.01 ^b	Grand Total		7.04 ± 12.36
	С	0.26 ± 0.03 ^c		C	0.67 ± 0.04^{c}			
	LSD	0.04		LSD	0.03			
	Total	4.22 ± 5.51		Total	1.90 ± 1.63			TWI of 1.20 BW 5.00 (raw or unprocessed)
Acha	Α	$0.77\pm0.04^{\text{ c}}$	Sorghum	Α	$4.93\pm0.04^{\text{ a}}$	WHO/EU/		
	В	1.36 ± 0.04 ^b		В	0.66 ± 0.07 ^b	FAO/CODEX		
	С	$2.75\pm0.06^{\text{ a}}$		С	0.31 ± 0.03 ^c	Limits		
	LSD	0.04		LSD	0.04			
	Total	1.63 ± 0.88		Total	1.97 ± 2.23]		

Table 1. Mean values of Ochratoxin levels in the grain samples

*A, B and C represent samples from Owerri Municipal, Owerri North, and Owerri West, respectively. *P= 0.05. *TWI = Tolerable Weekly Intake. *BW = Body Weight

Consumption of these grains may expose consumers to urinary tract cancer, renal damage, and other health problems if they are poorly prepared. It was discovered that there was a very significant difference (p = 0.05) in the amounts of Ochratoxins in the grain samples from A, B, and C. Some rice flour, on the other hand, was discovered to be devoid of the substance. Ochratoxins are a group of mycotoxins generated by a variety of Aspergillus species (mostly A. ochaceus and A. niger industrial strains) and some Penicillium species, most notably P. verrucosum and P. carbonarius. Ochratoxins are toxic fungi that are toxic to humans and animals (Fratamico et al., 2008). It is a mycotoxin that can be found in three secondary metabolite forms: OTA, OTB, and OTC. It is a mycotoxin that can be found in three secondary metabolite forms: OTA, OTB, and OTC. Penicillium and Aspergillus species are responsible for the majority of the production. Because Ochratoxin B is a nonchlorinated arrangement of Ochratoxin A, and because Ochratoxin C is the ethyl ester form of Ochratoxin A, the three kinds are distinguished from one another (Ashiq, 2015; Bayman & Baker, 2006; Jeswal & Kumar, 2015). Because of the toxicity of mycotoxins, their presence in food is carefully controlled and monitored by the appropriate regulatory authorities and organizations. The European Food Safety Authority (EFSA) adopted an updated scientific estimation relating to Ochratoxin A in Food on April 4, 2006, at the request of the European Commission. The updated scientific estimation took into account new scientific evidence and derived an acceptable weekly intake (TWI) of 120ng/kg (1.2 g/kg) body weight (Awuchi et al., 2021a). Ochratoxin has been identified as a potential human carcinogen, and it has been linked to urinary tract cancer and kidney damage in individuals from the Eastern European countries (Awuchi et al., 2021a). Ochratoxin exposure seems to be the most serious danger associated with fungus for grain eaters in Europe, according to recent research (Ashiq, 2015; Richard, 2007).

Proximate composition

The average values of the proximate composition of the grains are as shown in Table 2. Figure shows the proximate composition of the grains.



Figure 1. Mean proximate composition of the grains

Using the 0.05 significance level, the results in Table 2 reveal that there were statistically significant differences in the proximate composition (carbohydrates, protein, fat, fiber, ash, and moisture) between the grains. These were in agreement with the findings of Iwe et al (2016). However, with the exception of millet and acha, there was no statistically significant variation in the carbohydrate levels of the grain samples from A, B, and C.

Grain		% Crude Protein	% Crude Fat	% Crude Fibre	% Ash	% Moisture	% Carbohydrate
Rice	А	7.58 ± 0.04^{c}	$0.85\pm0.03^{\text{ b}}$	$0.94\pm0.02^{\text{ a}}$	$0.88\pm0.02^{\text{ b}}$	8.18 ± 0.58^{a}	81.54 ± 0.57^{a}
	В	8.97 ± 0.06^{a}	$0.88\pm0.02^{\text{ a}}$	0.89 ± 0.02^{a}	0.73 ± 0.02^{c}	7.87 ± 0.56^{a}	$80.66\pm0.61^{\text{ a}}$
	С	8.60 ± 0.60^{b}	0.76 ± 0.03^{c}	$0.97\pm0.05^{\text{ a}}$	0.95 ± 0.02^{a}	8.07 ± 0.03^{a}	80.65 ± 0.62^{a}
	LSD	0.29	0.02		0.02		
	Total	8.38 ± 0.69	0.83 ± 0.06	0.93 ± 0.05	0.86 ± 0.10	8.04 ± 0.42	80.95 ± 0.68
Cowpea	А	$20.19\pm0.04^{\rm \ a}$	$2.17\pm0.04^{\rm \ a}$	$4.88\pm0.09^{\rm \ a}$	3.44 ± 0.07 ^b	$9.46\pm0.02^{\text{ b}}$	59.87 ± 0.06^{a}
	В	20.69 ± 1.28^{a}	2.04 ± 0.03^{c}	$4.59\pm0.05^{\text{ b}}$	3.22 ± 0.04^{c}	10.61 ± 0.07 ^a	58.85 ± 1.24^{a}
	С	22.04 ± 1.23^{a}	$2.14\pm0.04^{\text{ b}}$	4.21 ± 0.05 °	3.57 ± 0.06^{a}	$9.32\pm0.52^{\text{ b}}$	58.71 ± 1.72^{a}
	LSD		0.03	0.05	0.05	0.25	
	Total	20.98 ± 1.22	2.12 ± 0.07	4.56 ± 0.29	3.41 ± 0.16	9.80 ± 0.66	59.14 ± 1.19
Acha	А	8.33 ± 0.02^{a}	$0.93 \pm 0.02^{\text{ b}}$	1.88 ± 0.02^{b}	$1.85 \pm 0.03^{\text{ b}}$	11.69 ± 0.03^{a}	$75.32 \pm 0.02^{\circ}$
	В	7.57 ± 0.02^{b}	1.16 ± 0.02^{a}	1.93 ± 0.04^{a}	$1.52 \pm 0.02^{\circ}$	10.40 ± 0.59^{b}	77.43 ± 0.61 ^b

 Table 2. Mean Values of the Proximate Composition of the Grain Samples

ISSN 2721-1304 (Print), ISSN 2721-1207 (online) Copyright © 2021, Journal La Lifesci, Under the license CC BY-SA 4.0

	C	$7.24 \pm 0.05^{\circ}$	0.88 ± 0.02^{c}	$1.68 \pm 0.05^{\circ}$	1.90 ± 0.02^{a}	$9.69 \pm 0.06^{\circ}$	78.62 ± 0.11^{a}
	LSD	0.03	0.02	0.03	0.02	0.28	0.29
	Total	7.71 ± 0.49	0.99 ± 0.13	1.83 ± 0.12	1.75 ± 0.18	10.59 ± 0.93	77.12 ± 1.48
Maize	A	8.76 ± 0.03^{a}	$4.11 \pm 0.02^{\circ}$	$2.73\pm0.60^{\rm \ a}$	1.59 ± 0.04^{a}	10.38 ± 0.03^{a}	72.43 ± 0.61 a
	В	8.72 ± 0.35^{a}	$4.22 \pm 0.02^{\ b}$	$2.53\pm0.04^{\rm \ a}$	$1.08 \pm 0.02^{\text{ b}}$	$9.82 \pm 0.60^{\text{ b}}$	73.63 ± 0.89^{a}
	C	$8.23 \pm 0.03^{\ b}$	$4.50\pm0.03^{\text{ a}}$	2.64 ± 0.01 ^a	1.54 ± 0.35^{a}	9.13 ± 0.06 °	73.96 ± 0.37^{a}
	LSD	0.17	0.02		0.17	0.29	
	Total	8.57 ± 0.31	4.28 ± 0.18	2.63 ± 0.32	1.40 ± 0.30	9.78 ± 0.62	73.34 ± 0.90
Millet	Α	10.89 ± 0.02^{a}	5.11 ± 0.05 °	$2.13 \pm 0.03^{\circ}$	$1.83 \pm 0.02^{\circ}$	8.37 ± 0.03^{a}	$71.68 \pm 0.03^{\circ}$
	В	10.21 ± 0.04 ^b	$5.42 \pm 0.02^{\text{ b}}$	$2.39\pm0.03^{\text{ a}}$	1.94 ± 0.04 ^b	6.69 ± 0.04 ^c	73.35 ± 0.06^{a}
	С	10.11 ± 0.03 °	$5.69\pm0.05^{\text{ a}}$	$2.20 \pm 0.02^{\text{ b}}$	$2.09\pm0.05^{\text{ a}}$	6.84 ± 0.05 ^b	73.07 ± 0.04^{b}
	LSD	0.03	0.03	0.02	0.03	0.03	0.03
	Total	10.40 ± 0.37	5.41 ± 0.25	2.24 ± 0.12	1.95 ± 0.12	7.30 ± 0.80	72.70 ± 0.78
Sorghu m	А	9.87 ± 0.04^{a}	$3.16 \pm 0.06^{\circ}$	2.21 ± 0.04 ^b	1.69 ± 0.04 °	$10.43 \pm 0.06^{\text{b}}$	72.27 ± 0.65^{a}
	В	9.33 ± 0.55^{a}	3.56 ± 0.06^{a}	$2.22 \pm 0.03^{\text{ b}}$	1.76 ± 0.05 ^b	11.00 ± 0.03^{a}	72.13 ± 0.53^{a}
	С	9.48 ± 0.53 a	$3.22 \pm 0.06^{\ b}$	2.37 ± 0.06^{a}	1.89 ± 0.02 a	9.71 ± 0.07 ^c	73.34 ± 0.42^{a}
	LSD		0.05	0.04	0.03	0.04	
	Total	9.52 ± 0.47	3.31 ± 0.19	2.27 ± 0.08	1.78 ± 0.09	10.38 ± 0.56	72.58 ± 0.74
Ground nut	А	$24.00 \pm 0.13^{\text{ b}}$	42.35 ± 0.41 ^a	$3.59 \pm 0.05^{\text{ b}}$	$3.08\pm0.09^{\text{ a}}$	$8.43\pm0.04^{\rm \ a}$	18.55 ± 0.29^{a}
	В	$22.72 \pm 0.58^{\circ}$	41.63 ± 1.04^{a}	3.74 ± 0.05^{a}	$2.92\pm0.05^{\text{ a}}$	9.07 ± 0.55 a	19.93 ± 1.50^{a}
	С	24.87 ± 1.05^{a}	41.55 ± 0.56^{a}	3.11 ± 0.07 ^c	3.10 ± 0.10^{a}	8.59 ± 0.61 ^a	18.78 ± 1.28^{a}
	LSD	0.57		0.04			
	Total	23.86 ± 1.11	41.84 ± 0.73	3.48 ± 0.29	3.03 ± 0.11	8.70 ± 0.50	19.09 ± 1.19
Total		12.83 ± 6.33	8.40 ± 13.86	2.56 ± 1.11	2.03 ± 0.85	9.23 ± 1.32	64.99 ± 19.93

*A, B and C represent samples from Owerri Municipal, Owerri North, and Owerri West, respectively. *P=0.05

Table 2 and Figure 2 show that the moisture content of the grain samples varied from 6.65 to 11.72 percent (acha had the highest average moisture content, 11.69 0.03%), while millet had the lowest, 6.69 0.04% (see Table 2 and Figure 2 for more information). It was found that the greatest (11.72 percent) and lowest (6.65 percent) moisture content were found in acha from Owerri city and millet from Owerri North, respectively. The difference in moisture content between the grain samples from the three local government regions was statistically significant at p = 0.05, according to the results. The relatively low moisture level of the grains suggests that they will be stable throughout storage and shelf life. Adebayo-Oyetoro et al. (2011) reported the proximate, functional, and pasting characteristics of FARO 44 rice, African yam bean, and brown cowpea seeds mixed flour, while Iwe et al. (2016) reported the proximate, functional, and pasting properties of OFADA rice. It is stated in the American Association of Cereal Chemists' recognized techniques for assessing several aspects of flour's characteristics, that the greater the moisture content of a meal is, the smaller the amount of dry solids in the meal is. Flour specifications often restrict the amount of moisture in the flour to 14 percent or less. At room temperature, flours that have a moisture content more than 14 percent are not stable, and as a result, microorganisms that are present in them will proliferate, resulting in the production of offensive smells and tastes (Iwe et al., 2016; Chinaza, 2019; Twinomuhwezi et al., 2020).

Carbs: At a p value of 0.05, there was a statistically significant variation in the carbohydrates content of millet and acha from various local governments. A statistically significant difference did not exist between the carbohydrates contained in the other grains. Table 2 and Figure 3

show that rice had the greatest average carbohydrate content, while groundnut had the lowest average carbohydrate content. Grains with increasing average carbohydrate content were groundnut, cowpea, sorghum, millet, maize, acha, and rice in that order, with groundnut being the highest. Despite the fact that carbohydrates are very important for the body's metabolic and physiological processes (Awuchi & Amagwula, 2021), they have also been connected to metabolic disorders such as obesity when eaten in large quantities over an extended length of time (Chinaza et al., 2020a,b; Tufail et al., 2021; Yasmin et al., 2021)

It was discovered that the average protein content of the grains differed significantly (p = 0.05) between the two groups. Groundnut and cowpea have a higher protein level than sorghum, rice, acha, millet, and maize, which all have lower protein contents (see Table 2 and Figure 4). Due to the fact that legumes (groundnut and cowpea) have been proven to have higher protein content than their cereal counterparts (rice, acha, millet, sorghum and maize), this statistically significant difference in protein content was anticipated. Legumes are a high-protein food that is easy to prepare (Udeogu & Awuchi, 2016). The greatest average crude protein concentration was found in groundnut, whereas the lowest was found in acha. Proteins are required for the development and repair of tissues (Ahaotu et al., 2020a,b; Awuchi et al., 2021b)





Figure 2. Mean moisture content of grains from each LGA

Figure 3. Mean carbohydrates content of grains from each LGA

ISSN 2721-1304 (Print), ISSN 2721-1207 (online) Copyright © 2021, Journal La Lifesci, Under the license CC BY-SA 4.0



Figure 4. Mean crude protein content of grains from each LGA

Fats: Other grains, with the exception of groundnut, had relatively low fat content (see Table 2 and Figure 5), ranging from 0.73 percent in rice to 5.73 percent in millet from Owerri west (see Table 2). A significantly higher fat content was found in groundnut, with the greatest percentage (42.68 percent) found in groundnut from Owerri north. The fat content of the grains differed significantly (p = 0.05) from one another in a visible manner. Rice, millet, cowpea, acha, sorghum, and maize have low fat content, which may be due to the fact that cereals, certain legumes, and tubers store energy as starch rather than lipids, as do some legumes and tubers (Iwe et al., 2016). The low-fat content is beneficial since it ensures that the goods will have a longer shelf life because all fats and fat-containing meals include some unsaturated fatty acids and, as a result, are possibly susceptible to rancidification (Chinaza et al., 2018).

Ash content: The average ash content of the grains varied from $0.73\ 0.02$ percent to $3.57\ 0.06$ percent, with rice having the lowest average percent ash content and cowpea having the highest average percent ash content, respectively (see Table 2 and Figure 6). With the exception of groundnut, the difference in ash content of the grains from A, B, and C was statistically significant at p = 0.05 for all three groups. The ash content of the grains may be used to estimate the total mineral content of the grains by analyzing the ash content (Twinomuhwezi et al., 2020). When organic components and moisture are converted into CO2 and nitrogen oxides, the total inorganic components are calculated (Awuchi et al., 2020d). A mineral is an essential element (trace element) that is needed by organisms (including humans) in small amounts in order to sustain processes that are vital for life (Awuchi et al., 2020e). They may act as cofactors to enzymes, allowing them to perform more efficient and effective metabolic functions. They are also components of some biomolecules, including hemoglobin, myoglobin, Adenine Triphosphates, NADP, chlorophyll, and nucleic acids, among others. They are found in high concentrations in hemoglobin, myoglobin, and Adenine Triphosphates.



Figure 5. Mean crude fat content of grains from each LGA



Figure 6. Mean ash content of grains from each LGA



Figure 7. Mean crude fibre content of grains from each LGA

Fiber: With the exception of rice and maize, a statistically significant difference (p = 0.05) in the average fiber content of the grains was found. Cowpea and rice have the greatest and lowest average fiber contents, respectively, when compared to other crops. Rice, acha, millet, sorghum, maize, groundnut, and cowpea were the crops with the highest fiber content, followed by acha, millet, sorghum, and cowpea (see Table 2 and Figure 7). From 0.93 percent to 4.96 percent, the crude fiber content of the grains was measured. Cowpea and groundnut fiber contents were not very impressive, despite their relative high fiber content. This is due to the high crude fiber content of legumes, which may explain why they are so nutritious. Crude fiber slows the release of glucose into the bloodstream and lowers intracolonic pressure, lowering the risk of colon cancer, diverticulosis, and other gastrointestinal problems (Chinaza, 2019; Twinomuhwezi et al., 2020). The presence of fiber, despite the fact that it is indigestible, is critical in maintaining the integrity of the gastrointestinal (GI) tract as well as overall health. It helps to avoid constipation and diverticular illness, as well as aids the management of body weight, blood glucose, and cholesterol levels in the bloodstream. Increased fiber helps to reduce the risk of obesity and improves intestinal transit, in addition to lowering the risk of cardiovascular disease.

Functional properties

The results of the functional properties of the grains are shown in Table 3.

Grains		Oil Absorption Capacity	Water Absorption Capacity	Bulk Density	Swelling Index	Emulsion Capacity	Foaming Capacity
		ml/g	ml/g	g/ml		%	%
Rice	Α	$1.25 \pm 0.03^{\text{ b}}$	3.30 ± 0.01^{a}	0.72 ± 0.01 a	1.40 ± 0.01 ^c	$46.20 \pm 0.06^{\circ}$	8.51 ± 0.03 ^b
	В	1.31 ± 0.01 ^a	3.18 ± 0.02 °	0.71 ± 0.00^{a}	1.46 ± 0.02 b	$47.26\pm0.05^{\text{ a}}$	8.55 ± 0.04^{a}
	C	$1.25 \pm 0.02^{\text{ b}}$	3.26 ± 0.03^{b}	0.71 ± 0.01 ^a	1.53 ± 0.04^{a}	$46.82 \pm 0.08^{\text{ b}}$	8.43 ± 0.04 ^c

Table 3. Mean Values of the Functional Properties of Grain Samples

ISSN 2721-1304 (Print), ISSN 2721-1207 (online) Copyright © 2021, Journal La Lifesci, Under the license CC BY-SA 4.0

	LSD	0.02	0.02		0.02	0.05	0.03
	Total	1.27 ± 0.03	3.24 ± 0.06	0.71 ± 0.01	1.47 ± 0.06	46.76 ± 0.46	8.49 ± 0.06
Cowpea	Α	$2.21\pm0.02^{\text{ b}}$	$1.85 \pm 0.02^{\text{ b}}$	$0.71 \pm 0.00^{\text{ b}}$	1.06 ± 0.04^{a}	63.48 ± 0.08 ^c	14.17 ± 0.03^{a}
	В	$2.10 \pm 0.06^{\circ}$	$1.87 \pm 0.01^{\text{ a}}$	$0.72\pm0.00^{\rm \ a}$	$1.14\pm0.05^{\rm \ a}$	67.68 ± 0.15^{a}	14.24 ± 0.15^{a}
	С	$2.28\pm0.03^{\rm \ a}$	$1.67 \pm 0.03^{\circ}$	0.70 ± 0.00^{c}	1.07 ± 0.02^{a}	$67.31 \pm 0.10^{\text{ b}}$	$14.26\pm0.03^{\text{ a}}$
	LSD	0.03	0.02	0.003		0.09	
	Total	2.20 ± 0.09	1.80 ± 0.10	0.71 ± 0.01	1.09 ± 0.05	66.15 ± 2.02	14.22 ± 0.09
Acha	Α	$1.82\pm0.01^{\text{ a}}$	2.64 ± 0.04^{a}	0.77 ± 0.01 a	1.21 ± 0.02^{a}	46.12 ± 0.04 ^b	8.11 ± 0.02^{a}
	В	$1.76\pm0.03^{\text{ b}}$	$2.47\pm0.04^{\text{ b}}$	0.77 ± 0.01 a	$1.17 \pm 0.03^{\text{ b}}$	$46.34\pm0.06^{\rm \ a}$	8.01 ± 0.03 ^c
	С	$1.77\pm0.02^{\text{ b}}$	$2.42\pm0.03^{\text{c}}$	$0.73\pm0.00^{\text{ b}}$	1.13 ± 0.04 ^c	44.84 ± 0.11 ^c	$8.07\pm0.04^{\text{ b}}$
	LSD	0.02	0.03	0.01	0.02	0.06	0.03
	Total	1.78 ± 0.03	2.51 ± 0.10	0.76 ± 0.02	1.17 ± 0.04	45.77 ± 0.70	8.06 ± 0.05
Maize	Α	$2.15\pm0.02^{\rm \ a}$	$3.04\pm0.03^{\text{ b}}$	$0.61\pm0.00^{\text{ b}}$	$1.26\pm0.03^{\text{ b}}$	48.79 ± 0.16^{a}	9.85 ± 0.15^{a}
	В	$2.10\pm0.02^{\text{ b}}$	3.10 ± 0.03^{a}	0.64 ± 0.01^{a}	1.29 ± 0.02^{a}	$48.57 \pm 0.03^{\text{ b}}$	9.67 ± 0.03^{b}
	С	2.04 ± 0.03^{c}	$3.01 \pm 0.02^{\circ}$	0.64 ± 0.00^{a}	$1.20 \pm 0.03^{\circ}$	$46.21 \pm 0.08^{\circ}$	9.58 ± 0.04^{c}
	LSD	0.02	0.02	0.01	0.02	0.09	0.07
	Total	2.10 ± 0.05	3.05 ± 0.04	0.63 ± 0.02	1.25 ± 0.04	47.86 ± 1.24	9.70 ± 0.14
Millet	Α	$1.87\pm0.02^{\text{ b}}$	$2.08\pm0.05^{\rm \ a}$	0.67 ± 0.00^{a}	1.31 ± 0.04 ^b	$57.86 \pm 0.29^{\circ}$	7.31 ± 0.04^{a}
	В	$1.96\pm0.02^{\rm \ a}$	$2.16\pm0.04^{\rm \ a}$	$0.68\pm0.01^{\text{ a}}$	$1.35\pm0.03^{\text{ a}}$	$59.45\pm0.56^{\rm \ a}$	$7.14\pm0.14^{\rm \ a}$
	С	$1.77\pm0.02^{\circ}$	2.06 ± 0.06^{a}	$0.67\pm0.02^{\text{ a}}$	1.24 ± 0.03 °	58.41 ± 0.24 ^b	7.25 ± 0.06^{a}
	LSD	0.02			0.03	0.32	
	Total	1.87 ± 0.08	2.10 ± 0.06	0.67 ± 0.01	1.30 ± 0.05	58.57 ± 0.78	7.23 ± 0.11
Sorghum	Α	$2.11\pm0.02^{\text{ b}}$	$3.26\pm0.05^{\rm \ a}$	0.66 ± 0.00^{a}	1.05 ± 0.04 ^c	43.06 ± 0.04 ^b	$7.56\pm0.03^{\text{ b}}$
	В	$2.20\pm0.02^{\rm \ a}$	$3.16\pm0.04^{\text{ b}}$	0.67 ± 0.03^{a}	$1.10 \pm 0.02^{\text{ b}}$	44.69 ± 0.02^{a}	7.51 ± 0.01 ^c
	С	$2.12\pm0.03^{\text{ b}}$	3.25 ± 0.03^{a}	0.66 ± 0.01^{a}	1.27 ± 0.03^{a}	43.18 ± 0.28^{b}	7.61 ± 0.01^{a}
	LSD	0.02	0.03		0.02	0.13	0.01
	Total	2.15 ± 0.05	3.22 ± 0.06	0.66 ± 0.02	1.14 ± 0.10	43.64 ± 0.80	7.56 ± 0.05
Groundnut	Α	$2.20\pm0.03^{\text{ b}}$	$1.67 \pm 0.03^{\text{ b}}$	$0.57\pm0.01^{\text{ a}}$	1.35 ± 0.03^{a}	$52.61 \pm 0.02^{\circ}$	8.66 ± 0.04^{a}
	В	2.33 ± 0.02^{a}	1.58 ± 0.00^{c}	0.55 ± 0.01 ^c	1.29 ± 0.02^{a}	55.03 ± 0.07^{a}	8.57 ± 0.02^{b}
	C	2.12 ± 0.02^{c}	$1.76\pm0.03^{\text{ a}}$	$0.56\pm0.00^{\text{ b}}$	$1.33\pm0.02^{\text{ a}}$	$53.10 \pm 0.02^{\text{ b}}$	8.31 ± 0.06 °
	LSD	0.02	0.02	0.01		0.03	0.03
	Total	2.22 ± 0.09	1.67 ± 0.08	0.56 ± 0.01	1.32 ± 0.03	53.58 ± 1.11	8.51 ± 0.16
Total		1.94 ± 0.32	2.51 ± 0.63	0.67 ± 0.06	1.25 ± 0.13	51.76 ± 7.68	9.11 ± 2.23

*A, B and C represent samples from Owerri Municipal, Owerri North, and Owerri West, respectively. *P=0.05

As indicated in Table 3, there was a statistically significant variation in the foaming capacity of the grain flours from the three local governments, with the exception of millet and cowpea. The foaming capacity of a protein refers to the amount of interfacial area that can be produced by the protein while it is in a foaming state (Fennama, 1996). Cowpea flour had the greatest average foaming capacity, with cowpea from Owerri West (14.26 0.03 percent) having the highest value, while millet flour had the lowest average foaming capacity. Cowpea flour was found to have the highest average foaming capacity. Foam is a colloidal suspension of different gas bubbles that is suspended in a liquid or solid. Tiny liquid coatings encircle small bubbles, preventing them from escaping (Suresh et al., 2014).

Bulk density: There was just a minor difference in bulk density between the two groups. In Table 3, it can be shown that there was a statistically significant difference in bulk density in groundnut and cowpea from A, B, and C, while there was no statistically significant difference in bulk density in the remaining grain samples, at p = 0.05. Groundnut had the lowest average bulk density, with groundnut from Owerri north having the lowest (0.55 0.01 g/ml), while acha

had the greatest average bulk density, with groundnut from Owerri south having the lowest (0.55 0.01 g/ml). According to certain theories, the minor change in bulk density may be due to the differential in starch concentration (Iwe et al., 2016). According to Iwe & Onuh (1992) and Iwe & Onadipe (2001), higher starch content resulted in increased bulk density in the aforementioned studies. Bulk density is also dependent on factors such as the method of measurement, geometry, dimension, surface properties of materials, and so on. It can be increased when the particles are smaller, compactible, properly tapped or vibrated, and packaged in the appropriate packaging material, all of which can be beneficial (Awuchi et al., 2020b). It is possible that this is the reason why the average bulk density of acha and rice is considerably greater. It is important to note that bulk density reflects the relative capacity of packing material needed (Iwe et al., 2016). The greater the bulk density, the denser the package material required to accommodate it. It indicates the porosity of a product, which has an effect on the package design and may be used to determine the kind of packing material needed for the product (Iwe & Onadipe, 2001).

Efficacy in emulsion formation and stabilization: Protein, as a surface active agent, has the ability to create and stabilize emulsions by creating electrostatic repulsion on the oil droplet surface (Kaushal et al., 2012). According to the results in Table 3, the average values of the grains' emulsion capacity varied from 43.06 0.04 percent to 67.68 0.15 percent on a percentage basis. When it came to emulsion capacity, there was a considerable variation between the grain samples. Cowpea flour had the greatest average emulsion capacity, while sorghum flour exhibited the least. It is possible that the high emulsion capacity of cowpea flour is related to the high protein content of the flour.

Swelling index: The mean values of the swelling index of the grain flours varied very little from one another. The greatest value, $1.53\ 0.04$, was found in the rice sample, while the lowest value, $1.05\ 0.04$, was found in the sorghum sample. The average values of the swelling indices of cowpea and groundnut from the three local governments did not exhibit a statistically significant difference, while the mean values of the other grain samples did indicate a statistically significant difference at p = 0.05. In certain high-grade preparations, such as bread goods, swelling capacity is considered a quality indicator (Iwe et al., 2016). Non-covalent bonding between molecules inside starch granules is indicated by the presence of amylopectin, which is a factor of the ratio of -amylose to amylopectin (Awuchi et al., 2020b,f).

A significant difference (variations) was found between the grain samples, with the exception of the millet samples, at p = 0.05 in the Water Absorption Capacity (WAC) of the grain samples, as shown in Table 3. The WAC varied from 1.58 to 3.29 milliliters per gram of protein. Groundnut and rice samples were found to have the lowest and highest levels of phosphorus, respectively. WAC is a functional characteristic that is required in all food compositions, but is particularly important in those that involve dough handling (Lorenz & Collins, 1980). Protein concentrations, degree of interaction with water, and structural features of the grain samples all have the potential to cause apparent differences in WAC across the grain sample samples. The loose connection between amylose and amylopectin in the starch granules, as well as reduced associative forces that help to maintain the granular structure, may have resulted in the consequence. A product's ability to absorb water is critical in the bulking and stability of the product, as well as in baking applications (Lorenz and Collins, 1980). Therefore, in baking applications, the sample that has the greatest water absorption capacity is the one that is most readily identified (Iwe et al., 2016). WAC increases have always been associated with increased amylose leaching and solubility, as well as degradation to the crystalline structure of starch, according to research. More hydrophilic components, such as polysaccharides, are likely to be found in flour with the greatest WAC. Since protein exhibits

both hydrophilic and hydrophobic tendencies, it can interact with water in foods in a variety of ways (Suresh et al., 2014).

Oil Absorption Capacity (OAC) is a measure of a material's ability to absorb oil. Having a high oil absorption capacity is a key functional characteristic that enhances the mouthfeel of food items while also being retentive to their taste (Adebowale & Lawal, 2004). There was a significant variation in the OAC of the grain samples compared to one another. The average OAC in the groundnut sample was 2.22 0.09 ml/g, whereas the average OAC in the rice sample was 1.27 0.03 ml/g. The groundnut sample had the greatest average OAC, while the rice sample had the lowest average OAC. This suggests that the amount of fat in the flours may have had an impact on the OAC of the flours. Protein and fat are the two most important proximal components that influence OAC. Both hydrophilic and hydrophobic components make to a protein's structure. Interaction between non-polar amino acid side chains and hydrocarbon chains in lipids is known as hydrophobic interaction (Jitngarmkusol et al., 2008).

Influences of Ochratoxins on the nutritional compositions of the grain samples

Table 4 shows the correlation results of total ochratoxins and proximate composition.

Model	R	R Square
% Carbohydrates	.925	.856
% Moisture	.410	.168
% Ash	.699	.489
% Crude fibre	.702	.493
% Crude fat	.971	.943
% Crude protein	.897	.805

Table 4. Correlation of Ochratoxins and proximate composition

Ash: According to Table 4, the correlation between percent ash and Ochratoxins was 0.699, which showed a moderately strong connection. The R2 value indicated how the Ochratoxins were able to explain a significant portion of the overall variance in the % ash content. When the R2 value of 0.489 was obtained, it indicated that the effect of the Ochratoxins on the percent moisture content of the grains was moderate. It was discovered that the presence of Ochratoxins was responsible for 48.9 percent of the variance in percent ash, with the remaining 51.1 percent possibly attributable to other factors. That the presence of Ochratoxins had a substantial impact on the percent ash content of the grains was shown by this finding.

Carbohydrates: An R-value of 0.925 in Table 4 showed a significant degree of connection between the two variables. According to the R square value, the Ochratoxins may explain how much of the overall variance in the percent carbs might be explained by them. The correlation coefficient (R2) of 0.856 showed that the impact of the Ochratoxins on the percent carbohydrate content of the grains was very strong. In other words, the synthesis of Ochratoxins is responsible for 85.6 percent of the shift in the carbohydrates content of the grains, with the remaining 14.4 percent being attributable to other causes.

Raw fiber: The R-value in Table 4 was 0.702, which showed a moderately high degree of association between the Ochratoxins in the grains and the percent crude fiber content in the grains. It was determined that the Ochratoxins had a little negative impact on the percent crude fiber content of the grains by calculating an R2 value of 0.493. In other words, the synthesis of Ochratoxins resulted in a 49.3 percent increase in the percent crude fiber content of the grains.

Crude protein: The results in Table 4 reveal an R-value of 0.897, indicating a very high degree of correlation between crude protein and total protein. When the R2 value of 0.805 was calculated, it indicated that the Ochratoxins had a significant impact on the percent crude protein content of the grains. The presence of the Ochratoxins resulted in a change in the crude protein content of the grains of about 80.5 percent.

Crude fat: The R-value in Table 4 showed the degree of connection between the Ochratoxins and the percent crude fat content of the grains. Ochratoxins and crude fat: The R-value of 0.971 showed a very high degree of correlation between the two variables. According to the R square value, the Ochratoxins might explain how much of the overall variance in % crude fat could be explained by their presence. When the R2 value was calculated, it was found to be very high, indicating that the Ochratoxins had a significant impact on the percent crude fat content of the grains. In other words, the presence of Ochratoxins resulted in a 94.3 percent increase in the crude fiber content of the grains, with the remaining 5.7 percent likely attributable to a variety of other reasons.

% Moisture Content: The R-value in Table 4 showed the degree of connection between the Ochratoxin levels and the percent moisture content of the grains. When compared to the percent carbs, an R-value of 0.410 indicated that there was a minimal connection. Using the R square value, researchers were able to determine how much of the overall change in percent moisture could be explained by Ochratoxins. The R2 value of 0.168 indicated that the impact of the Ochratoxins on the percent moisture content of the grains was insignificant, according to the data. The addition of the Ochratoxins had a negligible effect on the moisture content of the grains in this experiment.

Influences of Ochratoxins on the functional properties of the grain samples

Table 5 shows the correlation results of total ochratoxins and functional properties.

Model	R	R Square
WAC	.682	.465
Bulk density	.738	.545
Swelling index	.501	.251
Emulsion capacity	.498	.248
Foaming capacity	.507	.257
OAC	.604	.365

 Table 5. Correlation of Ochratoxins and functional properties

Result in Table 5 indicates an R-value of 0.501, indicating a moderate connection between the swelling index (SI) and the Ochratoxin concentrations. As shown by the R2 value of 0.251, the Ochratoxins had little impact on the SI of both grains flours and whole grains flours. It was discovered that the presence of Ochratoxins in the flour was responsible for 25.1 percent of the variance in the SI of the flour, with the remaining percent perhaps attributable to other factors.

A moderate connection between oil absorption capacity (OAC) and the Ochratoxins was found in the results of Table 5, with a R value of 0.604, indicating a moderate association between oil absorption capacity (OAC) and the Ochratoxins. The R2 value of 0.365 indicated that the impact of the Ochratoxins on the OAC of the grains flours was only marginally positive, while the value of 0.365 indicated a somewhat negative effect. It was discovered that the presence of the Ochratoxins in the flour was responsible for 36.5 percent of the variance in the OAC of the flour, with the remaining percent being attributable to other factors. This demonstrates that the presence of Ochratoxins had a somewhat negative impact on the Oil Absorption Capacity of the grains, which is a good thing.

Fructose-degrading capacity (FC): The R value in Table 5 showed how well the Ochratoxins and the foaming capacity of the grain flour were correlated. The correlation coefficient (R) was 0.507, indicating a modest degree of connection. Ochratoxins were shown to be responsible for a significant portion of the overall difference in foaming capacity, as indicated by the R square value. When the R2 value was 0.257, it was determined that the impact of the Ochratoxins on the FC of the grains was only marginally significant. In other words, the generation of Ochratoxins was responsible for 25.7 percent of the variance in the FC of the grains, while the remaining 74.3 percent might be attributed to a variety of other variables.

Table 5 revealed a significant degree of connection between bulk density and R value, with a R value of 0.738. The R2 value of 0.545 indicated that the Ochratoxins had a little above-average impact on the bulk density of the grains, which was slightly above the average.

Amount of water that can be absorbed (Water Absorption Capacity (WAC)): The R value in Table 5 showed the degree of connection between the ochratoxin levels and the water absorption capacity (WAC) of the grain flours. A correlation coefficient of 0.682 showed a moderately high degree of connection. The R2 value indicated that the Ochratoxins were responsible for a significant portion of the overall variance in the WAC of the flours. Having an R2 of 0.465 indicated that the Ochratoxins had a little to moderately positive impact on the WAC of the grains in the experiment. As a result of the synthesis of Ochratoxins, the WAC of the grains changed by 46.5 percent as a result of the process.

Grain flour emulsion capacity (EC): The degree of connection between the Ochratoxins and the emulsion capacity of the grain flours is shown in Table 5. The correlation coefficient (R) was 0.498, indicating a moderate degree of correlation, while the R2 value of 0.248 showed that the impact of the Ochratoxins on the EC of the flours was very low. In other words, the presence of Ochratoxins produced a 24.8 percent variance in the EC of the flours, with the remaining 74.8 percent variation perhaps attributable to other reasons.

Conclusion

A class of poisonous secondary metabolic products generated by microorganisms of the fungal kingdom, namely Penicillium and Aspergillus species, ochratoxins are toxic secondary metabolic products. This group of Ochratoxins is frequently found in grains before to, during, and after harvest time. The total ochratoxin levels detected in the grains varied from 0.09 to 54.41 micrograms per kilogram of grain, with the majority of them above the WHO/EU permitted limit of 5.00 micrograms per kilogram. Consumption of these grains, if they are poorly processed, may increase the chance of developing liver cancer, urinary tract cancer, and renal disease. As a result of the presence of Ochratoxin, the nutritional and functional characteristics of grains were altered to varying degrees of importance, according to the findings. Some rice samples were found to be devoid of ochratoxin. It was discovered that the presence of the Ochratoxin had a substantial impact on the percentages of carbs, proteins, and lipids found in the grains samples tested. In the case of grains, it has a moderate effect on the percentage of crude fiber and percent ash present, and a little impact on the percent moisture content of the grains present. Using the results, it was discovered that the presence of the Ochratoxin had a modest impact on the functional characteristics of the grains (bulk density; SI; WAC; FC; EC; OAC).

Competing Interest

The authors declare no conflict of interest.

Authors' Contributions

Professor Owuamanam, I. C., and Professor Ogueke, C. C. were the principal investigators, directed the sampling and analytical methods used for the research, among other responsibilities. Chinaza G. Awuchi carried out the study, did the statistical analyses, and prepared the reports and manuscript under the guidance and direction of Professor Owuamanam, I. C., and Professor Ogueke, C. C. All authors approve this version for submission.

Acknowledgment

All thanks go to Dr. Thompson E. Nwankwere, and Dr. Ebere Udeogu for their profound contributions during the research

References

- Adebayo-Oyetoro, A. O., Olatidoye, O. P., Ogundipe, F. O., Ogundipe, O. O., Balogun, O. I., Bamidele, F. A., & Faboya, A. O. (2011). Evaluation of proximate composition and functional properties of Ofada rice (Oryza sativa) flour blended with bambara groundnut (Vigna subterranean L). *Journal of Agriculture and Veterinary Sciences*, 3, 60-66.
- Adebowale, K. O., & Lawal, O. S. (2004). Comparative study of the functional properties of bambarra groundnut (Voandzeia subterranean), jack bean (Canavalia ensiformis) and mucuna bean (Mucuna pruriens) flours. *Food Research International*, 37(4), 355-365.
- Ahaotu, N.N., Bede, N.E., Umejesi, T.J., Echeta, C.K., & Awuchi, C.G. (2020a). Studies on the Functional Properties of Malted Soy-garri as affected by Moisture Variation during Storage. *European Academic Research*, 8 (8): 4616-4625.
- AOAC (2000). Ochratoxin in Roasted Coffee Immunoaffinity Column HPLC Method First Action 2000. The Association of Official Analytical Chemist, Washington D.C., USA
- AOAC (2005). Method of analysis of the AOAC, 15th ed. Washington D.C., USA.
- Ashiq, S. (2015). Natural occurrence of mycotoxins in food & feed: Pakistan perspective. *Comprehensive Reviews in Food Sci & Food Safety*, 14, 159–175.10.1111/crf3.2015.14.issue-2.
- Awuchi, C.G. & Amagwula, I.O. (2021). Biochemistry and Nutrition of Carbohydrates. *Global Journal of Research in Agriculture and Life Sciences*, 1(1): 4-12.
- Awuchi, C.G., Amagwula, I.O., Priya, P., Kumar, R., Yezdani, U., Khan, & M.G. (2020a). Aflatoxins In Foods And Feeds: A Review On Health Implications, Detection, And Control. Bulletin of Environment, Pharmacology, and Life Sciences, 9(9): 149-155
- Awuchi, C.G., Igwe, V.S., & Amagwula, I.O. (2020d). Nutritional Diseases and Nutrient Toxicities: A Systematic Review of the Diets and Nutrition for Prevention and Treatment. *International Journal of Advanced Academic Research*, 6 (1); 1 – 46.
- Awuchi, C.G., Igwe, V.S., & Echeta, C.K. (2019). The Functional Properties of Foods and Flours. *International Journal of Advanced Academic Research*, 5 (11); 139 160.

- Awuchi, C.G., Igwe, V.S., Amagwula, I.O., & Echeta, C.K. (2020e). Health Benefits of Micronutrients (Vitamins and Minerals) and their Associated Deficiency Diseases: A Systematic Review. *International Journal of Food Sciences*, 3(1): 1 – 32.
- Awuchi, C.G., Ondari, E.N., Ogbonna, C.U., Upadhyay, A.K., Baran, K., Okpala, C.O.R., Korzeniowska, M., & Guiné, R.P.F. (2021a). Mycotoxins Affecting Animals, Foods, Humans and Plants: Types, Occurrence, Toxicities, Action Mechanisms, Prevention and Detoxification Strategies—A Revisit. *Foods*, 10, 1279. https://doi.org/10.3390/foods10061279
- Awuchi, C.G., Owuamanam, I.C., Ogueke, C.C., & Hannington, T. (2020f). The Assessment of the Physical and Pasting Properties of Grains. *European Academic Research*, 8 (2): 1072 – 1080.
- Awuchi, C.G., Owuamanam, I.C., Ogueke, C.C., Hannington, T. (2020b). The Impacts of Mycotoxins on the Proximate Composition and Functional Properties of Grains. *European Academic Research*, 8 (2): 1024-1071.
- Awuchi, C.G., Twinomuhwezi, H., & Awuchi, C.G. (2021c). "Hyphenated Techniques." In C Egbuna, K Patrick-Iwuanyanwu, MA Shah, JC Ifemeje, A Rasul (Ed.). Analytical Techniques in Biosciences. Amsterdam, Netherlands. Elsevier. DOI: 10.1016/B978-0-12-822654-4.00015-4
- Awuchi, C.G., Twinomuhwezi, H., Igwe, V.S., Sarvarian, M., & Amagwula, I.O. (2021b). Protein Functions and Structure Prediction with I-TASSER, a Novel Technology. *Journal of Chemical, Biological and Physical Sciences*. 11(4): 555-567. DOI: 10.24214/jcbps.B.11.4.55567
- Battacone, G., Nudda, A., & Pulina, G. (2010). Effects of ochratoxin A on livestock production. *Toxins*, 2(7), 1796-1824.
- Bayman, P., & Baker, J. L. (2006). Ochratoxins: a global perspective. *Mycopathologia*, *162*(3), 215-223.
- Cavin, C., Delatour, T., Marin-Kuan, M., Holzhäuser, D., Higgins, L., Bezencon, C., ... & Schilter, B. (2007). Reduction in antioxidant defenses may contribute to ochratoxin A toxicity and carcinogenicity. *Toxicological Sciences*, 96(1), 30-39.
- Chinaza, G.A. (2019). Proximate Composition and Functional Properties of Different Grain Flour Composites for Industrial Applications. *International Journal of Food Sciences*, 2(1); 43 – 64.
- Chinaza, G.A., Chinelo, K.E., & Victory, S.I. (2020a). Diabetes and the Nutrition and Diets for Its Prevention and Treatment: A Systematic Review and Dietetic Perspective. *Health Sciences Research*. 6 (1): 5-19.
- Chinaza, G.A., Clifford, I.O., Chika, C.O., & Victory, S.I. (2019). Evaluation of Patulin Levels and impacts on the Physical Characteristics of Grains. *International Journal of Advanced Academic Research*, 5 (4); 10 – 25.
- Chinaza, G.A., Erick, N.O., Chigozie, E.O., James, S.C., Waheed, A.R., Sonia, M., & Charles, O.R.O. (2021b). Grain Processing Methods' Effectiveness to Eliminate Mycotoxins: An Overview. Asian Journal of Chemistry, 33(11): 1-9
- Chinaza, G.A., Erick, N.O., Hannington, T., Victory S.I., & Ikechukwu, O.A. (2021a). Aflatoxin B1 Production, Toxicity, Mechanism of Carcinogenicity, Risk

Management, and Regulations. Archives of Animal and Poultry Sciences. 1(4): 555568.

- Chinaza, G.A., Igwe, V.S., & Amagwula, I.O. (2020b). Ready-to-Use Therapeutic Foods (RUTFs) for Remedying Malnutrition and Preventable Nutritional Diseases. *International Journal of Advanced Academic Research*, 6 (1); 47 81.
- Chinaza, G.A., Ikechukwu, O.A., Victory, S.I., & Allan, I.G. (2018). Effects of Repeated Deep Frying on Refractive Index and Peroxide Value of Selected Vegetable Oils. *International Journal of Advanced Academic Research*, 4 (4); 106 – 119.
- Fennama, R. O. (1996). *Food Chemistry* (3rd eds) Basel, Hong Kong: Marcel Dekker Inc. New York. Pp. 36 39.
- Fratamico, P., Arun, K.B., & Smith, J.L. (2008). *Foodborne Pathogens: Microbiology and Molecular Biology*. Horizon Scientific Press. ISBN 978-1-898486-52-7.
- Iwe, M., & Onadipe, O. (2001). Effect of the addition of extruded full-fat soy flour into sweet potato flour on the functional properties of the mixture. *Journal of Sustainable Agriculture and the Environment*, 3, 109–117.
- Iwe, M.O., & Onuh, J.O. (1992). Functional and sensory properties of soyabean and sweet potato flour mixtures. *Lebensmittelwissenschaft und Technologie*, 25, 569–573
- Iwe, M.O., Onyeukwu, U., & Agiriga, A. (2016). Proximate, functional & pasting properties of FARO 44 rice, African yam bean, and brown cowpea seeds composite flour. *Cogent Food and Agriculture*. 2: 1142409.
- Jeswal, P. & Kumar, D.H. (2015). "Mycobiota & Natural Incidence of Ochratoxin A, Aflatoxins, and Citrinin in Indian Spices Confirmed by the LC-MS/MS." *International Journal of Microbiology*. 2015: 242486. doi: 10.1155/ 2015/242486.
- Jitngarmkusol, S., Hongsuwankul, J., & Tananuwong, K. (2008). Chemical composition, functional properties & microstructure of defatted macademic flours. Food Chemistry. 2008; 110: 23–30. doi: 10.1016/j.foodchem.2008.01.050.
- Kaushal, P., Kumar, V., & Sharma, H.K. (2012). Comparative study of physicochemical, functional, and anti-nutritional and pasting properties of taro (*Colocasia esculenta*), rice (*Oryza sativa*), pigeon pea (*Cajanus cajan*) flour & their blends. *LWT-Food Science and Technology*. 48: 59-68.
- Lorenz, K., % Collins, F. (1980). Quinoa, starch physicochemical properties and the functional characteristics of starch, *Wiley*, 42, 81–86.
- Mateo, R., Medina, A., Mateo, E.M., Mateo, F., & Jiménez, M. (2007). An overview of ochratoxin A in beer and wine. *International Journal of Food Microbiology*. 20;119(1-2):79-83. doi: 10.1016/j.ijfoodmicro.2007.07.029.
- Niemiec, J. & Borzemska, W. (1994). "The effect of Ochratoxin A on the egg quality development of embryos and level of toxin in egg and the tissues of hens and chicks." *Journal of Animal and Feed Sciences*. 3 (4): 309–316.
- Onwuka, G. I. (2005). Functional properties in Food analysis and instrumentation. Naphtali Prints, Lagos. Pp 134-135.
- Richard, J. (2007). Some major mycotoxins and their mycotoxicoses An overview. International Journal of Food Microbiology, 119, 3-10.

- Robbins, C.A, Swenson L.J., Nealley M.L., Gots R.E., & Kelman, B.J. (2000). Health effects of mycotoxins in indoor air: a critical review. *Applied Occupational and Environmental Hygiene*. 15 (10): 77384.
- Suresh, C., Samsher, S., & Durvesh, K. (2014). Evaluation of functional properties of composite flours and sensorial attributes of composite flour biscuits. *Journal of Food Science and Technology*, 52(6): 3681–3688.
- Tufail, T., Ijaz, A., Noreen, S., Arshad, M.U., Gilani, S.A., Bashir, S., Din, A., Shahid, M.Z., Khan, A.A., Khalil, A.A., & Awuchi, C.G. (2021). Pathophysiology of Obesity and Diabetes. In: Egbuna C., Hassan S. (Eds.). *Dietary Phytochemicals* (pp. 29–42). Springer, Cham. https://doi.org/10.1007/978-3-030-72999-8_2
- Turner, N., Subrahma, S, & Piletsky, S. (2009). Analytical methods for the determination of mycotoxins: a review. *Analytica Chimica Acta*. 632 (2): 168–80.
- Twinomuhwezi, H., Awuchi, C.G., Mihigo, R. (2020). Comparative Study of the Proximate Composition and Functional Properties of Composite Flours of Amaranth, Rice, Millet, and Soybean. American Journal of Food Science and Nutrition. 6(1): 6-19.
- Udeogu, E. & Awuchi, C. G. (2016). Effects of some processing methods on hemagglutinin activity of lectin extracts from selected grains (Cereals and Legumes). IJAAR, 2 (12): 25.
- Yasmin, I., Khan, W.A., Naz, S., Iqbal, M.W., Awuchi, C.G., Egbuna, C., Hassan, S., Patrick-Iwuanyanwu, K.C., & Uche, C.Z. (2021). Etiology of Obesity, Cancer, and Diabetes. In: Egbuna C., Hassan S. (Eds.). *Dietary Phytochemicals* (pp. 1–27). Springer, Cham. https://doi.org/10.1007/978-3-030-72999-8_1