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Influence of Traditional Food Processing Systems on Food Safety, Chemical Compositions, and Functional Properties of Pumpkin (Cucurbita pepo L) Seed Flour

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ABSTRACT

Background: Plant foods contain almost all of the essential mineral and organic nutrients for human, as well as several unique organic phytochemicals that have been linked to the promotion of good health. This study investigated the influence of traditional food processing systems on the food safety, chemical compositions, and functional properties of pumpkin (*Cucurbita pepo L*) seed flour. **Methods:** Fresh mature pumpkin fruits were procured from an open market and taken to the laboratory. The pumpkin seed was divided into three different portions and subjected to traditional sand toasting, germination, and then processed into flour. The samples were analyzed for proximate composition, minerals, heavy metals, vitamins, functional properties, and microbial safety. **Results:** The result of proximate composition revealed that raw and traditionally processed pumpkin seed flour had 33.75-39.63% protein, 43.65-36.36% fat, 7.38-5.89% moisture, 4.85-6.04% ash, 1.89-2.25% fiber, and 5.94-14.49% carbohydrate. The mineral and heavy metal content were in the range of 7.17-4.13 mg/100 Iron, 117.88-221.55 mg/100 g Potassium, 7.41-9.01 mg/100 g Calcium, 45.67-55.65 mg/100 g Phosphorus, 0.89-4.67 mg/100 g Iodine, and 7.39-6.33 mg/100 g Zinc. The toxic/deleterious elements values for Hg, Pb, As, and Cd were 0.018-0.004 mg/100 g; 0.021–0.062 mg/100 g, 0.013–0.092 mg/100 g, and 0.0011-0.0051 mg/100 g, respectively. Cyanide, Nitrate, Phytate, and Oxalate contents of the pumpkin seed flour were 0.72-0.32 mg/100 g, 2.77–1.00 mg/100 g, 35.56–14.56 mg/100 g, and 0.073–0.030 mg/100 g, respectively. An appreciable amount of vitamins (Beta-carotene, vitamin C, and thiamine) were recorded with excellent functional properties. The microbial count was within the limit of national and international standards showing the processed pumpkin seed flour to be safe. **Conclusion:** It can be concluded that subjecting pumpkin seeds to traditional processing methods enhanced their nutritional properties and food safety quality. Therefore, adopting this traditional processing approach can be used in rural community nutrition and nutrition in emergency feeding programs in developing countries.

Keywords: *Germination; Toasting; Traditional food system; Anti-nutritional factors; Food safety*

Introduction

Plant foods contain almost all of the mineral and organic nutrients established as essential for human nutrition, as well as several unique organic phytochemicals that have been linked to

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the promotion of good health. This has led to a considerable increase in the demand for new nutritionally healthy, cost-effective, and sustainable viable foods of plant origin. "The use of these raw materials adds value to economic production, contributes to the formulation of new food products, and minimizes waste" (Lemus-Mondaca *et al.*, 2019). Pumpkin (*Cucurbita pepo L*) is botanically defined as a fruit although commonly regarded as a vegetable in consumer terms. *Cucurbita Pepo* is one of the underutilized crops belonging to the family *Cucurbitaceae*. Its existence is presently threatened due to neglect in Nigeria (Blessing *et al.*, 2011). Pumpkin is cultivated in Nigeria at a subsistence level with virtually no commercial importance. In Nigeria, it is a traditional vegetable crop, grown mainly for its leaves, fruits, and seeds and, consumed either by boiling the leaves and fruits or by roasting or baking the seeds. Flesh and the seeds of pumpkin are commonly used for culinary and medicinal purposes (Azizah *et al.*, 2009). When dried, seeds can be used as a thickener for soups and as snacks.

Pumpkin seeds are characterized by high levels of protein and oil. Several beneficial effects to human health have been attributed to pumpkin seeds, due to their macro and micronutrient content. Pumpkin seeds are a natural source of phytosterols and antioxidants vitamins such as tocopherols and carotenoids (Lemus-Mondaca *et al.*, 2019, Ryan *et al.*, 2007) and an excellent source of unsaturated fatty acids such as oleic and linoleic. These compounds are attributed to have physiological activity beneficial to the prostate and others such as being anti-parasitic for the intestine, thereby making pumpkin seeds natural functional food.

Plant protein and seed flours from legumes and oilseeds serve as a functional ingredient in the food system especially in developing countries where access to first-class protein and wheat flour is limited. However, most of the oilseeds and legumes used as protein and flour substitutes contain inherent inhibiting compounds and toxicants that compromise their food quality and safety. These intrinsic factors equally hinder their further utilization and contribution to food security, nutritional/diets diversity, and economic development. Various foods presuppose different processing techniques depending on the needs and end products required (Mbah *et al.*, 2012). Traditional food processing systems such as germination, fermentation, roasting/toasting, and soaking have been established to have the potential of reducing anti-nutrients, toxicant, improving the nutritional quality, and enhancing digestibility and bioavailability of essential micro and macronutrients (Adekanmi *et al.*, 2009, Adeoti *et al.*, 2017, Akintade *et al.*, 2019, Amadi *et al.*, 2019, Fagbemi, 2007, Fagbemi *et al.*, 2005, Maria and Hannah, 2019).

Sand toasting or roasting is one of the most convenient, simplest, and oldest traditional food processing techniques employed for legumes, oilseeds, and cereal processing. Traditionally, sand toasting of legumes and oilseeds are typically carried using sand placed in a pot over an open fire and the seeds are toasted by frequent stirring until the kernel is golden or caramel. During toasting, the far-infrared rays produced from the sand penetrate the grains/oilseed and aid in breaking down the starch, protein, and fats in the grains. It also enhances the color, aroma, flavor, shelf life, and consumer acceptance and reduces the bulk density (BD) and antinutrients present in cereals and legumes (Kora, 2019).

It is known that germination induces an increase in free limiting amino acids and available vitamins with modified functional properties of seed components. In addition, germination has been shown to improve the vitamins and protein quality and reduce flatulence and anti-nutritional factors in legumes, resulting in improved digestibility and utilization (Fagbemi *et al.*, 2005, Uwaegbute *et al.*, 2000).

The knowledge of how processing conditions modify the bioavailability of nutrients trapped in the food matrix and affect food safety, quality, and functional performance is indeed a critical factor to be considered in the development of food for various age groups in addition to the cost of raw ingredients, processing cost, and sensory

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acceptability of such foods (Abebe *et al.*, 2006, Maria and Hannah, 2019). This study contributes to the existing knowledge of pumpkin (*Cucurbita pepo L*) seed nutrition, food safety, and functionality and reports the influence of traditional sand toasting and germination processes on the nutritional quality, food safety, chemical compositions, and functional properties of pumpkin (*Cucurbita pepo L*) seed flour (Adekanmi *et al.*, 2009, Adeoti *et al.*, 2017, Akintade *et al.*, 2019, Amadi *et al.*, 2019, Fagbemi, 2007, Fagbemi *et al.*, 2005, Kora, 2019, Maria and Hannah, 2019, Mbah *et al.*, 2012).

Materials and Methods

Sources of materials: The fresh mature pumpkin fruits were procured from open market in Lafia Metropolis, Nigeria, and taken to the laboratory for processing using traditional methods.

Preparation of pumpkin seed flours: The pumpkin fruits were cut open using a sharp knife and the seeds were manually separated. The seeds were picked, washed, drained, and divided into three portions. The first portion was raw served as the control. The second and third portions were subjected to traditional sand toasting and germination and were processed into flour. The control seeds were milled raw without any treatment. The germinated, sand toasted, and raw flours were stored in labeled polythene bags in a cool dry place until used for various analysis.

Preparation of raw pumpkin seed flour: The washed and drained raw pumpkin seeds were sun-dried for 12 h on a raised platform to avoid dust and other debris contamination. The dried seeds were dehulled manually and then milled and sieved using a local mash to obtain fine flour. The fine pumpkin seed flour was packaged and stored in air-tight polythene Ziploc bags for further analysis.

Preparation of germinated *pumpkin seed flour:* Germination was carried out as shown in **Figure 1**. One kilogram of raw pumpkin seeds was washed in a 5% (w/v) sodium chloride (NaCl) solution to disinfect the seeds. The seeds were then soaked in tap water at room temperature using a ratio of 1:3 (w/v seed: water), in a plastic bucket for a total steeping time of 12 hours. The seeds were drained in a plastic basket and spread in a single layer on a moistened jute bag and allowed to germinate at room temperature for 72 hours. The germinated seeds were removed at 72 hours and sun-dried on a raised platform to avoid dust and other debris. The dried seeds were dehulled manually, winnowed, milled, and then sieved using a local mash to obtain fine flour. The fine germinated pumpkin seed flour was packaged in polythene Ziploc bags and stored in air-tight plastic containers and utilized for further analysis within 24 h.

Preparation of sand toasted pumpkin seed flour: The method is described and shown in **Figure 2**. The washed and drained raw pumpkin seeds were sun-dried for 12 h on a raised platform to avoid dust and other debris contamination. The dried seeds were traditionally toasted on a charcoal stove in a saucepan with hot fine sand for 10 min. The sand toasted pumpkin seeds were dehulled manually and winnowed to remove the hulls. The dehulled seeds were milled and sieved using a local mash to obtain fine flour. The fine sand toasted pumpkin seed flour was packaged in polythene Ziploc bags and stored in air-tight plastic containers and utilized for further analysis within 24 h.

Proximate analysis of raw and processed pumpkin seed flour: The proximate content of processed pumpkin seed flour was determined according to the Association of Official Analytical Chemists (Association of Official Analytical Chemists, 2012).

Determination of mineral content of raw and processed pumpkin seed flour: The mineral content of the samples was determined using energy dispersive X-ray fluorescence spectroscopy according to (Jenkins, 2000).

Determination of functional properties of raw and processed pumpkin seed flour: The bulk density (BD), foaming capacity (FS), foaming stability (FS), least gelation (LG), oil absorption capacity (OAC), and water absorption capacity (WAC) of processed pumpkin seed flour were

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determined according to Fagbemi method (Fagbemi *et al.*, 2005), while swelling index (SI) was carried out using the method described by the Ukpabi *et al.* (Ukpabi and Ndimele, 1990).

Determination of antinutritional factors of raw and processed pumpkin seed flour: Analysis of oxalate, phytate, hydrocyanic acid, and nitrate were carried by adopting methods described by AOAC (Association of Official Analytical Chemists, 2012).

Determination of vitamin content of raw and processed pumpkin seed flour: Vitamin A, C, and thiamin in the samples were determined using the method described by AOAC (Association of Official Analytical Chemists, 2012) using high-performance liquid chromatography.

Microbial analysis: Samples were analyzed microbiologically using standard methods described by Cheesbrough (Cheesbrough, 2005). The total coliform count was determined by the MPN index method using a 3-3-3 regimen. Colony count was done using a digital colony counter for bacteria and a hand lens for fungi. The total colony was expressed as colony-forming units in milliliters (CFU/ml).

Data analysis: All the results were obtained in triplicate, subjected to analysis of variance ANOVA and mean values were separated by New Duncan Multiple Range Test (NDMRT). Means and standard deviations of all the samples were calculated and compared. SPSS for Windows program version 21.0 was used to analyze the obtained results.

Results

Influence of traditional food processing systems (germination and sand toasting) on the proximate composition of pumpkin seed flour: The influence of traditional food processing systems (germination and toasting) on the proximate composition of pumpkin seed flour is presented in **Table 1**. The crude protein content ranged from 33.75 to 39.63%. The fat ranged from 43.65 to 36.36%. Moisture content ranged from 7.38 to 5.89%. Ash content ranged from 4.85 to 6.04%. The fiber content ranged from 1.89 to 2.25% with carbohydrate content ranging from 5.94 to 14.49%. Mean ± SD values of triplicate determinations with different superscripts on the same row are significantly different at $P < 0.05$.

Influence of traditional food processing systems (germination and sand toasting) on the mineral and heavy metal contents of pumpkin seed flour: Nutritionally viable minerals and deleterious heavy metals contents of raw, germinated, and sand toasted pumpkin seed flour are presented in **Table 2**. Germination and sand toasting significantly ($P < 0.05$) increased mineral content with Iron (7.17-4.13 mg/100 g), Potassium (117.88-221.55 mg/100 g), Calcium (7.41-9.01 mg/100 g), Phosphorus (45.67-49.99 mg/100 g), Iodine (0.89-4.67 mg/100 g), and Zinc (7.39-6.49 mg/100 g). The toxic/deleterious elements significantly reduced with values for Mercury (Hg), Lead (Pb), Arsenic (As), and Cadmium (Cd) ranging 0.018–0.004 mg/100 g, 0.021–0.062 mg/100 g, 0.013–0.092 mg/100 g, and 0.0011-0.0051 mg/100 g, respectively. Mean±SD values of triplicate determinations with different superscripts on the same row are significantly different at $P < 0.05$.

Influence of traditional food processing systems (germination and sand toasting) on the anti-nutritional factors of pumpkin seed flour: The anti-nutritional factors in the raw and treated flour samples are presented in **Table 3**. Cyanide, Nitrate, Phytate, and Oxalate contents ranged 0.72-0.32 mg/100 g, 2.77-1.00 mg/100 g, 35.56-14.56 mg/100 g, and 0.073-0.030 mg/100 g, respectively. Mean ± SD values of triplicate determinations with different superscripts on the same row are significantly different at $P < 0.05$.

Influence of traditional food processing systems (germination and sand toasting) on the vitamin content of flour: The effect of germination and sand toasting on the vitamin content of flour samples is presented in **Table 4**. The values of vitamin C, beta carotene, and thiamin ranged 1.37-2.06 mg/100 g, 127.37-272.64 µg/g, and 0.055-0.087 mg/100 g, respectively. Mean ± SD values of triplicate determinations with different superscripts on the same row are significantly different at $P < 0.05$.

Influence of traditional food processing systems (germination and sand toasting) on the functional properties of pumpkin seed flour: **Table 5** shows the functional properties of traditionally processed pumpkin seed flour. The values of OAC, WAC, OAC, and packed bulk density (PBD) ranged 98.74-118.69%, 138.22-177.82%, 1.34-1.75 g/ml, 1.68-1.7 g/ml, respectively. The values of loose bulk density (LBD), foam stability, least gelation, and foam capacity ranged from 1.41 to 1.46 g/ml, 9.39 to 11.09%, 47.72 to 64.38%, and 24.02 to 29.83%, respectively. Mean ± SD values of triplicate determinations with different superscripts on the same row are significantly different at $P < 0.05$.

Microbiological profile of sand toasted and germinated pumpkin seed flour: The microbial safety quality of the processed pumpkin seed flour is shown in **Table 6**. The total viable and fungi count ranged from 1.46×10^3 to 4.20×10^3 CFU/g and 1.76×10^2 to 4.97×10^2, respectively, with sand toasted flour having the least value.

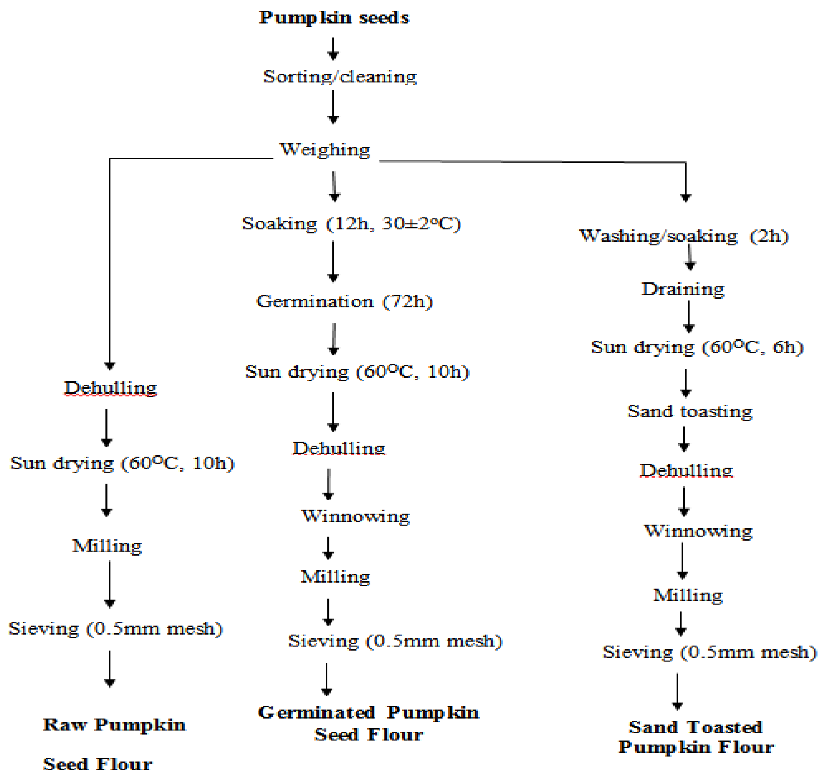

Figure 1. Flow chart for the production of pumpkin seed flour.

Table 1. Influence of traditional food processing systems (germination and sand toasting) on proximate composition of pumpkin seed flour (%).

Type of flour	Protein	Fat	Fiber	Ash	Moisture	Carbohydrate
Raw	36.27 ± 2.03[b]	43.65 ± 3.83[a]	1.89 ± 0.62[b]	4.85 ± 0.15[b]	7.38 ± 1.31[a]	5.94 ± 0.27[c]
Toasted	33.75 ± 0.74[c]	37.84 ± 0.84[b]	2.06 ± 0.44[a]	5.49 ± 0.86[ab]	6.36 ± 0.31[b]	14.49 ± 0.10[a]
Germinated	39.63 ± 0.62[a]	36.36 ± 5.55[c]	2.25 ± 0.91[a]	6.04 ± 0.46[a]	5.89 ± 0.51[b]	9.82 ± 1.67[b]

Values are Mean ± SD, values followed by the same alphabets in a row are not significantly different but those followed by different alphabets are significantly different $P < 0.05$.

Table 2. Influence of traditional food processing systems (germination and sand toasting) on the mineral and heavy

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metal contents of pumpkin seed flour (mg/100 g).

Type of flour	Fe	Ca	I	P	Zn
Raw	4.13±2.67[c]	7.41±2.43[c]	3.51±0.15[b]	49.99±0.23[b]	6.33±0.002[b]
Toasted	7.17±0.01[a]	9.01±0.10[a]	0.89 ±0.01[c]	45.67±0.03[c]	6.49±0.005[b]
Germinated	6.92±0.06[b]	8.25±0.16[b]	4.67±0.50[a]	55.65±0.009[a]	7.39±0.007[a]
	K	**Hg**	**Pb**	**As**	**Cd**
Raw	182.40±0.004[b]	0.018±0.015[a]	0.062±0.007[a]	0.092±0.001[a]	0.0051±0.007[a]
Toasted	117.88±0.008[c]	0.004±0.002[b]	0.021±0.003[b]	0.013±0.003[b]	0.0011±0.020[b]
Germinated	221.55±0.009[a]	0.003±0.005[b]	0.032±0.006[b]	0.027±0.005[b]	0.0031±0.030[b]

Values are Mean ± SD, values followed by the same alphabets in a row are not significantly different but those followed by different alphabets are significantly different $P < 0.05$.

Table 3. Influence of traditional food processing systems (germination and sand toasting) on the anti-nutritional factors of pumpkin seed flour (mg/100 g dry weight).

Type of flour	Cyanide	Nitrate	Phytate	Oxalate
Raw	0.72±0.03[a]	2.77±0.01[a]	35.56±1.00[a]	0.073± 0.03[a]
Toasted	0.25±0.03[b]	1.00±0.01[c]	14.56±1.10[c]	0.030± 0.03[b]
Germinated	0.32±0.05[b]	1.39±0.02[b]	17.56±1.20[b]	0.037± 0.04[b]

Values are Mean ± SD, values followed by the same alphabets in a row are not significantly different but those followed by different alphabets are significantly different $P < 0.05$.

Table 4. Influence of traditional food processing systems (germination and sand toasting) on the vitamin content of pumpkin seed flour.

Type of flour	Beta Carotene, µg/g	Thiamin, mg/100 g	Vitamin C, mg/100 g
Raw	241.52±0.02[b]	0.083±0.04[b]	2.06±0.03[a]
Toasted	127.37±0.03[c]	0.055±0.03[c]	1.37±0.02[c]
Germinated	272.64±0.01[a]	0.087±0.02[a]	1.87±0.03[b]

Values are Mean ± SD, values followed by the same alphabets in a row are not significantly different but those followed by different alphabets are significantly different $P < 0.05$.

Table 5. Influence of traditional food processing systems (germination and sand toasting) on the functional properties of pumpkin seed flour.

Type of flour	OAC (%)	WAC (%)	SI (g/ml)	PBD (g/ml)
Raw	98.74±0.07[c]	138.22±0.01[c]	1.57±0.06[ab]	1.70±0.03[a]
Toasted	118.69±0.04[a]	177.82±0.03[a]	1.34±0.08[b]	1.68±0.04[a]
Germinated	110.25±0.03[b]	167.36±0.02[b]	1.75±0.03[a]	1.69±0.05[a]
	LBD (g/ml)	**FS (%)**	**LG (%)**	**FC (%)**
Raw	1.46±0.09[a]	9.39±0.06[a]	61.05±0.07[b]	26.07±0.04[a]
Toasted	1.41±0.13[a]	9.37±0.15[a]	47.72±0.05[c]	24.02±0.03[a]
Germinated	1.45±0.05[a]	11.09±0.07[a]	64.38±0.06[a]	29.83±0.05[a]

Values are Mean ± SD, values followed by the same alphabets in a row are not significantly different but those followed by different alphabets are significantly different $P < 0.05$; OAC: Oil absorption capacity, WAC: Water absorption capacity, SI: Swelling index, PBD: Packed bulk density, LBD: Loose bulk density, FC: Foaming capacity, LG: Least gelation, FS: Foaming stability.

Table 6. Microbiological profile of sand toasted and germinated pumpkin seed flour (CFU/g).

Type of flour	Total viable count	Total fungi count
Raw	$4.20x10^{3} \pm 0.43^{a}$	$4.97x10^{2} \pm 1.23^{a}$
Toasted	$1.46x10^{3} \pm 1.25^{c}$	$1.76x10^{2} \pm 0. 67^{c}$
Germinated	$4.06x10^{3} \pm 0.45^{b}$	$4.85x10^{2} \pm 2.60^{b}$

Values are Mean ± SD, values followed by the same alphabets in a row are not significantly different but those followed by different alphabets are significantly different *P* < 0.05.

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Discussion

Influence of traditional food processing systems (germination and sand toasting) on the proximate composition of pumpkin seed flour: There were significant differences in protein content among the pumpkin seed flour samples. The protein values agreed with the findings of Fagbemi study (Fagbemi, 2007). Hence, *Cucurbita pepo* seed is an excellent source of protein that could be used to combat protein-energy malnutrition (PEM) in developing countries. The increase in protein content of germinated pumpkin seed flour may be attributed to protein formation through the liberation of bound proteins as a result of hydrolytic enzymes during germination. This result is consistent with the work of Enujiugha study (Enujiugha *et al.*, 2003). The decrease in protein content with sand toasting may be attributed to the Maillard reaction, which is an interaction between the carbonyl group of a reducing sugar and the free amino acid or protein (Maria and Hannah, 2019).

The fat content in this study decreased with all processing method applied. The decrease in fat content in the roasted and germinated sample might be attributed to the increased activities of lipolytic enzymes during toasting and germination, which hydrolyzes fat components into fatty acids and glycerol. This was contrary to the report of the other study (Maria and Hannah, 2019) but agreed with the findings of other one study (Akintade *et al.*, 2019) for fermented, germinated, and roasted pumpkin seed. The decrease in fat content obtained in this study might be of advantage during the storage of flour samples, as low lipid levels are known to increase the shelf life of a product. This is because rancidity will be retarded for shelf stability. Also, weight-conscious people will find sand toasted and germinated pumpkin seed flour a healthy option for calories.

The ash content was significantly in sand toasted and germinated pumpkin seed flour when compared to the raw sample. This could be attributed to the concentration of organic compounds during roasting. This result is consistent with the finding of Maria *et al.* study (Maria and Hannah, 2019) but differs from the report of other study (Akintade *et al.*, 2019). The lowest carbohydrates recorded in the raw samples was not in line with the study of Maria *et al.* (Maria and Hannah, 2019) who reported a decrease in carbohydrates by all processing techniques employed. However, it is consistent with the finding of Akintade study (Akintade *et al.*, 2019) for pumpkin seed flour. The variation in the carbohydrate content may be due to alterations in other components (protein, fat, ash fiber, and moisture). The observed decreases in carbohydrate and oil contents with germination could be attributed to their utilization as energy sources during the germination process.

Influence of traditional food processing systems (germination and sand toasting) on the mineral and heavy metal content of pumpkin seed flour: The traditional food processing systems of sand toasting and germination increased all the selected minerals. However, the raw pumpkin seed flour recorded higher in Iodine, Phosphorus, and Potassium compared to sand toasted pumpkin seed flour. This was not in line with the study by Akintade (Akintade *et al.*, 2019). Minerals were observed to be higher in germinated pumpkin seed flour than sand toasted pumpkin seed flour. This may be due to the decomposition of anti-nutrient

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like phytate, thereby releasing the bound nutrient which also led to improved ash content. The study was consistent with the other studies (Adeoti *et al.*, 2017, Amadi *et al.*, 2019, Ihemeje *et al.*, 2015, Maria and Hannah, 2019). Iron and Calcium were significantly higher in sand toasted seed flour when compared to other treatments. The increase in Iron content might be that the hydrolytic enzymes released more free Iron from its organic complexes. This study was consistent with two studies (Amadi *et al.*, 2019, Nnorom *et al.*, 2015), but inconsistent with Akintade (Akintade *et al.*, 2019) that reported a decrease in Calcium and Iron after roasting. This may be due to differences in processing methods. Akintade employed boiling of pumpkin seed with salt before roasting, in which leaching of solid matter may occur. Considering the nutritional beneficial micronutrients (Fe, Ca, I, P, Zn, and K); seeds are a fairly source of the minerals and demonstrate the potential of the seed in resolving hidden hunger. Most heavy metals are toxic to humans, even at low concentrations. Hg, Pb, As, and Cd concentrations were relatively lower, showing seeds are generally low in toxic heavy metals. A similar result was obtained by Fagbemi study (Fagbemi, 2007).

Influence of traditional food processing systems (germination and sand toasting) on the anti-nutritional factors of seed flour: Toasting was observed to be more effective in the deactivation of anti-nutrients than germination, as phytate and oxalate are heat-labile. This is not in line with the report of Maria study (Maria and Hannah, 2019) who observed germination to be more efficient. The presence of anti-nutrients in human diets affects nutrient absorption in infants and adults (Maria and Hannah, 2019). Toasting and germination have been effective in deactivating nutrients, making pumpkin seed flour safe for both infant and adult food formulations. This result is consistent with the findings of the other study (Fagbemi *et al.*, 2005). Maria stated that "bioavailability of nutrients in processed food products could be enhanced when they contain a minimal amount of residual anti-nutritional factors. This is of great concern in community nutrition and emergency feeding programs in developing countries where the consequences of anti-nutritional factors may worsen incidences of malnutrition and disease among infants and other vulnerable groups" (Maria and Hannah, 2019).

Influence of traditional food processing systems (germination and sand toasting) on the vitamin content of pumpkin flour: Vitamin A (Beta Carotene) and thiamin contents were significantly higher in germinated pumpkin seed flour than sand toasted and raw seed flours. This is contrary to the findings of Amadi study (Amadi *et al.*, 2019) that recorded a decrease of vitamin A content in germinated breadfruit flour. The raw pumpkin seed flour had the highest value of vitamin C. The vitamin C contents decreased drastically in the roasted sample, which could be attributed to the high toasting temperature. This would be expected that this chemical compound is easily degraded with heat. The vitamin C content in germinated pumpkin seed flour was also reported in the other studies (Amadi *et al.*, 2019, Devi *et al.*, 2015, Rahman *et al.*, 2016).

Influence of traditional food processing systems (germination and sand toasting) on the functional properties of pumpkin seed flour: The traditional food processing systems had a significant effect on the functionality of the pumpkin seed flour as presented in **Table 4**. Sand toasted and germinated seed flour had the lowest least BD, which is not in line with the findings of Maria study (Maria and Hannah, 2019). The PBD was higher in raw seed flour and decreased with processing treatments. This agrees with the findings of mentioned study (Maria and Hannah, 2019) and could be attributed to a decrease in the weight of the flour as a result of the breakdown of complex denser compounds (Gernah *et al.*, 2011, Maria and Hannah, 2019). However, lower BD is desirable as it offers packaging advantages and is nutritionally important for convalescent child feeding. It increases the energy obtainable from a diet (Adepeju *et al.*, 2011, Maria and Hannah, 2019). Hence, sand toasted and germinated pumpkin seed flours may be useful in the manufacture of high

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nutrient-dense weaning foods. There was a significant increase in water and OAC with processing treatment. The polar amino acid residues of proteins or charged site chains account for the sharp difference in water absorption as reported by Jitngarmkusol study (Jitngarmkusol *et al.*, 2008). The increase in WAC indicates that sand toasted pumpkin seed flour could be used as a thickener in the food system, which is consistent with the Fagbemi study (Fagbemi *et al.*, 2005). The sand toasted pumpkin seed flour had the highest OAC in comparison to germinated and raw pumpkin seed flour. This could be due to proteins denaturation and constituent dissociation as a result of heating that exposes the interior non-polar residues of the molecules. Also, increased activity of lipolytic enzymes releasing more free fatty acids during sprouting as the result of the unfolding a polar amino acid of seed protein that encourages hydrophobicity could be responsible for the increase in OAC of germinated pumpkin seed flour (Fagbemi *et al.*, 2005, Ihemeje *et al.*, 2015, Odoemelam, 2005).

The ability of the protein to form a gel is measured using least gelation capacity (LGC). The LGC was lowest in sand toasted pumpkin seed flour which could be due to a reduction in protein as a result of the Maillard reaction. However, germination increased the gel formation capacity of the flour corresponding to increasing in protein. The low OAC in toasted pumpkin seed flour could be attributed to the dextrinization of starch to sweet-tasting molecules by dry heat during toasting. The functional properties recorded in this study are in line with the findings of the other studies (Akintade *et al.*, 2019, Giami and Bekebain, 1992, Hasmadi *et al.*, 2020, Lin *et al.*, 1974, Yasumatsu *et al.*, 1972).

Microbiological profile of sand toasted and germinated pumpkin seed flour: The pumpkin seed flours were microbiological safe for consumption and application in product development. The microbial counts were within the limit of local and international standards for microbial safety. Heat and quality attributes such as high oil content and low moisture content are factors that could be responsible for the lower microbial count observed in this study. Oil has an inhibitory effect that limits growth of aerobic microorganisms by sealing up air pores for aerobic microorganisms' respiration. Sand toasting had the least microbial count that could be attributed to the effect of heating that destroys microorganisms. The result of this study is in agreement with the findings of the others (Eke and Elechi, 2021, Ike *et al.*, 2020).

Conclusion

The present study shows traditional food processing systems of sand toasting and germination to have significant improvement in nutrient density, food safety, and functional properties. Processed pumpkin seed flour contained an appreciable amount of protein, fiber, fat, carbohydrates, minerals, and vitamins (Beta-carotene, vitamins C, and thiamine). The levels of anti-nutrients and toxic heavy metals were reduced with excellent functional properties. The microbial count was within the limit of national and international standards showing the processed pumpkin seed flour to be safe. It can be concluded that subjecting pumpkin seeds to traditional processing methods enhanced its nutritional properties and food safety quality. Therefore, adopting this traditional processing approach can be used in rural community nutrition and nutrition in emergency feeding programs in developing countries, where the consequence of heavy metals, anti-nutritional factors, and microbial contamination may worsen the incidence of malnutrition and disease.

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Conflict of interest

No conflict of interest declared.

Authors' Contribution

Elechi J.O.G designed and conducted research; Sule J.I provided essential reagents/materials necessary for the research and analyzed data; and Elechi J.O.G and Sule J.I wrote the paper. Elechi J.O.G had primary responsibility for final content. Both authors read and approved the final manuscript.

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