Effect of dehulling, fermentation, and roasting on the nutrient and anti-nutrient content of sorghum and pearl millet flour

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Abstract—Sorghum and pearl millet contain anti-nutritional factors such as tannins and phytic acid, which limits their use in processed food products. Pre-treatment processes of these traditional grains such as dehulling, roasting, and fermentation, have potential to reduce the anti-nutritional factors. However, there is death in data on their efficacy. Therefore, this study aimed to evaluate the effect of dehulling, fermenting and roasting on the proximate, micronutrient and anti-nutritional content of sorghum and pearl millet flour from Zimbabwe. The grains were roasted, dehulled, fermented, and milled into flour. Four treatments namely, (1) unprocessed sorghum (control 1), (2) processed sorghum, (3) unprocessed pearl millet (control 2) and (4) processed pearl millet were prepared. The treatment samples were evaluated for proximate and mineral content using standard methods of analysis (AOAC, 2000) and inductively coupled plasma atomic emission spectrophotometry (ICP-OES) method, respectively. Phytates and tannins were measured using the UV Spectrophotometer method. The alkaloid content was determined gravimetrically. Data were analysed statistically using ANOVA at 95% probability. Sorghum processing i.e. dehulling, fermenting, roasting and milling significantly increased the protein content (from 15.0±0.08 to 20.0±3.98%) and ash content (from 3.6±0.05 to 4.2±0.52%) (p<0.05). No significant difference in fat content was observed between the processed and unprocessed sorghum and pearl millet (p>0.05). Similarly, the protein content of pearl millet significantly increased after processing (from 3.6±0.05 to 25±2.875) (p<0.05), while carbohydrate and ash content reduced significantly after processing (p<0.05). The processed pearl millet had significantly higher moisture content than the unprocessed (p<0.05). No significant difference in the calcium, iron and sodium content was recorded between the processed and unprocessed sorghum (p>0.05). The magnesium content decreased significantly after processing sorghum (p<0.05). However, potassium and zinc content increased significantly after processing (p<0.05). Processing pearl millet significantly increased in the calcium, potassium and zinc content (p<0.001). No significant difference in the iron content was observed between the two treatments (p>0.05). The magnesium and sodium content decreased significantly after processing (p<0.001). Processing pearl millet and sorghum significantly reduced the phytic acid content and tannin levels (p<0.05). The alkaloid content of the processed sorghum decreased significantly, while no significant difference in alkaloid content was recorded between processed and unprocessed pearl millet (p>0.05). Processing of sorghum and pearl millet is recommended since it improves the nutritional composition and lowers anti-nutritional factors.

Keywords— Fermentation, dehulling, roasting, anti-nutrients, sorghum, pearl millet

I. INTRODUCTION

Sorghum (Sorghum bicolor), and other traditional grains such as millet have the potential to alleviate chronic food insecurity in semi-arid areas because of their drought tolerance (Mukarumbwa and Mushunje 2010). One of the major impediments for adopting sorghum as a staple vis-à-vis most cereal based products is its lower nutritional status and inferior organoleptic qualities, which are attributed to the presence of anti-nutritional factors for example tannins and phytic acid (Thilakarathna, Madhusankha, and Navaratne 2022). Sorghum originated from the Northeast quadrant of Africa, in countries like Ethiopia, Eritrea, Somalia, Djibouti, Sudan and Egypt (Adebo, 2020). Nowadays the crop is being grown all over the world. Sorghum is one of the crops that grows well in most parts of Africa and in all regions in Zimbabwe. The 20th century saw an intensification of agriculture that was driven by an economic rationality. However, factors such as climate change, air pollution, lack of fresh water, and the extinction of species and habitats, have sometimes irreparably harmed the environment,
endangering the food security of many countries (Helali, Ouertani, and Dhraief 2023). Sorghum cereal is drought resistant, and it is the fifth most important cereal with regards to production after rice, wheat, maize and barley. In Sub-Saharan Africa it is the most grown cereal after maize (Eggen et al. 2019). Unlike many major cereal crops, sorghum has genetic traits with phytochemicals that have antioxidant capacity which makes them more important in alleviating cancers and other disorders (Awika and Rooney 2004).

Pearl millet (Pennisetum glaucum) another traditional grain crop, is considered an important staple diet in Africa’s arid and semi-arid regions (Traore et al. 2022). Pearl millet is drought tolerant and can grow in sandy soils with a low pH and low fertility content due to acidic nature of such soils. Pearl millet is among the best tropical cereals with superior micronutrient and protein composition and a healthy source of energy (Ogbonna, Abuajah, and Udofia 2012). Essential amino acid profile of pearl millet is richer in lysine methionine and threonine than in corn (Osman 2011a). Pearl millet possesses many anti nutritional factors including tannins, phytic acid and enzyme inhibitors. Mineral bio availability, protein and starch digestibility are impaired by the presence of anti nutritional factors in pearl millet. Various methods have been used to upgrade the nutritional status of cereal grains including processes like fermentation. Food preservation by fermentation has been employed since time immemorial. It is inexpensive, provides much needed nutrients, increase nutrient bioavailability, destroys undesirable components including natural toxins and pathogenic microorganisms, and enhances the flavour (Simatende 2016). The fermentation plays a role in the degradation of some complex compounds into simpler compounds (Masahid, Belgis, and Agesti 2021). Africa, in particular Southern Africa has continuously received less than the normal rainfall expected to give a good yield in the production of the popular staple cereals like maize and wheat (Yarnell 2008). Droughts are now more frequent than before in Africa and in order to address the issue of food insecurity, increase in production of these drought resistant crops can become a game changer to improve food security. Though the productivity and production of the crop are not increased as expected due to many limitations in the generation of demand-driven technologies and innovation up-scaling in an integrated and impact-oriented (Degefa, Abebe, and Biru 2023). Value addition of these grains can make them more attractive and increase their acceptance in the food market. The grains possess functional properties and novel products from these grains are highly likely to penetrate the market given the consumer shift towards healthy foods. Food is the most important basic need for humans to survive (Habibah, Albaar, and Rasulu 2021)

Traditional and modern methods can be employed to lower the anti-nutritional factors in small grains. Milling, soaking, fermentation, debranning, soaking and autoclaving are among the several methods and techniques used in reducing the anti-nutritional factors in small grains. By using various methods alone or in combinations, it is possible to reduce the level of anti-nutrients. (Samtiya, Aluko, Dhewa 2020). This method of coupling has not been fully investigated in developing countries.

The aim of this study was to investigate the effects, roasting, dehulling, fermentation and milling on selected minerals, protein content and anti-nutritional content of sorghum and pearl millet flour. This study is part of a larger project in investigating prospects of value addition to underutilised traditional grains in Zimbabwe.

II. METHODS

A. Process flow chart

Fig. 1. Flowchart of research

B. Sample preparation

Samples of sorghum and pearl millet were bought directly from farmers in Buhera district, Manicaland province in Zimbabwe (19.3211° S, 31.4399° East). Sorghum and pearl millet samples were sorted to remove physical foreign particles like stones and sticks. The sorghum and millet grains were dehulled using a dehuller MH200 (Kurima Agriculture products, Zimbabwe) and were subsequently fermented separately at room temperature (32 ± 2°C) for 72 hours in the presence of 0.2 % ascorbic acid. The fermentation water was decanted. The fermented grain was dried using a hot air oven at 60°C for 10 hours. The grains were then roasted using a gas roaster stove (Model LQ200, Nanyang, Heinan, China) until a roasting flavour smell could be smelt. The fermented grains were put in a miller (Ruzha brand /R70) to produce the milled sorghum and pearl millet flour. The flours were then packed in sealed plastic bags which were then stored in refrigerators for analysis.

C. Proximate analysis of the sorghum and pearl millet flour

Standard methods by the Association of Analytical Chemist (AOAC) 2016 (Paterne et al. 2019) were employed for the proximate analysis of the grains. The Kjeldahl and Soxhlet method was used for protein and fat determination, respectively. Total carbohydrate was determined by the difference method. Moisture and ash content was determined using the oven and furnace method, respectively. The reagents used were of analytical grade imported from the Sigma Aldrich (St. Lois, MO, USA).

D. Mineral analysis

The flour mineral content was measured by using standard methods of AOAC (2016). Inductively coupled plasma atomic emission spectrophotometry (ICP-OES) (Model ICAP 6000, Thermo-Fischer Scientific, Tokyo, Japan) was used to
determine the concentration of zinc, iron, calcium, potassium, sodium and magnesium content in the sorghum and pearl millet flour. Flour samples (2 g) were heated at 550 °C. The resulting ashes were dissolved in 100 ml 1 M HCl and used for mineral analysis. Zinc was determined at a wavelength of 213.9 nm, iron was determined at a wavelength of 522 nm, calcium was determined at a wavelength of 550 nm, sodium was determined at a wavelength of 589.0 nm, potassium was determined at a wavelength of 766.5 nm and magnesium was determined at a wavelength of 610 nm. Standard solutions were prepared using analytical grade reagents and then serial diluted.

D. Phytic acid determination

The method of Haug and Lantzsch (1983), modified by Onyango et al. (2005) was used to measure phytates. Samples of weight 2 g were put into 250 ml conical flask and 100 ml of 2 % HCl was added until soaking of the samples was achieved in the conical flasks for a period of 3 hours. The solution was then filtered through a double layered hardened filter paper. Each respective filtrate (50 ml) was placed in 0.50 ml measuring conical flask and 107 ml of distilled water was added in each case to give proper acidity. Ammonium thiocyanate (NH4SCN) solution (10 ml of 0.3 %) was added into each solution. The solutions were then titrated with standard iron (III) chloride solution containing 0.00195 g Fe per ml. A slightly brownish-yellow colour which persisted for 5 minutes was denoted as the end point. The percentage of phytic acid was calculated using the formula:

\[
\text{Percentage Phytic Acid} = \frac{\text{Titre value} \times 0.00195 \times 1.19 \times 100 \times 3.55}{\text{Wt. of sample}}
\]

E. Alkaloid’s determination

The alkaloid content was determined gravimetrically (Haborne, 1973). Sample of weight 5 g were weighed using a weighing balance and put into 50 ml of 10 % acetic acid solution in ethanol. The solution was adequately shaken and left to stand for 4 hours before filtration. Evaporation of the filtrate to a quarter of the original volume was done on a hot plate. Dropwise discharge of concentrated ammonium hydroxide was added to precipitate the alkaloids. Previously weighed filter papers were used to precipitate the alkaloids, with a facilitated washing using 1% ammonium hydroxide solution. Drying of the filter papers containing the filtrate was done in an oven at a temperature of 60 °C for a duration of 30 minutes. The dried filter papers were then transferred into desiccators and allowed to cool. Reweighing of the filter papers was done until a constant weight was recorded. Alkaloid content was expressed as the difference between the filter paper containing the residue and the recorded weight of the unused filter paper, expressed as a percentage of the sample weight analyzed. Three runs were done for each grain sample and average readings were recorded for each experiment.

F. Tannin determination

The vanillin-HCl method of Price and Butler (1977) using spectrophotometer (Thermo Scientific, Model Evolution 300/600, London) at 500 nm, was used to determine the tannin content. The tannin content was expressed as catechin equivalent using a standard curve.

G. Data analysis

Data were tested for normality using Shapiro Wilk Test. All data passed the normality test. Means on proximate, micronutrient and anti-nutritional composition of the treatments were compared using ANOVA in Genstat Version 18. Where significant differences were observed the means were separated using Turkey’s Test at 95% probability

III. RESULTS

Sorghum grain had an average protein, ash, fat, moisture and carbohydrate content of 15.0±0.08, 3.6±0.05, 8.5±0.09 and 69.0±0.08, respectively. Significant differences in the proximate composition was recorded after processing (dehulling, fermenting and roasting) sorghum (p<0.05) (Table 1). Sorghum processing i.e. dehulling, fermenting and roasting significantly increased the protein and ash content (p<0.05), from 15 to 20% and 3.6 to 4.2%, respectively. No significant difference in fat content was observed between the processed and unprocessed sorghum (p>0.05). The dehulled, fermented and roasted sorghum had significantly higher moisture content (11.0±1.78) than the unprocessed sorghum (8.5±0.09) (p<0.05) (Table 1).

<table>
<thead>
<tr>
<th>TABLE 1: EFFECT OF FERMENTATION, DEHULLING AND ROASTING ON PROXIMATE COMPOSITION OF SORGHUM FLOUR.</th>
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<tr>
<td>Treatments</td>
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<tr>
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<tr>
<td>Sorghum P</td>
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<tr>
<td>Sorghum U</td>
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<td>P value</td>
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<td>LSD</td>
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<td>CV (%)</td>
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Means followed by different superscript letters in a column are significantly different at p <0.05. Key: P - Processed (dehulling, fermenting and roasting) and U – Unprocessed (dried sorghum)

Pearl millet grain had mean protein, ash, fat, moisture and carbohydrates content of 20±0.07, 3.5±0.08, 4.8±0.04, 8.2±0.07 and 69.0±3.0%, respectively. Processing pearl millet i.e. dehulling, fermenting and roasting resulted in significant changes in the proximate composition (p<0.05) (Table 2). The protein content significantly increased after dehulling, fermenting and roasting pearl millet from 20 to 25% (p<0.05). Significant reduction in carbohydrate and ash content was recorded after processing pearl millet (p<0.05) from 69 to 64% and 3.5 to 1.1%, respectively. No significant change in the fat content was recorded after processing the pearl millet (p>0.05). The processed pearl millet treatment had significantly higher moisture content than the unprocessed treatment (p<0.05) (Table 2).
Sorghum grain had the following levels of selected minerals in mg/100g: Ca (29.0±0.008), Mg (1.4±0.01), K (21.0±0.004), Fe (0.7±0.01), Na (202.0±0.13) and Zn (0.06±0.01). There were significant differences between the micronutrient composition of processed and unprocessed sorghum (p<0.001) (Table 3). No significant difference in the calcium, iron and sodium content was recorded between the processed and unprocessed sorghum (p>0.05). The magnesium content significantly reduced after processing sorghum (p<0.001), from 21 to 111% and 0.06 to 0.47%, respectively (Table 3). Processing sorghum and pearl millet i.e. dehulling, fermenting and roasting significantly reduced the phytic acid content (p<0.05), from 0.66 to 0.41% and 1.32 to 0.74%, respectively (Fig. 2). Sorghum had the following mean mineral content in mg/100g: Ca (210±0.002), Mg (98.0±0.002), K (244±0.009), Fe (2.2±0.21), Na (386.0±0.013) and lastly Zn (0.32±0.05). The micronutrient composition of processed and unprocessed pearl millet varied significantly (p<0.05) (Table 4). Processing resulted in a significant increase in the calcium, potassium and zinc content (p<0.001) from 210 to 300 mg/100g, 244 to 261mg/100g and 0.32 to 4.2 mg/100g, respectively. No significant difference in the iron content was recorded between the two treatments (p>0.05). The magnesium and sodium content decreased significantly after processing (p<0.001) (Table 4). Processing pearl millet and sorghum had the following levels of selected minerals in mg/100g: Ca (210±0.002), Mg (98.0±0.002), K (244±0.009), Fe (2.2±0.21), Na (386.0±0.013) and lastly Zn (0.32±0.05). The micronutrient composition of processed and unprocessed pearl millet varied significantly (p<0.05) (Table 4). Processing resulted in a significant increase in the calcium, potassium and zinc content (p<0.001) from 210 to 300 mg/100g, 244 to 261mg/100g and 0.32 to 4.2 mg/100g, respectively. No significant difference in the iron content was recorded between the two treatments (p>0.05). The magnesium and sodium content decreased significantly after processing (p<0.001) (Table 4).
IV. DISCUSSION

We set out to investigate the effect of dehulling, fermentation and roasting on nutrient and anti-nutrient content of sorghum and millet flour. In overall dehulling, fermentation and roasting of sorghum and pearl millet grains improved the nutritional content and lowered the anti-nutritional content significantly. In a similar study by Prospects. (2020), the ash content and protein content of untreated sorghum were lower than those recorded in this study. The ash content was 4.2% and protein content 20% while their recorded values were 1.98% and 9.35%, respectively. Badi. (2004) obtained the following values; ash, 52%, carbohydrates 71.8%, fat 4.50%, protein 14.86%. Gerrano et al. (2014) recorded 1.44% ash, 3.32% fat, and 9.95% protein content. Variations in proximate analysis can arise due to use of different methods, temperatures and other experimental exposures. Another possible reason for the differences in the observed results could be attributed to different climatic conditions exposed to the crops because of different locations. We used official AOAC methods and our results compare well with studies by other researcher. However, from all these studies it was observed that dehulling, roasting and milling improved the ash and protein content of the sorghum flour although the fat and carbohydrate content were lowered.

It can be suggested that some minerals like Zn and Na are leached during fermentation. Fe and Mg increased during fermentation; this could be due to the decrease in binding substances like phytic acid. The observed mineral content after processing is showing some positive results, giving high prospects of having sorghum and pearl millet flours as competent ingredients to be in cooperated in many nutritious foods. The obtained range of phytic acid percentage of 1.32% in non-fermented sorghum is in agreement with the levels found by Ogbonna, Abuajah, and Udofia. (2012) where they found a range between 0.88% to 2.21% in four different sorghum cultivars from Sudan. Reduction in phytic acid content after fermentation was also noted for sorghum in the same study, a reduction by 36% was noted after 72 hours while, in the current study, a reduction of 56% after 72 hours in pearl millet was observed. The increase in protein content after fermentation has been attributed to the increase in nitrogen content released when microorganisms utilise carbohydrates for energy (Province and Sciences 2017). A similar observation was reported by Pranoto. (2013), where proteolysis of protein during fermentation resulted in production of peptides and amino acids. The decrease in carbohydrate content in all treatments can be attributed to the fermentation process, microbes use energy and nutrients in their activities during the fermentation process hence a decrease in carbohydrates is imminent as they are the core source of energy. The lowering of pH during fermentation is responsible for the reduction of phytic acid. The optimal pH for cereal phytase is around 5.0, where phytase will have high activity during fermentation (Osman 2011b). At this low pH, phytase activity hydrolyses phytic acid to inorganic phosphate and inositol (Towo, Matuschek, and Svanberg 2006). The enzyme microbial phytase produced by microorganisms helps to hydrolyse phytic acid during the fermentation process which then accounts for the lowering of phytic acid in the fermented products.

The decrease in phytic acid and tannins in roasted, fermented and dehulled sorghum and pearl millet flours is a welcome development to the food industry as these flours can now be processed and in cooperated as ingredients in novel foods. This was not possible before because they were considered nutritionally inferior (Rooney and Miller 1982). Baby foods which meet the nutritional demands can now be formulated...
using these ingredients (Deribe and Kassa 2021). Products like pastas and soups can also be formulated using these ingredients (Alavi, Mazumdar, and Taylor 2018).

We used the titrimetric method in the determination of anti nutrients namely phytic acid. The advantages of the titrimetric method used in phytic acid determination over the spectrometric method is that it is a simple procedure and has got low costs as compared to spectrophotometric methods. The major setback in this method is that there are changes of inconsistent molar ratios and also interferences with other inositols but this also applies to spectrophotometric methods. The use of sensors (electrochemical biosensors and fluorescence probes) is extremely expensive (Bizzotto et al. 2018).

The coupling method of combining dehulling, fermentation and roasting was done without investigating the effectiveness of the individual processes on the reduction of anti nutrition factors and increase in mineral availability. This limits knowledge on how effective each method is in reducing anti nutrients and in increasing mineral availability. We therefore, recommend further studies investigating the individual effect of each processing method.

V. CONCLUSION

Pre-treatment of sorghum and pearl millet by the methods of dehulling, fermentation and roasting increased the protein, moisture, fat and ash content of these grains. Pre-treatment of the grains proved to be a viable way to increase the nutrient content hence can be a positive way to help eliminate some malnutrition related disorders in populations that mainly rely on these grains as a main source of carbohydrates. The decrease in anti-nutrients which are undesirable such as alkaloids, tannins and phytic acid after pre-treatment of sorghum and pearl millet, proves that coupling of these processing methods is effective in enhancing the nutritional value of sorghum and pearl millet, coining of policy on the encouragement of food processors involved with these grain is recommended.

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