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Iron, Zinc and Total Phenolic Content in Cooked Biofortified NUA 45 and Gloria Sugar Beans: The Case of Zimbabwe

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ABSTRACT

Biofortification involves breeding of staple crops that are micronutrient-dense and high yielding for example iron-biofortified beans. The biofortification of crops has been found to be cost effective and feasible to fight micronutrient deficiencies. The aim of the study was to determine the iron, zinc and total phenolic composition for the biofortified NUA 45 and Gloria sugar beans in raw as well as cooked samples with and without previous soaking. Determination of iron and zinc content in the raw, cooked bean grain samples with and without soaking was carried out by Inductively Coupled Plasma (ICP) Optical Emission Spectrometry whereas the determination of Total Phenolic Content was done using the Folin-Ciocalteau colorimetric method. The highest zinc concentration (0.295ppm) was found in raw Gloria sugar beans whilst the raw NUA 45 sugar beans had the highest iron content (0.755ppm) and total phenolic composition (352.8mg/100g). For the cooked bean samples Gloria sugar beans had the highest zinc concentration (0.247ppm) cooked after soaking. NUA 45 sugar beans had no significant difference on the zinc content after being cooked with and without soaking. Amongst the cooked samples the biofortified NUA 45 sugar beans cooked after soaking had the highest iron content (0.373ppm) as well as the highest total phenolic content in the beans cooked without soaking (200.5mg/100g). Biofortified NUA 45 and Gloria sugar beans proved to be good sources of iron, zinc and phenolic compounds. However, cooking with and without soaking diminished the iron, zinc and the total phenolic composition of the cooked NUA 45 and Gloria sugar beans.

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Introduction

Most of the global population survive on diets that mainly consists of staple food crops with poor dietary diversity. Limitations in dietary diversity correlates to low household income to buy more nourishing foods such as animal products, fruits, and vegetables. Micronutrient deficiencies result when there is poor consumption and absorption of minerals as well as vitamins to sustain good health and development. Nutrition specific interventions address the immediate causes of undernutrition, such as inadequate dietary intake, and some of the underlying causes, like feeding practices and access to food for example increasing dietary diversification, micronutrient supplementation, modification of food choices and fortification. Nutrition-sensitive interventions address the underlying determinants of malnutrition and include biofortification [1-5].

Biofortification is a process that focuses on breeding of micronutrient dense staple crops such as maize, beans, millet and cassava. Biofortification can be achieved through conventional plant breeding, agronomic approaches such as soil fertilization as well as genetic engineering. When biofortified staple crops are consumed regularly they produce measurable improvements in human health and nutrition. Furthermore, biofortification has been found to be cost effective and feasible to fight micronutrient deficiencies [6-7].

Pulse crops biofortified using conventional plant breeding approaches have been released by Harvest Plus to help overcome micronutrient deficiencies in developing countries.The common bean is produced in both developed and developing countries and is an important source of nutrients such as protein, carbohydrates, some vitamins, and micronutrients as well as phenolic compounds which gives common bean seeds diverse colours.Phenolic compounds include phenolic acids, flavonoids, stilbenes, coumarins and tannins. Phenolic compounds play an important role in human health because they possess antioxidant activity related to anti-diabetic, anti-obesity, anti-inflammatory, anti-mutagenic and anti-carcinogenic properties. Beans is a popular crop because it is easy to produce, flavourful and enhances soil fertility through nitrogen fixation [8-11].

Biofortified beans can contain double the amount of iron as compared to non-biofortified beans. Most ordinary beans do contain some iron and zinc, but the levels are too low to significantly improve iron status. NUA 45 sugar beans is a biofortified variety that was released in 2010 and it is a rich source of iron (95 mg/kg) as well as zinc (38 mg/kg) and fits well in crop rotation systems.

The common preparation methods for beans include soaking, and boiling and micronutrients can be lost during preparation due to chemical degradation as well as physical loss through leaching of minerals. Studies have been done on retention of iron, zinc for conventional and some biofortified beans. However, the studies to date have not reported the retention of iron, zinc and total phenolic composition of NUA45 sugar beans and Gloria sugar beans after different treatment conditions [12-14].

No studies have been done in Zimbabwe that focus on the retention of micronutrients and phenolic compounds of the commonly consumed biofortified bean variety and the Gloria sugar bean variety. Furthermore, owing to the high bean consumption in Zimbabwe, the importance of iron, zinc and phenolic compounds in human nutrition, and the high numbers of children and women deficient in these micronutrients, this study was carried out on the iron and zinc content as well as the total phenolic content in the two bean varieties after cooking. Commonly applied treatment conditions such as cooking without previous soaking as well as cooking with previous soaking of beans were used in the study to demonstrate if sufficient amounts of iron, zinc and phenolic compounds are being retained after cooking to improve health status of individuals. The study aimed to determine the amount of iron, zinc and Total Phenolic Content in a biofortified variety (NUA 45 Sugar beans) and in a commercial non-biofortified variety of the common bean (Gloria Sugar beans), using both raw samples and cooked samples with and without previous soaking.

Methodology

Bean varieties

Two different varieties of the common beans (biofortified and non-biofortified) were selected for this study. These varieties included the biofortified NUA 45 sugar beans and the non-biofortified Gloria sugar beans. The two bean varieties were grown in Makoni district, Zimbabwe in January 2022 and harvested in March 2022 respectively. The harvested bean crops were dried and stored in hermetic bags at room temperature until they were used for the study.

Cleaning Procedure for Beans and Materials

The dry beans were cleaned to remove dirt, broken beans, and disease infested beans. The materials (pots and lids) used for sample preparation were decontaminated from free minerals by overnight bathing in a 5% HCl solution with ultrapure water [15].

Sample Preparation

The determination of the iron, zinc and Total Phenolic Composition of NUA 45 sugar beans, and the Gloria sugar beans was done under the following conditions: raw beans, regular pot cooking after soaking as well as regular pot cooking without water soaking as shown in figure 1.

Sample Characteristics

Sample 1- raw NUA 45 sugar beans
Sample 2- raw Gloria sugar beans
Sample 3 – NUA 45 sugar beans cooked without soaking
Sample 4 -NUA 45 sugar beans cooked after soaking
Sample 5 – Gloria sugar beans cooked without soaking
Sample 6 - Gloria sugar beans cooked after soaking

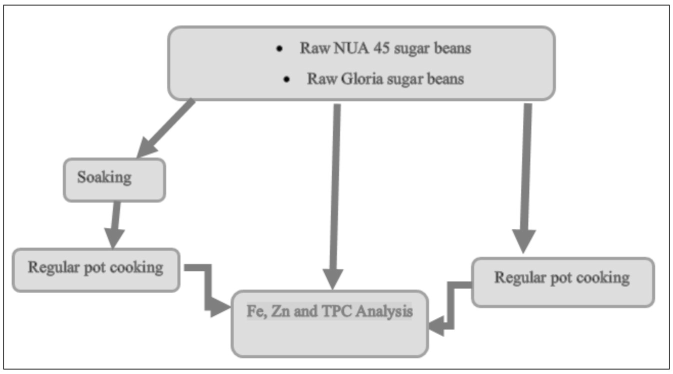

Figure 1: Overview of The Preparation Methods for The Bean Samples

The processing procedure for Cooking Without Soaking and Cooking After Soaking samples was adapted from a study with modifications by [16].

The Raw Bean Samples

The raw bean samples were selected manually for each variety and kept in clean glass receptacles. The bean grains were washed with deionized water and all the water was discarded after washing. Drying of beans was performed in an oven at 60°C, with no air circulation, overnight. The raw beans of each variety were ground in a zirconium ball mill, until a sufficient amount was obtained for analysis [17].

Samples Cooked Without Soaking (CWS)

Samples 3 and 5 were cooked without soaking by boiling 100 g of each variety of beans in 500 ml of water until they felt soft between fingers, after which they were drained. 500 ml of hot water was added during cooking to correct the loss of water by evaporation during cooking. Regular pot cooking was done with the lid half open. Cooking times for each sample were recorded. Samples for analysis of iron and zinc were taken after cooling down the beans for 30 minutes at room temperature.

Samples Cooked After Soaking (CAS)

100g of each variety of beans was soaked in 500 ml of deionised water in a glass beaker for 16 hours for samples 4 and 6. The soaking water was discarded, and the soaked beans was cooked using same procedure as the CWS samples. The CAS samples were taken after cooling down the beans for 30 minutes at room temperature [17].

Analysis of Iron and Zinc

The cooked samples (both CAS and CWS) were dried thoroughly at 80°C for at least 12 hours and were placed in a desiccator to keep samples dry. The samples were ground to a flour and the ground samples were dried again at 80 ◦C for at least 12 hours and put in a desiccator until further analysis. Determination of Iron and Zinc content in the raw and cooked bean samples was performed using Inductively Coupled Plasma Optical Emission Spectrometry using Spectro Analytical Instrument Spectro flame P at Standards Association of Zimbabwe (SAZ) laboratories in Harare, Zimbabwe [15].

Total Phenolic Content (TPC) analysis

Determination of TPC in NUA 45 sugar beans and the Gloria sugar beans was done under the following conditions: raw beans, beans prepared by regular pot cooking with and without previous water soaking.

The broth was removed after cooking followed by drying the cooked beans at 60 ºC in the ventilated oven until the beans were completely dried. The cooked and dried samples were ground to obtain homogenous samples and stored at 4ºC until further analysis. The extraction method used was according to the method by. The supernatants, considered to be polyphenol extracts, were stored at -20 ºC until analysis for their total phenolic content. The procedure for analysis of TPC of the samples was based on the Folin-Ciocalteau colorimetric method that was used in a study by. The TPC analysis was done at Midlands State University Biochemistry laboratories in Gweru, Zimbabwe. The absorbance was measured at 765 nm using a spectrophotometer against the distilled water blank. A standard curve using 0 – 500 mg/L gallic acid was constructed. The total phenolic contents were expressed as mg of gallic acid equivalent (GAE)/100 g extract [16].

Data Analysis

Data was analysed using XLSTAT trial version 2022.3.1.1323. One-way analysis of variance (ANOVA) was used to compare iron content, zinc content and the total phenolic content between raw and different processed beans, and between the two varieties of beans. Results were expressed as means and standard deviations and P value of < 0.05 was considered to be statistically significant.

Results

Cooking Times

The cooking times of the two bean varieties, with and without previous soaking in water, is shown in Table 1. The Gloria sugar beans cooked without previous soaking needed the most cooking time (240 minutes) and the NUA 45 sugar beans with previous water soaking needed the least amount of time (90 minutes) to get cooked. Samples cooked after previous soaking had the least cooking times for both varieties compared to the samples cooked without soaking.

Table 1: Cooking times of beans with and without previous soaking in water Cooking time in minutes

Variety	Without previous soaking	With previous soaking
NUA 45 sugar beans	150 minutes	90 minutes
Gloria sugar beans	240 minutes	150 minutes

Iron, Zinc and Total Phenolic Content

Raw NUA 45 sugar beans had the highest iron content (0.755ppm), followed by the raw Gloria sugar beans (0.697ppm), Table 2. For the cooked beans, the highest iron content was found in NUA 45 sugar beans (0.373ppm) cooked with previous soaking followed by Gloria sugar beans cooked without soaking (0.258ppm).

The zinc contents were highest in the raw Gloria sugar beans followed by Gloria sugar beans cooked after soaking (0.295ppm and 0.247ppm) respectively (Table 2). Gloria sugar beans cooked without previous soaking had the least zinc content(0.183ppm). The zinc contents were 0.238ppm in raw NUA 45 sugar beans and 0.124ppm after being cooked in a regular pot, with and without soaking, respectively.

Table 2 also illustrates that total phenolic content was highest in the raw NUA 45 sugar beans followed by raw Gloria sugar beans (352.8 mg/100g and 332.1 mg/100g). The least TPC were in NUA 45 sugar beans cooked with previous soaking (134.8mg/100g). After cooking without previous soaking, the NUA 45 sugar beans (200.5 mg/100g) presented higher phenolic content values than the Gloria sugar beans (175.4 mg/100g) cooked without soaking.

Table 2: Iron, Zinc and Total Phenolic Content in Raw and Cooked Beans

	Fe (ppm)	Zn (ppm)	TPC (mg/100g)
NUA 45 Raw	0.755 (±0.002476)a	0.238 (±0.003036)c	352.8 (± 4.596)a
Gloria Raw	0.697 (±0.003896)b	0.295 (±0.0007575)a	332.1 (± 6.081)b
NUA 45 CWS	0.247 (±0.00351)e	0.124 (±0.001082)e	200.5 (± 0.460)c
Gloria CWS	0.258 (±0.00809)d	0.183 (±0.0009961)d	175.4 (± 1.858)d
NUA 45 CAS	0.373 (±0.004375)c	0.124 (±0.00228)e	134.8 (± 0.830)f
Gloria CAS	0.236 (±0.00234)f	0.247 (±0.00459)b	137.7 (± 1.307)e

Different letters in the same column differ significantly

Discussion

Cooking Time

Cooking time depends mainly on the cultivar studied and the soaking time. In this study the soaking time for the two varieties was the same, hence the difference in cooking time could be attributed to the difference in cultivars. Soaking before cooking reduces the cooking time as mentioned by hence the reduction of the cooking times noted for each variety after soaking (Table 1). NUA 45 sugar beans cooked after soaking had the shortest cooking time (90 minutes). A study by [15,18-19] also showed that the biofortified varieties had the shortest cooking times than the non biofortified varities used.

Iron Content

Raw NUA 45 sugar beans had the highest iron content (0.755ppm), followed by the raw Gloria sugar beans (0.697ppm). However, iron can be lost into cooking water hence the decrease in iron content in the cooked beans for both varieties as compared to the raw beans. For the cooked beans, the highest iron content was found in NUA 45 sugar beans (0.373ppm) cooked with previous soaking followed by Gloria sugar beans cooked without soaking (0.258ppm). This could be due to soaking which was found to increase the extractability of iron from the food matrix, compared to cooking without soaking. A significant difference was found in the iron contents of the cooked bean grains for both varieties with and without water soaking [17,20].

Zinc Content

The two varieties analysed presented zinc contents ranging from 0.124ppm (NUA 45 sugar beans cooked with and without soaking) to 0.295 ppm (raw Gloria sugar beans). Zinc content in cooked Gloria sugar beans were lower than in raw Gloria sugar beans. This shows that boiling with or without prior soaking of common bean reduces its zinc retention as indicated in a study by [19].

Gloria sugar beans cooked after soaking contained higher Zn content (0.247ppm) as compared to the Gloria sugar beans cooked without soaking. Another study showed similar findings in which zinc retention was higher in boiled common bean with previous soaking compared to common bean without previous soaking. Zinc concentrations have been found to be higher in cooked (boiled and pressure cooked) versus raw beans, regardless of when or whether prior soaking had been carried out [17,19].

Zinc retention in boiled common bean with previous soaking were found to be higher than boiled common bean without previous soaking [15]. However, this was not the case for NUA 45 beans in this study which had no significant difference on the zinc content after being cooked with and without soaking. The zinc content of NUA 45 sugar beans cooked without soaking and that cooked with soaking was 0.124ppm.

The higher zinc content in boiled Gloria sugar beans with previous soaking compared to boiled Gloria sugar bean without previous soaking, observed in this study could be explained by soaking. soaking increases the extractability of zinc from the food matrix, compared to without soaking [20].

Total Phenolic Content

Generally, the two bean varieties showed a reduction of total phenolic content after cooking regardless of the samples being cooked with or without previous soaking. Processing such as cooking may promote degradation amongst the aromatic rings of the phenolic compounds, leading to polymerization reactions or structural breaks, which are reflected in the lower phenolic content of the cooked beans. Cooking after soaking seemed to be the preparation procedure that resulted in greater loss of total phenolic content in both NUA 45 beans and Gloria sugar beans compared to cooking without soaking. These results were in agreement with a previous study, whereby cooking without soaking was the most efficient preparation method in retaining the TPC. Factors such as the soaking and draining stage can reduce the total phenolic content of the beans. A possible explanation to the loss in total phenolic content can be due to leaching of the soluble phenolic into soaking water and cooking water which were discarded in this study, chemical transformation, and formation of phenolic-protein complex within thermal and stress condition. Beans can only be eaten after cooking. However, analyses of raw beans are required to identify the nutritional content of the beans. Boiling is the most basic preparation method applied in bean processing and results in loss of total phenolic content in food. If the reference was based on the data of raw beans then the actual intake of total phenolic content would be overestimated [16,21-23].

Conclusions

The biofortified NUA 45 sugar beans and the Gloria sugar beans proved to be good sources of iron, zinc and phenolic compounds. However, cooking with and without soaking diminished the iron, zinc and the total phenolic composition of the cooked NUA 45 and Gloria sugar beans. More research is needed in comparing iron, zinc and total phenolic compositions of different varieties of biofortified beans after regular pot cooking with and without soaking with water.

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Consent for Publication

All authors of this paper have given their consent in the publication of this research paper.

Availability of Data and Materials

All data and materials are available upon reasonable request from the corresponding author.

Competing Interests

Authors have declared that no competing interests exist.

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Author Contribution

Ropafadzo Chirimubwe, Amiel Mugari, Ruth Nyoka and Phyllis Nyamande contributed towards study design, data collection, statistical analysis, data presentation and article compilation.

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