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Effect of Different Processing Techniques on Nutritional and Anti-nutritional Composition of Winged bean (*Psophocarpus tetragonolobus L. Dc.*) Seed

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Abstract
The high cost of plant protein inclusion in human and animal diets and the need to find alternative sources to traditional protein sources is on the increase. Winged bean (*Psophocarpus tetragonolobus*), a tropical underutilized legume was therefore analysed for its nutritional and anti-nutritional contents using different processing techniques. Two kilograms (2kg) of winged bean seeds were purchased, and samples were divided into different parts and subjected to two different processing techniques; baking at 250°C and boiling at 100°C for 20 minutes. An unprocessed portion was also separated as control. Samples from each processing technique and control were subjected to analysis for nutritional and anti-nutritional factors using standard laboratory methods. The crude protein content of winged beans from baking, boiling and control was 17.93%, 19.59 and 12.55% respectively. Crude fat was 11.25%, 9.25% and 10.40% in that order. The highest anti-nutritional factor of 2788.45 mg/100g and 3254.57 mg/100g was recorded for processed and unprocessed tannin composition also. Oxalate values reduced by 22% and 19.74%, while, phytate content reduced by 20% and 18.12% for the processed boiled and baked after baking and boiling respectively. Nutritive and anti-nutritive compositions of the processed and unprocessed winged beans were significantly different at $p>0.05$. Processing is therefore a viable means of increasing the nutritive value and in turn reducing the anti-nutritive values of winged beans.

Keywords: Winged beans, Nutritional value, Crude protein, Crude fibre, Food security

Introduction

Winged beans (*Psophocarpus tetragonolobus*), also known as cigarillas, goa beans, four-angled beans, four-cornered beans, manila beans, princess beans, asparagus beans or dragon beans, is a tropical leguminous crop which originated from New Guinea (Patrush *et al.*, 2017). Winged beans are mainly cultivated on small scale in hot, humid equatorial countries of South and South East Asia (Hussein *et al.*, 2020). The crop is widely recognised by farmers and consumers in Southern India and Thailand for its variety of uses, rich nutrients, and edibility (Singh *et al.*, 2013; Lawal, 2019). The leaves, flowers, pods, green seeds, dried seeds, and tuberous roots of winged beans are all edible and nutritious (Lawal, 2019). According to Mohanty (2020) and Calvindi (2020), the tender young shoots, flowers and leaves of winged beans may be eaten raw or cooked as green vegetables. The protein content of the flowers appears to be fairly high compared with edible flowers of other better-known tropical plants such as banana and *Sesbania grandiflora* (Agathi). The tubers of winged beans had also been reported to have an extraordinarily high protein content of 8 – 20% (dry weight) compared to 1 – 5 % contained in cassava, potatoes, and some common cultivars of root crops. The half-ripe seeds can be removed from the pod and cooked like peas or kidney beans, flowers can be eaten raw, fried or steamed. The crop had also been reported to be rich in carbohydrates, edible oil, essential amino acids, protein and vitamin A to the tune of these reported for soybeans (Amoo *et al.*, 2011; Singh *et al.*, 2013; Abberton *et al.*, 2020; Calvindi, 2020). Winged beans "milk" and flour are suitable as dietary treatments in protein-deprived children. According to Calvindi *et al.* (2020), winged beans can also be employed for the treatment of cancer, infection, diabetes, eye and migraine diseases, asthma and muscle weaknesses. The beneficial use of winged beans can also be harnessed to ensure healthy reproductive health and pregnancy in women and promotion of immunity, respiratory health, bone health and male fertility (Singh *et al*., 2019).

Research into use of underutilized protein-rich legumes would go a long way to address looming food insecurity, protein shortages, and the high cost of protein, especially in developing countries where animal protein is far from the reach of a majority of the populace (Raai *et al*., 2020). The winged bean has a promising future and potential for expanded use, especially in the humid tropics due to its hardness and high protein contents (FAO, 2015; Massawe *et al*., 2016; Tanzi *et al.*, 2019; Raai *et al*., 2020). Earlier studies have been made on winged beans as a minor pulse and grossly underutilized crop. Singh *et al*. (2019) investigated the variation in the antioxidant, phenol and flavonoid properties of different winged bean genotypes using chemometric analysis. Singh *et al*. (*ibid*) further identified that AMBIKAWB-II-I, AMBIKA-13-4 B and MWBS-16-26 genotypes were superior based on the investigated traits. Makeri *et al*. (2017) characterized protein fractions of winged bean seeds in comparison with soybeans, using physiochemical, functional and structural analysis. This study concluded that winged beans had some better functional properties than soybeans and also compared favourably with those reported for most legume seeds. However, soybeans were discovered to be more digestible than winged beans due to the low content of β-sheets in the former. Despite some of these studies, none has addressed the processing techniques for winged beans. This research was therefore designed to investigate the best processing technique for winged beans for human and animal consumption. The suitability of processed winged bean seeds as a source of protein, and its different nutritional and anti-nutritional compositions resulting from different processing techniques were investigated in this study.

Materials and Methods

Seed Sources

Two kilograms (2kg) of winged beans (*Psophocarpus tetragonolobus*) seeds were purchased from Ipata market; an open grain market in the Ipata area of Ilorin, Kwara State, Nigeria. Ipata market is located on longitude of 4.56° E and latitude of 8.50° N. The seed colour was brownish white at the point of purchase.

Experimental Design

The experiment was conducted using a completely randomized design with three treatments. The treatments include; two processing techniques (baking and boiling) and an unprocessed portion of seeds as control. Each processing lasted for 20 minutes. Chemical analyses were thereafter carried out in the laboratory in two replicates to identify the effects of processing on nutritive and anti-nutritive contents of winged beans.

Processing of Seeds

Purchased two kilograms' seeds were cleaned by winnowing of all associated physical dirt and foreign material present with the seeds. Dry basis moisture contents of seeds were tested and obtained before processing, using standard laboratory methods. Seeds were thereafter subjected to different processing as treatments, before analysing the nutritional and anti-nutritional characteristics of the seeds. Five hundred grams (500g) of seeds were employed for each processing technique.

Baking Process

Five hundred grams (500g) of clean winged beans were baked in a flamemax electric commercial, 15kg capacity oven for a period of 20 minutes after moisture content determination. A maximum uniform temperature of 250^{o}C was maintained throughout the oven cabin. Winged bean seeds were placed in five relatively flat stainless containers having 100g capacity each and steered after 10 minutes in other to ensure uniform distribution of baking temperature throughout the winged bean.

Boiling Process

Five hundred grams (500g) of seeds to be boiled were washed thoroughly under running water to remove all physical dirt and unwanted colouration from the seeds. Seeds were thereafter subjected to boiling in clean water using a new and thoroughly washed 10 litres capacity stainless pot. Seed quantities were placed in 2L of clean water and boiled on a liquefied natural gas stove for a period of 20 minutes. A maximum temperature of 100^{o}C was maintained throughout the process. Leftover water was drained from the beans after 20 minutes, before laboratory analysis for nutritive and anti-nutritive composition

Unprocessed Winged Beans- Five hundred grams (500g) of winged beans were also separated and left unprocessed after cleaning and removal of foreign materials. This was necessary to serve as a control experiment in other to ascertain the effects of boiling and baking processing techniques on the nutritive and anti-nutritional composition and changes of processed winged beans.

Chemical Laboratory Analysis

Six nutritive and four anti-nutritive compositions of winged bean were tested to determine some effects of processing techniques on winged bean. The nutritive composition tested include; percentage dry matter, percentage moisture content after processing, percentage crude fibre, percentage crude fat, percentage crude protein and percentage ash content. In like manner, anti-nutritive compositions tested include saponin, phytate, oxalate and tannin contents of winged beans. Each composition tested was done in two replicates and averages as well as standard deviations determined. Standard analytical methods specified by the Association of Official Analytical Chemists (AOAC, 2005) were used in this study. Percentages of crude protein, crude fat, crude fibre, ash and dry matter contents of processed and unprocessed winged beans were determined using AOAC standards of 988.05, 2003.06, 958.06, 942.05 and 967.08 respectively. Saponin, phytate, oxalate and tannin contents of processed and unprocessed winged beans were determined using procedures highlighted by Ejimeke *et al*. (2014).

Statistical Analysis

Results obtained from analysing the nutritive and anti-nutritive contents of processed and unprocessed winged beans were subjected to descriptive statistics and one-way Analysis of Variance (ANOVA) to determine the differences between the means and variance of each composition considered. The means were separated using the Least Square Difference (LSD). These were done using SPSS (version 25.0) at a 95% level of confidence.

Results and Discussion

Nutritive Composition of Processed and Unprocessed Winged Bean

Table 1 highlighted the nutritive composition of processed and unprocessed winged beans. Highlighted nutritive content include; percentages of dry matter, moisture content, crude protein, crude fibre, crude fat and ash contents. The crude fibre content in the processed seeds ranged from 18.10% (baked) to 19.59% (boiled). Crude fibre contents of processed winged beans increased by 7% and 5.55% as a result of boiling and baking processes respectively. Similarly, crude protein contents were made more available by processing. Boiling increased the percentage of crude protein by 7.04%, while, the baking process increase available crude protein by 5.38%. On the contrary, the percentage of ash content was reduced by the baking and boiling processes. However, crude fat content was reduced by baking, but increased during the boiling process. This may have resulted in high heat involved in the baking process, thus, subliming some crude fat contents of winged beans. Percentage ash contents decreased by 1.05% from baking, and by 2.15% when boiled. Percentage crude fat on the other hand increased by 0.85% when boiled, but reduced by 1.15% when baked. As expected, boiling increased the percentage seed moisture content by 9.38 %, but reduced the percentage of dry matter by the same amount. The baking process also increased the moisture content by 3.78% and the percentage of dry matter by the same quantity.

The percentage of crude fibre in processed seeds was consistent with those reported by Singh *et al.* (2012) as 12.65% in fully mature seeds and 2.76% in tubers. Comparatively, lower percentages of 1.23% and 0.85% were reported by Leach *et al*. (1959) for brown rice and wheat flours respectively. David *et al.* (2015) reported a crude fibre percentage of 3.21% for Asomdwee cowpea flour and Chinma and Gernah (2007) reported a crude fibre percentage of 8.19% for pigeon pea, 9.58% for cowpea, and 4.61% for mung beans flour. These are all lower than the percentage of crude fibre obtained from processed and unprocessed winged beans. These imply winged contain much more fibre than other commonly consumed legumes and cereals. The percentage of crude fibre obtained from processed winged beans was significantly different from the percentage of crude fibre from unprocessed winged beans ($p<0.05$). There was however insignificant difference between boiled and baked winged beans in terms of the percentage of crude fibre contents.

The percentage of crude protein in the processed winged bean is in line with the protein contents reported for winged beans by Kantha and Erdman (1984), ranging from 17–19%. This range is also in tandem with the crude protein contents reported for some other legumes including; cowpea (22.5%), pigeon pea (22.4%), and lima beans (23.3%) (Morris *et al.*, 1999; Adegboyega *et al.*, 2019). The percentage of crude protein reported for soybeans however almost doubled the percentage of crude protein in processed winged beans (Morris *et al.*, 1999; Adegboyega *et al.*, 2019). The values exceed 14.70% and 12.86% previously reported for wheat flour by Morris *et al.* (1999). These suggest flours made from winged beans could be suitably blended with wheat or cowpea flour as composite flours for increased protein content. Relatively high protein contents of winged beans could thus play significant roles in human and animal nutrition improvement. Similar to crude fibre contents, the percentage of crude protein obtained from processed and unprocessed winged beans is significantly different at 0.05 level. However, no significant difference exists between the percentage of crude protein obtained from the boiled and baked winged beans.

Singh *et al.* (2012) reported lower crude fat content of 0.47% in fully mature winged bean seeds. Lepcha *et al.* (2017) however, reported crude fact contents ranging from 15 – 20.4%. Boye *et al.* (2010) reported a range of 5.76 to 6.87% for chickpeas. Winged bean oil resulting from its processed crude fat contents is highly valuable for frying; due to its better thermal conductivity and oxidative features than soybeans oil (Makeri *et al*., 2016). Winged bean oil however has more saturated fatty acids content; thus less preferred than soybeans oil (Makeri *et al*., 2016). Differences in the percentage crude fat contents of the processed and unprocessed winged beans are statistically insignificant since $p>0.05$. Food ash contents indicate its mineral contents. Processed and unprocessed ash contents of winged beans were comparatively higher than those reported for mung beans flour (2.53%), chickpea flour (2.53%), pigeon pea (4.58%), processed cowpea (4.73%) and mucuna beans flour (3.25%) (Olalekan and Abosede, 2010). Processing, however, reduced the mineral content of winged beans, but no significant difference exists between the percentage ash contents of unprocessed and baked winged beans, while the boiling process resulted in significantly percentage reduced ash contents.

Anti-Nutritive Composition of Processed and Unprocessed Winged Beans

Table 2 highlights the anti-nutritive composition of processed and unprocessed winged beans. Highlighted anti-nutritive content includes percentages of Saponin, phytate, oxalate and tannin. Despite all the positive nutrition benefits offered by winged beans, some anti-nutritional factors may however limit their usage and consumption when not well processed. Like many legumes (beans), winged beans contain phenolics, tannins, phytic acid, flatulence factors, saponins, and hydrogen cyanide (Adegboyega *et al*., 2019). Some of these compounds, especially tannins and phenolics specifically inhibit enzyme activity by forming complexes with food proteins, thus reducing such protein's quality (Kantha *et al.*, 1986). Tannins have been described to have cross-links with proteins, which leads to a reduction in in-vitro protein digestion. They have been traced to inhibition of digestive enzymes, increased excretion of endogenous protein, and unfavourable effects on the digestive tract (Parmar *et al.*, 2017). Oxalate forms soluble and insoluble salts with minerals, especially insoluble chelate complex with dietary calcium (Akhtar *et al.*, 2011). Calcium oxalate which is an insoluble crystal can easily accumulate in the renal glomeruli, thus causing renal diseases such as kidney stones (Massey, 2007). Soluble oxalates are, however, more available in plant and food products than insoluble oxalates (Chai and Leibman, 2004). More attention is therefore usually placed on soluble oxalates.

Phytate, as an antioxidant, binds to some dietary minerals and interferes with the availability of such minerals (Graf *et al*., 1987). Phytate content in winged beans was estimated to be similar to that of soybeans. Pythates occur naturally as compounds in many kinds of cereals and legumes (Thrane *et al.*, 2017). Phytate has anti-nutritional abilities in humans and animals with a single stomach compartment, through its strong chelation of Ca, Fe, and Zn. This process forms unabsorbed insoluble complexes and thus leads to Fe and Zn deficiency (Raboy, 2001). They restrain the bioavailability of minerals and the digestibility of proteins in humans and animals (Spier *et al*., 2018). Phytate removal could increase phosphorus bioavailability and also enhances the nutritional value derived from meals (Kumar *et al.,* 2018). However, phytate also has positive nutritional roles as an antioxidant and anticancer agent. They suppress the formation of hydroxyl formation mediated by iron which is complexed by phytate (Graf *et al*., 1987; Graf and Eaton, 1993).

It had, however, been evidenced from the literature that the use of moist heat or soaking in water above room temperature over a while, could effectively and safely eliminate these substances without reducing their nutritional composition (Doss *et al*., 2011). This claim was also corroborated by results obtained from this research. Twenty minutes of baking reduced the saponin, phytate, oxalate and tannin contents of processed winged beans by 5.98mg/g (22%), 0.023mg/g (20%), 0.023mg/g (22%) and 4.66 mg/g (14%) respectively. Statistical comparison from ANOVA and LSD showed a significant reduction in measured anti-nutritive contents of processed winged beans. These suggest further baking time might drastically reduce the anti-nutritive contents of winged beans to make it fit for consumption

Twenty minutes of boiling reduced saponin, phytate, oxalate and tannin contents of processed winged beans by 8.02 mg/g (29.7%), 0.02 mg/g (18.12%), 0.02 mg./g (19.74%) and 6.61 mg/g (20.31%) respectively. From Table 2, the boiling process removed the anti-nutritive composition of winged beans better than the baking process. Saponin, phytate, oxalate and tannin contents of boiled and baked winged beans differed by 9.71%, 2.63%, 1.83% and 7.01% respectively. Saponin, phytate, oxalate and tannin contents of the processed winged beans are significantly different from those of unprocessed winged beans. In the same vein, saponin, phytate, oxalate and tannin contents of the boiled winged beans differed significantly from those of baked winged beans. These differences further strengthen the claim that the use of moisture in the presence of heat could effectively deal with the major anti-nutritive composition of winged beans better than only heat. Based on these results boiling is a more recommended processing technique for winged beans, especially in the removal of their anti-nutritive compositions.

Conclusion

This study evaluated the effect of two processing techniques on the nutritional and anti-nutritional compositions of winged beans. It was observed that the two processing techniques had significant impacts on the nutritional and anti-nutritional compositions of the processed seeds. Percentage of crude fibre and crude protein were made more available in winged beans by the two processing techniques, while crude fat was reduced by baking but increased by boiling. Percentage ash contents which indicate mineral availability in food substances were reduced by both means of processing. Crude fibre contents of the seeds after processing were higher than that of most other legumes this thus positioned winged beans as a functional food with health benefits. Furthermore, both processing techniques had reduced effects on the anti-nutritional composition of winged beans. However, the boiling process removed these unwanted components better than baking. These imply winged bean flour has the potential to be incorporated into food formulations as a functional ingredient. With these promising potentials of winged beans and the added values brought by processing, further research on the effect of processing time on different properties and contents of winged beans is recommended. These will further position winged beans in the limelight of being utilized rather than their presently underutilized position. Further research on their agronomic performance and optimal inclusion levels in food formulations are as well recommended.

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Table 1: Nutritive Composition of Processed and Unprocessed Winged Beans

Sample ID	% DM	% MC	% Ash	% Crude Fat	% Crude Protein	% Crude Fibre
Baked	90.12±0.00^{b}	9.88±0.00^{b}	3.90 ± 0.42^{a}	9.25±1.34^{a}	17.93±1.10^{a}	18.10±0.84^{a}
Boiled	84.52±0.00^{c}	15.48±0.00^{a}	2.80±0.28^{c}	11.25±0.49^{a}	19.59±0.82^{a}	19.55±0.78^{a}
Unprocessed	93.90±0.00^{a}	6.10±0.00^{c}	4.95±0.07^{a}	10.40±0.00^{a}	12.55±0.07^{b}	12.55±0.07^{b}
p value	0..00	0.00	0.01	0.20	0.006	0.004

+ % DM is the percentage of dry matter, % MC is the percentage of moisture content
++ Figures with the different superscripts on the same column are significantly different

Table 2: Anti-Nutritive Composition of Processed and Unprocessed Winged Bean

Sample ID	Saponin(mg/100g)	Phytate(mg/100g)	Oxalate(mg/100g)	Tannin(mg/100g)
Baked	2102.3±2.79^{b}	8.89±0.03^{c}	8.54±0.02^{c}	2788.45±2.25^{b}
Boiled	1898.12±4.35^{c}	9.13±0.03^{b}	8.70±0.01^{b}	2593.07±2.79^{c}
Unprocessed	2700.00±1.42^{a}	11.15±0.07^{a}	10.84±0.03^{a}	3254.57±2.17^{a}
p-value	2.66E-07	3.85E-05	3.25E-06	2.34E-07

+ Figures with the same superscripts on the same column are not significantly different