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Functional Properties and Heavy Metal Content of Some Swallows made from Edible Flours

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Abstract
The globally reported increase in the incidence of chronic diseases, such as diabetes, cardiovascular disease, and certain cancers has prompted the search for more diet alternatives which could be incorporated in the prevention and management of these conditions. In Nigeria, many indigenous menus include swallows, which are usually consumed alongside native soups. This study aimed at determining the functional properties and heavy metal content of some swallows made from cereal flours-finger millet (*Eleusine coracana*), black fonio (*Digitaria iburua*), and rye (*Secale cereale*) and also cassava (*Manihot esculenta*) flour. The cereal samples were purchased from local markets, sorted, washed, dried and prepared for analyses; the cassava flour was purchased as a finished product. The flours were then used to prepare swallows by pouring them into boiling water. Standard methods were used to investigate the functional properties and heavy metals present in the food samples. Laboratory results were analyzed using Microsoft Excel and Statistical Package for Social Sciences (SPSS). The results show that rye swallow had significantly ($p<0.05$) higher bulk density (0.63g/ml) than the others. Cassava flour had the highest water absorption capacity (2.24±0.06 ml/g) while black fonio flour had the lowest (1.85±0.00 ml/g). Gelatinization temperatures in degrees centigrade ranged from 64.67±0.08 (rye) to 70.58±0.04 (black fonio). The heavy metals were present in very minimal amounts, with aluminium having slightly higher values, although all the values were within safe limits. Consequently, with appropriate preparation methods, adoption of these cereal flours as swallows is recommended for improved nutrition outcomes.
Keywords: functional properties, heavy metals, swallows, cereals, cassava flour

Introduction

Modern industrialization has brought about nutrition transition both in developing and developed countries worldwide, and this has not come without some consequences on health and longevity. Thus, there is need for proper investigation of both nutritional and non-nutritional content of popularly consumed foods. Also, less popular foods such as cereals, legumes, and vegetables are being promoted to replace highly starchy foods to reduce the problem of diabetes and obesity. Nowadays, even vegetables like cabbage and carrots are now being used to prepare swallows for people with restricted diets. The increase in incidence rates of diet-related NCDs has brought about emphasis on the prevention/management of diseases using dietary intervention (Venkat-Narayan *et al.*, 2010). Furthermore, the formulation of single flours and composite flours from fibre-rich cereal grains and low glycemic index (GI) foods such as wheat, guinea corn (also known as sorghum), and oats has been proven beneficial for the prevention and management of diet-related diseases. These diseases include diabetes and hypertension (Noorfarahzilah *et al.*, 2014).

Food wholesomeness is also an aspect of food safety that must not be taken for granted. Food contains both nutrients, non-nutrient and anti-nutrient compounds which all contribute to the overall quality of the food. Some non-nutrients such as herbicides, insecticides, food additives find their way into foods, either intentionally or unintentionally. Food processing methods such as post-harvest handling, storage and food preparation techniques play a major role in reducing the concentrations of these non-nutrient constituents of food (Gandebe *et al.*, 2017). Understanding the various processing methods and their effects is essential in ensuring that edible flours meet both culinary and nutritional expectations.

Washing, a fundamental step in flour processing, serves the purpose of removing impurities, soil, and potential contaminants. Adequate washing, when performed with clean water, helps eliminate external debris and microbial contaminants, contributing to the overall safety of the flour (Ndife *et al*., 2013). Furthermore, soaking is a traditional method employed in the preparation of various flours. For instance, in the case of cassava flour, soaking cassava roots before processing helps reduce cyanogenic glycosides, enhancing the safety of the flour. Additionally, soaking can lead to nutrient enhancement by promoting the release of bioactive compounds and reducing anti-nutrients (Oladunmoye *et al*., 2018). Cooking is a prevalent method for preparing indigenous flours. Heat application during cooking promotes the gelatinization of starch, enhancing its digestibility and making essential nutrients more accessible (Chung *et al*., 2018).

Milling, a pivotal step in the processing of indigenous edible flours, holds paramount importance in determining the particle size distribution and overall quality of the final product. Traditional stone milling, a practice deeply rooted in many indigenous flour processing methods, involves grinding grains between two stones. While this method retains certain nutritional aspects, it may result in variable particle sizes, affecting the consistency and texture of the flour (Devi *et al*., 2015). Modern roller milling, an advanced technique widely adopted in commercial flour production, offers a more controlled approach to particle size reduction. The use of roller mills allows for precise adjustment of grinding settings, resulting in flours with consistent particle sizes and improved functional properties (Posner & Hibbs, 2005). The particle size of flour influences various functional attributes, including water absorption, dough rheology, and baking performance. Finer particles generally exhibit increased water absorption capacity, affecting the hydration kinetics during dough formation (Ozsisli *et al*., 2019).

However, the significance of these flours extends beyond their role in culinary practices. They are renowned for their affordability and accessibility, addressing the dietary needs of various communities, including those residing in remote and underserved regions. Yet, these grains have come under scrutiny due to potential contamination by heavy metals and anti-nutritional factors. An emerging concern revolves around the potential contamination of these indigenous flours by heavy metals (Baba & Bhat, 2020). Heavy metals like Cadmium (Cd), Aluminium (Al), Lead (Pb), Arsenic (As), and Nickel (Ni) are introduced into the grains through environmental exposure, possibly via soil, water sources, and atmospheric deposits. Prolonged exposure to elevated levels of these heavy metals poses severe health risks, particularly to neurological development and the well-being of specific demographic groups (Obioh *et al*., 2019). This raises queries regarding the extent of heavy metal contamination in indigenous flours and its potential implications for public health.

These cereals serve beyond mere sustenance, and are also used in a variety of culinary functions, from the preparation of baked products, to the making of wholesome porridge and the thickening of stews. Over the years, cassava (*Manihot esculenta*) has emerged as a dominant staple of primary or secondary importance in many developing countries of Sub-Saharan Africa. Cassava is a starchy staple whose roots are very rich in carbohydrates, a major source of energy. Cassava flour's effective functionalities are facilitated by its amylose (20–30%) and amylopectin (70–80%) contents (Naziri *et al*., 2014). It is widely used for the production of foods such as breakfast cereals, cookies, noodles, pastries such as breads, cakes, muffins, and doughnuts (Akinlonu, 2011). Cassava flour appears to be of lower nutritional value than cereals, legumes, and even some other root and tuber crops, such as yams (Latham, 2019); hence exploring other kinds of foods that can be used interchangeably is a good idea especially for the purpose of promoting good health. Summarily, this research sought to investigate the functional properties of four edible flours which can be used as swallows, and to determine their heavy metal concentrations as well.

Materials and Methods

Ethics

For this study, ethical clearance was applied for and obtained from the Faculty Animal Research Ethics Committee (FAREC-FBMS), Faculty of Basic Medical Sciences, University of Calabar, Cross River State in July, 2023.

Collection of Samples

The four samples were purchased from three different locations in Nigeria: cassava flour (Calabar), rye (Jos), black fonio, and finger millet (Benue). All samples were conveyed to the Department of Human Nutrition and Dietetics, University of Calabar, Calabar for sample preparation. The cereal samples (finger millet, black fonio and rye) were selected, washed using clean water and dried under the sun for three days. The sun-dried grains were ground into fine powder using a miller (Retsh ZM 200 miller, Germany). The purchased cassava flour was also opened and placed in a clean ziploc bag. The flours were placed and sealed in labelled ziploc bags and sent to the laboratory for analyses.

Sample digestion and determination Heavy metals

Heavy metal concentrations of the samples were determined using Atomic Absorption Spectrophotometric method as described by AOAC (2010). The samples were first digested using acid mixture of nitric acid, perchloric acid and sulphuric acid. The digest was later filtered then diluted to reach the 50 ml mark with distilled water. Preparation of standard solutions of the elements was prepared from Standard solution containing 1000 ppm of each element in 2N nitric acid solution. Each element was calibrated and measured against a blank, and this was done at different working wavelengths as stated in the Table A below:

Experimental procedure for Determination of Functional properties

Determination of Bulk Density (BD)

The Bulk Density of the flour samples was determined by the method of Onwuka (2018). A 10ml capacity graduated measuring cylinder was weighed and filled with the sample to the 10ml mark. The bottom of the cylinder was tapped gently but repeatedly on a laboratory bench until there was no further diminution of the sample level after filling to the 10ml mark. The cylinder with the sample was weighed.

Calculation:

$$\text{BD (g/ml)} = \frac{\text{Weight of sample (g)}}{\text{Volume of sample (ml)}}$$

Determination of Water/Oil Absorption Capacity (WAC/ OAC)

The method of Onwuka (2018) was used to determine the water and oil absorption capacity. One (1) gramme of each sample was weighed into conical graduated centrifuge tubes, then 10ml of water or oil was added to the weighed sample and mixed thoroughly. The sample was allowed to stand for 30 minutes at room temperature and then centrifuged at 5000 rpm for 30 minutes. After then, the volume of the free water/oil was read directly from the graduated centrifuge tube. The absorption capacity was expressed as gramme of oil or water absorbed per gramme of sample.

Calculation:

$$\frac{\text{Water}}{\text{oil}}\ \text{absorption capacity}\ \left(\frac{\text{g}}{\text{g}}\right) = \frac{\text{Volume of}\ \frac{\text{oil}}{\text{water}}\ \text{used} - \text{volume of free}\ \frac{\text{oil}}{\text{water}}}{\text{Weight of sample}}\ \text{x 100}$$

Determination of Swelling Index/ Capacity (SI)

The method described by Ukpabi and Ndimele (1990) was used. Ten (10) grammes of the flour sample were measured into a 300ml clean, graduated measuring cylinder and the volume was noted. Then 150ml of distilled water was added to the flour sample; the cylinder was swirled and allowed to stand for four hours. The final volume after swelling was recorded. The percentage swelling was calculated as:

$$\text{\% Swelling capacity} = \frac{\text{Final volume} - \text{Initial volume}}{\text{Initial volume}}\ \text{x 100}$$

Foam Capacity (FC) and Foam Stability (FS)

Foam capacity and stability of the flour samples were determined according to the method described by Onwuka (2018). Two (2) grammes of the flour sample were blended with 100ml distilled water in a blender and the suspension was whipped at 1600 rpm for 5 minutes. The whipped mixture was transferred into 250ml measuring cylinder. The blender was rinsed with 10ml distilled water, and the sample was gently added to the measuring cylinder. The percentage increase in volume of foam after whipping was expressed as the foam capacity, while the volume of foam at 15, 30, 60 and 120 seconds after whipping were recorded and used to determine foam stability. These were expressed using the formula as follows:

$$\text{Foam capacity (\% volume increase)} = \frac{\text{Volume after whipping} - \text{Volume before whipping}}{\text{Volume before whipping}}\ \text{X 100}$$

$$\text{Foam stability (\%)} = \frac{\text{Foam volume after time `t'}}{\text{Initial foam volume}}\ \text{X 100}$$

Emulsion Stability/Capacity (ES/EC)

The emulsion stability and emulsion capacity of the flours was determined by the method described by Chandra and Samsher (2013). The emulsion (1 g sample, 10 ml distilled water and 10 ml soybean oil) was prepared in a calibrated centrifuged tube. It was then centrifuged at 2000 × g for 5 min. The ratio of the height of the emulsion layer to the overall height of the mixture was computed to determine emulsion activity in percentage. The emulsion stability was determined by heating the emulsion in the calibrated centrifuged tube in a water bath at 80°C for 30 minutes, chilling for 15 minutes under running tap water, then centrifuging at 3000 rpm for 15 minutes. The emulsion stability, reported as a percentage, was obtained by dividing the height of the emulsified layer by the overall height of the mixture.

Gelatinization Temperature (GT)

The method of Onwuka (2018) was adopted in the determination of gelatinization temperature. Ten (10) grams of the flour sample was dispersed in distilled water in a 250ml test tube and made up to 100ml flour suspension. In a bath of boiling water, the aqueous solution was heated while being constantly stirred. Until the suspension started to gel, the heating and stirring were maintained. Then 30 seconds after the gelatinization was noticed, the corresponding temperature was recorded as the gelatinization temperature of that sample.

Data analysis

The analytical findings from the laboratory were collated, input into a Microsoft Excel 2013 spread sheet, and evaluated. The results were presented as mean ± SEM. Utilizing the Statistical Package for Social Sciences (SPSS version 20.0), Analysis of Variance (ANOVA) was performed, with significance set at $p< 0.05$.

Data availability Statement

This article contains the results that corroborate the study's conclusions. The authors are willing to provide any other information upon request.

Results and Discussion

Results

Functional properties of the edible flours

In Table 1, the functional properties of the four edible flours used for swallows are reported. Cassava, finger millet and black fonio had the same value for bulk density (0.58g/ml), while rye flour had significantly ($p<0.05$) higher BD of 0.63g/ml. The water absorption capacity (WAC) ranged between 1.85 ± 0.00g/g (black fonio) and 2.24 ± 0.06g/g (cassava flour). With regard to the swelling index (SI) of the edible flours, cassava flour recorded significantly ($p<0.05$) higher value of 3.27 ± 0.01% than the others, while rye flour had the least SI value of 2.79 ± 0.01%. The Emulsion Capacity (EC) differed significantly ($p<0.05$) among the four edible flour samples, ranging from 19.88 ± 0.02% (black fonio) to 22.69 ± 0.05% (rye flour). Rye had significantly ($p<0.05$) the lowest Gelatinization Temperature (GT) of 64.67 ± 0.08°C while black fonio had the highest GT (70.58 ± 0.04°C).

Heavy metal content of the edible flours

From the results shown in Table 2, the heavy metals – cadmium (Cd), aluminium (Al), lead (Pb) and Chromium (Cr), were found in minimal quantities (with SEMs of 0.00) which fell within the Codex Alimentarius (2015) Tolerance range for the respective elements. Rye flour had significantly ($p<0.05$) higher content of Cadmium (0.08 mg/100g) and Chromium (0.08 mg/100g) than the other edible flours. By observation, black fonio recorded the least values for most of the heavy metals. Cd concentrations ranged from 0.01 mg/100g (black fonio) to 0.08 mg/100g (rye flour). For lead, cassava flour, black fonio flour and rye flour had the same value (0.01mg/100g) but finger millet had significantly ($p<0.05$) higher content of 0.07 mg/100g. Similarly, cassava flour, finger millet and black fonio flour had the same value for Chromium (0.02 mg/100g) while rye flour had a significantly ($p<0.05$) higher amount (0.08 mg/100g) than the other flour samples.

Discussion

The functional properties of flours have been said to be affected by heat processing (Hasmadi *et al*., 2020). The flour samples had relatively lower swelling power, water absorption capacity, and oil absorption capacity, when compared to results of other edible flours such as potatoes and maize flours as reported by Hasmadi *et al*., 2020; hence they may not be utilized in goods as humectants, thickeners, stabilizers, or flavor retainers effectively.

Certain factors have been said to influence functional properties of flours; “protein content of foods is believed to be generally responsible for functional properties, such as emulsification, nitrogen solubility, oil, foaming, and water absorption” (Kinsella, 1979; Hasmadi *et al*., 2020). A flour's low protein level and lack of gluten are considered drawbacks when it comes to using it exclusively in culinary items, particularly ones where the dough's flexibility is crucial to the final product's quality. The amount and quality of wheat gluten as well as the type of product in question determine how much wheat flour is needed in composite flours to obtain a certain result (Mepba *et al*., 2007). The functional features establish whether composite flour blends are effective in baked goods where moisture is necessary for better handling, and in ground meat, doughnuts, and pancakes where the oil absorption property is crucial (Mepba *et al*., 2007). Kinsella (1979) defined functional properties as the important physicochemical characteristics that control the intricate relationship between molecule conformation, composition, and structure. The findings of this study showed that cassava had better WAC which makes it more easily soluble in water than the other edible flours. WAC refers to a substance's capacity to withstand gravity, such as binding, hydrodynamic, capillary, and physically entrapped water (Moure *et al*., 2006). This ability is exhibited by wheat and starch. Per Masood *et al*. (2011), farinographic experiments shown a considerable increase in water absorption capacity upon combining wheat flour with cowpea flour, fermented cowpea flour, and germinated cowpea flour. This is because wheat flour has increased the amount of protein and fibre in the diet. The water absorption capacity reported by Hasmadi *et al*., (2020) for raw winged bean flour (2.1 g/g) is close to the values obtained in this study. Variations in the water absorption capacity of the wheat flour-bran blends may be caused by the presence of distinct hydrophilic carbohydrates (dextrin, cellulose, arabinoxylans, etc.) as well as variable protein structures.

The GT shows that rye flour does not need to get to boiling point in water before it gelatinises. In addition, the foaming capability of many flours, including wheat flour, rice flour, green gram flour, and potato flour, was noted by Suresh and Samsher (2013). Green gram flour had the highest foam capacity (24.23%), followed by potato flour (6.84%), wheat flour (12.92%), and rice flour (3.52%). Due to its high protein content, which may reduce the surface tension at the water-air interface and create a continuous, cohesive layer surrounding the foam's air bubbles, green gram flour exhibited the largest foam capacity (Kaushal *et al*., 2012). The FC of wheat in their study is similar to the values obtained for the flours in this study. Mepba *et al*. (2007) found that product foamability is correlated with the rate of decrease in surface tension at the air/water interface caused by protein absorption. Compared with raw cowpea flour, germination increased the foam capacity of cowpea flour, but decreased the foam stability (Giami, 1993); the foamability of bean (cowpea) flour is a desirable characteristic in the production of many traditional cowpea-based food products in Nigeria such as 'akara' (bean cake) and 'moimoi' (bean pudding) (Hasmadi *et al*., 2020).

Du *et al*. (2014) assessed the bulk density of whole flours made from legumes such as pinto bean, lima bean, red kidney bean, black bean, navy bean, small red bean, black eye bean, mung bean, lentil, and chickpea. According to them, the bulk density of these legume flours varied from 0.543 g/ml to 0.816 g/ml, which is similar to the 0.58 g/ml range found in this study. According to Ikpeme-Emmanuel *et al*. (2009), As the bulk density value decreases, the more flour particles can remain together, resulting in a higher energy content from such a diet. Where certain flours lack some required functional attributes for culinary purposes, they are usually combined with other flours which possess the desired functional properties, thereby producing a composite flour. Many composite flours are rich in micronutrients, as the nutrient contents of the individual flours complement themselves.

Lastly, regarding the heavy metals, all the values obtained in this study were within the Tolerance range stated by Codex Alimentarius (2015) meaning these flours are safe for consumption. Heavy metal contamination can cause serious and fatal health effects, especially when toxicity occurs as a result of bioaccumulation; hence, it must always be guarded against in order to maintain safe food quality. For instance, acute lead toxicity can result in fatigue, irritability, insomnia, headaches, loss of appetite, high blood pressure, and vertigo. While chronic toxicity can result in neurological disorders, cognitive impairments, premature births, brain injury, kidney dysfunctions, liver damage, paralysis, and even death. (Briffa *et al*., 2020; Jaishankar *et al*., 2014). Likewise, cadmium toxicity can lead to high blood pressure, reproductive problems, decreased fetal growth, premature births, iron deficiency, gastrointestinal disorders, bone fractures, renal dysfunction, and lung cancer (Geng & Wang, 2019; Yu *et al*., 2011).

Limitation of the Study: The main limitation of this study was insufficient funds to analyze other heavy metals and also carry out some shelf-life studies. This will be looked into subsequently.

Conclusion

The cereal flours demonstrated relatively good functional properties, although cassava flour seemed to exhibit better functional properties, which could be due to its industrial processing. With these functional properties, it is possible to make these cereal flours into swallows (stiff porridge) which may be used to eat native soups just like the popular garri is used: even if it means adding some amount of a natural thickening agent like psyllium husk (especially to rye). This may offer better nutrition outcomes, particularly for people managing NCDs such as diabetes mellitus type 2 and obesity, since these cereal flours have less starch but more protein and fibre content. The functional qualities of flours are a crucial component in the production of a variety of high-quality food items in terms of appearance, organoleptic features, and customer acceptability. This could also be applicable in the baking industries, and their consumption should be promoted. Appropriate processing methods have also been proven to reduce heavy metal contamination to minimal levels, making these cereals safe for consumption.

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Table A: Digestion and determination Heavy metals

	Elements	**Wavelengths (in nm)**
1	Cadmium (Cd)	228.0
2	Aluminium (Al)	245.8
3	Lead (Pb)	263.0
4	Chromium (Cr)	216.5

Calculation:

Element: Au/A5 X ppm of Curve X Vf/Vx X 1000/W
where Au is Absorbance of sample
A5 = Absorbance of standard
Vf = Total volume of extract
Vx = Volume of extract used
W = Weight of sample used

Table 1: Functional properties of the different edible flours made into swallows

Flours	BD (g/ml)	WAC (g/g)	S1 (%)	EC (%)	ES (%)	GT (^{o}C)	FC (%)
Cassava	0.58 ±0.00	2.24 ±0.06	3.27 ±0.01	21.56 ±0.04	18.38 ±0.04	65.28 ±0.14	16.67 ±0.13
Finger Millet	0.58 ±0.00*	1.91 ±0.01*	2.89 ±0.00*	20.69 ±0.05*	16.68 ±0.12*	68.33 ±0.09*	15.71 ±0.01*
Black Fonio	0.58 ±0.00*	1.85 ±0.00*	2.81 ±0.02*^{,a}	19.88 ±0.02*^{,a}	15.39 ±0.01*^{,a}	70.58 ±0.04*^{,a}	15.81 ±0.01*
Rye	0.63 ±0.00*^{,a,b}	1.95 ±0.01^{b}	2.79 ±0.01*^{,a}	22.69 ±0.05*^{,a,b}	18.85 ±0.05*^{,a,c}	64.67 ±0.08*^{,a,b}	14.38 ±0.04*^{,a,b}

**** Values are presented as mean ±SEM, n = 3.   * = significantly different from cassava flour at $p<0.05$***
a = significantly different from finger millet flour at $p<0.05$ b = significantly different from black fonio flour at $p<0.05$

Table 2: Heavy Metal concentrations of the edible flours

Flours	Cadmium (mg/100g)	Aluminium (mg/100g)	Lead (mg/100g)	Chromium (mg/100g)
Cassava	0.02 ±0.00	0.07 ±0.00	0.01 ±0.00	0.02 ±0.00
Finger Millet	0.02 ±0.00*	0.07 ±0.00*	0.07 ±0.00*	0.02 ±0.00*
Black Fonio	0.01 ±0.00^{*,a}	0.06 ±0.00^{*,a}	0.01 ±0.00^{a}	0.02 ±0.00^{*,a}
Rye	0.08 ±0.00*^{,a,b}	0.05 ±0.00*^{,a,b}	0.01 ±0.00*^{,a,b}	0.08 ±0.00*^{,a,b}
Acceptable Levels	0.05	0.05	0.01	0.1
Tolerance Range	0.05 - 2.00	0.05 - 1.00	0.01 - 3.00	0.1 - 0.2

**** Values are presented as mean ±SEM, n = 3.   * = significantly different from cassava flour at $p<0.05$***
a = significantly different from finger millet flour at $p<0.05$ b = significantly different from black fonio flour at $p<0.05$
Acceptable levels and Tolerance range (Codex Alimentarius, 2015)