



## Micro-Mineral Retention and Anti-Nutritional Compounds Degradation during Bean Cooking Process

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### Abstract

The objective of this study was to determine the effect of soaking, boiling and frying on retention of micro-minerals as well as degradation of polyphenols and phytic acids. Soaking of the beans did not significantly ( $p > 0.05$ ) decrease mineral, total polyphenol and phytic acid content of beans. While boiling of beans significantly ( $p < 0.05$ ) decreased the concentration of total polyphenols and phytic acid but not the mineral content of the beans. Frying of boiled beans decreased the total polyphenol content but increased the concentration of phytic acids in beans. Data obtained in this study indicates that cooking of beans without discarding of soaking water and broth results into greater retention of minerals but frying may be detrimental to mineral bioavailability because it leads to increase in phytic acid content.



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### Introduction


The common dry bean, *Phaseolus vulgaris* L., is a food legume grown and consumed in many countries. In developing countries like Rwanda, beans are primary sources of proteins as they supply 65% of proteins compared to 4% from animal sources<sup>1</sup>. Beans also contribute to a large amount of energy, 32% and micronutrients (iron, zinc, pro-vitamin A, folic acid and pyridoxine) that promote normal human body growth and development<sup>2,3</sup>. About 2

billion people (over 30% of the world's population) are anemic, mainly due to iron deficiency, and in resource-poor areas, this is frequently exacerbated by infectious diseases<sup>4</sup>. Due to high prevalence of iron and zinc deficiencies in many parts of the developing world, efforts are being made through bio-fortification to develop high iron and zinc containing bean varieties. Iron deficiency decreases mental and psychomotor development in children, increases both morbidity and mortality in young

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children and mothers and, diminish work performance and decrease resistance to infection<sup>5,6,7</sup>. Major health problems associated with zinc deficiency include; (i) impairment of brain function and mental development, (ii) disturbances of the immune system and susceptibility to deadly infectious diseases and (iii) delays in physical development especially stunted growth<sup>8</sup>. Lack of micronutrients such as vitamin A, zinc and iron affect at least a half of the world's population<sup>9</sup>. Insufficient intake, consumption of low nutrient dense foods and poor bioavailability has been attributed to these mineral micronutrient deficiencies. The most affordable way of combating Fe and Zn malnutrition in impoverished populations is to consume micronutrients rich foods and beans have been identified as a suitable food to combat iron and zinc deficiency.

Beans are a source of anti-nutritional compounds such as polyphenols, enzyme inhibitors (trypsin inhibitors), phytic acid and oligosaccharides such as raffinose and stachyose<sup>10</sup>. Phytate (inositol phosphates), a common constituent of cereals and legumes, is regarded as an anti-nutrient because of their ability to complex with mineral micronutrient leading to insoluble mineral-phytate complexes<sup>11</sup>. As phytate is not utilized by non-ruminants including humans, there is a risk of inadequate mineral supply when consuming legume based diets<sup>12,13</sup>. Polyphenols also hinder bioavailability of nutrients in beans especially tannins have strong ability to form complexes with minerals<sup>14</sup>.

Besides development of high iron and zinc containing bean varieties, there is need to assess the retention of iron and zinc during the cooking process since the cooking method used may lead to loss of minerals. There could also be an increase in mineral content during the cooking of beans since in most instances spices, cooking oil, tomatoes and other ingredients are added to the beans. Therefore, the objective of this study was determine the effects of soaking, boiling, re-boiling, frying, and refrying on retention of minerals especially iron, zinc, and degradation of total polyphenol and phytic acid content of beans.

## **Material and Methods**

### **Description of Samples**

Four different batches of carioca bean varieties were sampled and colour coded pink, white, blue and

green. All bean samples had the same phenotypic characteristics (size, colour and shape). Samples coded pink and white were semi climbing carioca landrace bean varieties (G4825) originally obtained from Brazil but cultivated in Columbia. While samples coded blue and green were a mixture of 7 different carioca bean lines namely; SMC 33, SMC 34, SMC 41, SMC 44, SMC 45, SMC 109 and SMC 111 bred at International Center for Tropical Agriculture (CIAT) headquarters in Bali, Columbia. Samples were wholesome (undamaged), of uniform size, free of extraneous matter such as plant debris and stones and meet the criteria for dry beans intended for human consumption as per the East African Standards<sup>15</sup>.

### **Bean Cooking Process**

For each batch of beans, 11 kg was weighed and washed with mineral free water at least three times. Two different methods were used for cooking the beans (Fig 1). Batches colour-coded pink and white were cooked using the first method while batches colour-coded blue and green were cooked using the second method. In the first method the beans were boiled without prior soaking while in the second method beans were boiled after soaking. The soaking water was not discarded but used in the bean boiling process. The boiling time of beans colour coded white and pink was 3.5 h while for the batches coded blue and green was 4 hours. The boiled beans were divided into two portions; one portion was fried and consumed immediately for lunch and the other portion was kept for consumption during dinner. In bean frying, the cooking oil was heated up in a sauce pan, together with onions, carrots, seasoning agent (Maggie®; Nestle, Nairobi, Kenya), celery, and green pepper until the colour of the mixture turned chocolate brown. Boiled beans were added to the fried ingredients and the combination of the fried ingredients and beans was boiled for further 2 minutes while stirring continuously. The quantities of ingredients used during frying process were as follows; cooking oil 450 ml, onions 450 g, celery 225 g, green pepper 450 g and seasoning agent 16 g. After about 5 hours, the portion for dinner was re-boiled and sub-divided into two portions; one portion was served directly for dinner while the other portion was fried before serving. These procedures were conducted in order to depict the actual bean cooking and consumption systems in East and

Central Africa. The water used for washing of beans, cooking and final rinsing of utensils was distilled and de-ionized (mineral free grade). Cooking materials were of stainless still grade or materials that cannot induce mineral contamination of the beans.

#### **Sample Preparation**

About 50 g of raw, boiled, re-boiled and fried bean samples were dried using a freeze dryer (Lyotrap Freeze-Drying Machine, Greenfield-Oldham, England). Dried bean samples were finely ground into powder form using a domestic grinder (Philips, Model No. 2161, Netherlands). The grinder was thoroughly cleaned, rinsed with distilled and de-ionized water and dried to prevent carry over contamination in between the subsequent grinding process. The bean powders were then transferred into factory clean and airtight air tubes (BD Biosciences, Belgium). The tubes were placed inside air tight zip lock bags and stored at -20°C until use.

#### **Determination of Moisture Content**

Moisture content of raw, boiled and fried bean samples was determined using the oven drying method<sup>16</sup>.

#### **Determination of Minerals**

For each sample, 10 g was dried in an oven at 70°C for 18 hours to eliminate moisture. Bean powder (0.25 g) was weighed in a digestion tube. Then 2.5 ml of an acid mixture of nitric acid (70%; Sigma-Aldrich, St Louis, USA) and perchloric acid (70%; Sigma-Aldrich, St Louis, USA) in a ratio of 2:1 was added to test tube. Digestion tubes containing samples and acid mixture were then inserted into a block heater (Grant type BT5D, Cambridge Instruments, UK). Initial heating was conducted at 60°C for 15 min and then the temperature was increased to 120°C for 75 min. Samples were then cooled to room temperature, filtered using a filter paper of diameter 125 mm (Whatman® No.1; Maidstone, England) and diluted to 25 ml with mineral free water. Minerals analysed included; calcium, copper, iron, potassium, magnesium, manganese, phosphorous, selenium and zinc. The minerals were quantified using inductively coupled plasma atomic emission spectroscopy (Shimadzu, Japan). A wheat sample obtained from National Institute of Standards and Technology (NIST; USA) with known concentrations of iron and zinc was

used to ascertain the accuracy and compatibility of the mineral analysis methods used in this study. A Bean sample with known concentration of iron and zinc was also included in each analysis to check for variations in measurements conducted at different times. Furthermore, the mineral extractability from the samples was ascertained by spiking quality control samples with a known concentration of iron and zinc and checking if the data obtained accurately depicts the quantity added. Standards solutions of the minerals were prepared at 0.0, 0.2, 0.6, 0.8 and 1.0 mg/Kg. The concentrations of minerals in the various beans were calculated using the standard curve data.

#### **Determination of Total Polyphenols**

##### **Preparation of Bean Extracts**

Extracts from raw bean samples were prepared using acidified methanol (1% conc. HCl in methanol) hereafter referred to as solvent. Each sample (0.12 g) was extracted with 12 ml solvent in three phases as follows: 4 ml of solvent was added to 0.12 g of the sample in a beaker covered with aluminium foil. The sample was stirred for 1 hour, transferred to a 14 ml plastic centrifuge tube, centrifuged at 3500 rpm for 10 min (25°C) and decanted, keeping the supernatant. The sample residue was rinsed again with 4 ml of the solvent, stirred for 15 min, centrifuged again as above and decanted, keeping the supernatant. The residual polyphenols in the pellet was re-extracted as described above. The supernatants were combined and kept at 4 °C until analysis.

##### **Determination of Total Phenolic Content (TPC) of Beans**

The total phenolic content in bean extracts was determined using the Folin-Ciocalteu method [17] as described by Waterman and Mole<sup>18</sup>. Bean methanolic extract (0.1 ml) or 0.1 ml gallic acid solution in acidified methanol (0.0 - 1.0 mg/ml) (used as a standard) was added to a 10 ml plastic tube containing 2 ml distilled water. Folin-Ciocalteu's phenol reagent (0.5 ml) was added and after 2 min, 1.5 ml of 20% (w/v) sodium carbonate solution was added. The content was mixed and made up to volume (10 ml) with deionised water. The volumetric flask was then covered and mixed thoroughly by inverting several times and allowed to stand for 2 hours from time sodium carbonate was added.

The absorbance was measured at 760 nm using a UV/Visible Spectrophotometer (M501 Single beam scanning UV/vis spectrophotometer: Camspec, Cambridge, UK). Results were expressed as gallic acid equivalents (CE, mg catechin equivalents/g sample).

### Determination of Phytic Acid

#### Extraction of Phytic Acid

For each sample, 0.2 g of bean powder was transferred into a 100 ml beaker followed by addition of 10 ml of 0.5 N HCl. The mixture was stirred using a magnetic stirrer for 1 hour, then filtered using a filter paper. About 1.5 ml of the filtrate was transferred into a micro-centrifuge tube and centrifuged for 15 min, 4000 rpm at room temperature (Sigma Laborzentrifugen, Model No. 1-14, Germany). The supernatants containing the phytic acid extracts were either used immediately or stored at -20°C for later use.

#### Analysis of Phytic Acid

The method described by Dost and Tokul19 was used with some modifications. Iron (III) thiocyanate solution was prepared by mixing 25 ml of 100 µg/ml iron (III) chloride solution and 25 ml of 500 µg/ml ammonium thiocyanate, followed by addition of concentrated HCl (200 µl). The volume of the mixture was adjusted to 100 ml by addition of distilled water. Standard solutions of 0, 10, 25, 50, 75, 100, and 125 mg/L were prepared. Standard solutions at each concentration (1ml) were

transferred into test tube followed by addition of 2 ml of iron (III) thiocyanate solution. After mixing, 1.5 ml was transferred into micro-centrifuge tubes, stirred in water bath at 40°C for 2.5 hours, cooled to room temperature and then centrifuged at 4000 rpm for 15 min. The prepared solutions (20 µl) were injected on HPLC fitted with a UV-vis detector (Shimadzu LC-20AD, Japan). The mobile phase used was a mixture of 2 solutions: 30% acetonitrile (Sigma-Aldrich, St Louis USA) in water containing 0.1 M of nitric acid (HNO<sub>3</sub>) and 25% methanol in which 75% was distilled water. Flow rate was 1 ml/min and detection was done at 460 nm. A reverse-phase C-18 column (Nucleosil 100-5; Chromatographie Service GmbH, Germany) was used.

#### Statistical Analysis

Each determination was conducted on three separate samples and analyzed in triplicate. Average values for each parameter was obtained. The data obtained from the experiments were analyzed using one-way ANOVA with the aid of statistical computer software (SPSS Inc. Released 2007, SPSS for windows version 16.0, Chicago, SPSS Inc.)

### Results and Discussions

#### Retention of Minerals during the Cooking Process

The concentration of minerals in the different batches of beans as well as the effect of cooking procedures such as soaking, boiling and frying on retention of minerals is shown in Table 1.

Table 1: Mineral content of raw, soaked, boiled and fried beans AB

Bean Type	Treatment	Mineral concentration(mg Kg <sup>-1</sup> )								
		Ca	Cu	Fe	K	Mg	Mn	P	Se	Zn
Pink-Normal bean	Raw	1139.1±	6.2±	46.8±	11174.0±	1241.7±	14.8±	4159.1±	0.1±	31.2±
		93.3 <sup>a</sup>	0.5 <sup>a</sup>	4.3 <sup>a</sup>	723.0 <sup>a</sup>	98.1 <sup>a</sup>	1.3 <sup>a</sup>	327.7 <sup>a</sup>	0.0	3.1 <sup>a</sup>
	Boiled for lunch	1125.6±	6.4±	49.8±	10246.1±	1260.2±	14.6±	4046.3±	nd*	32.5±
		23.3 <sup>a</sup>	0.2 <sup>a</sup>	3.0 <sup>a</sup>	351.0 <sup>a</sup>	35.6 <sup>a</sup>	0.5 <sup>a</sup>	146.2 <sup>a</sup>		0.9 <sup>a</sup>
	Boiled for dinner	1125.3±	6.3±	54.1±	10863.1±	1282.0±	15.0±	4239.5±	nd	33.4±
		11.8 <sup>a</sup>	0.2 <sup>a</sup>	13.0 <sup>a</sup>	273.1 <sup>a</sup>	42.9 <sup>a</sup>	0.4 <sup>a</sup>	87.3 <sup>a</sup>		1.2 <sup>a</sup>
Boiled and Fried for lunch	1067.7±	5.8±	56.0±	10254.0±	1232.5±	13.9±	3970.2±	nd	31.3±	
	85.3 <sup>a</sup>	0.5 <sup>a</sup>	1.5 <sup>a</sup>	321.8 <sup>a</sup>	67.9 <sup>a</sup>	0.6 <sup>a</sup>	204.4 <sup>a</sup>		1.3 <sup>a</sup>	
Boiled and Fried for dinner	1026.4±	6.0±	59.3±	10324.7±	1159.7±	14.3±	3984.6±	nd	32.1±	
	15.8 <sup>a</sup>	0.0 <sup>a</sup>	8.3 <sup>a</sup>	252.2 <sup>a</sup>	13.9 <sup>a</sup>	0.2 <sup>a</sup>	96.1 <sup>a</sup>		1.0 <sup>a</sup>	
White-	Raw	1168.7±	6.6±	54.3±	11744.6±	1315.4±	15.8±	4479±	0.1±	33.3±

Normal		75.8 <sup>a</sup>	0.3 <sup>a</sup>	5.2 <sup>a</sup>	405.9 <sup>a</sup>	68.5 <sup>a</sup>	0.6 <sup>a</sup>	129.7 <sup>a</sup>	0.0	1.6 <sup>a</sup>
bean	Boiled for lunch	1127.9±	6.0±	52.5±	8954.9±	1139.2±	14.5±	3815.9±	nd	31.9±
		57.1 <sup>a</sup>	0.6 <sup>a</sup>	3.1 <sup>a</sup>	676.6 <sup>b</sup>	83.2 <sup>a</sup>	1.0 <sup>ab</sup>	302.9 <sup>a</sup>		1.9 <sup>a</sup>
	Boiled for dinner	966.3±	5.3±	42.4±	10008.4±	1131.1±	12.9±	3805.9±	nd	29.5±
		44.7 <sup>a</sup>	0.1 <sup>a</sup>	7.2 <sup>a</sup>	149.3 <sup>ab</sup>	21.2 <sup>a</sup>	0.3 <sup>b</sup>	49.6 <sup>a</sup>		0.6 <sup>a</sup>
	Boiled and Fried for lunch	1133.3±	6.4±	57.8±	11163.8±	1301.6±	14.7±	4356.9±	nd	34.1±
		44.7 <sup>a</sup>	0.3 <sup>a</sup>	2.0 <sup>a</sup>	415.9 <sup>a</sup>	61.8 <sup>a</sup>	0.7 <sup>ab</sup>	191.4 <sup>a</sup>		1.4 <sup>a</sup>
	Boiled and Fried for dinner	1089.5±	6.2±	58.6±	11101.3±	1276.3±	14.6±	4182.5±	nd	33.1±
		56.2 <sup>a</sup>	0.4 <sup>a</sup>	3.7 <sup>a</sup>	551.2 <sup>a</sup>	107.7 <sup>a</sup>	0.4 <sup>ab</sup>	154.7 <sup>a</sup>		2.0 <sup>a</sup>
Blue-Biofortified	Raw	1017.0±	9.8±	88.2±	12379.3±	1274.5±	17.6±	5374.8±	0.1±	42.9±
		57.3 <sup>ab</sup>	0.3 <sup>ab</sup>	3.5 <sup>a</sup>	118.8 <sup>a</sup>	47.3 <sup>ab</sup>	0.8 <sup>a</sup>	164.2 <sup>a</sup>	0.0	1.6 <sup>a</sup>
bean	Soaked	1148.3±	11.9±	83.8±	11651.4±	1341.1±	17.6±	5043.7±	nd	43.6±
		101.5 <sup>b</sup>	0.5 <sup>a</sup>	1.8 <sup>a</sup>	129.1 <sup>ab</sup>	16.1 <sup>a</sup>	0.4 <sup>a</sup>	42.4 <sup>ab</sup>		0.0 <sup>a</sup>
	Boiled for lunch	1058.4±	9.7±	77.8±	11545.8±	1264.6±	18.3±	4992.7±	nd	44.2±
		69.3 <sup>ab</sup>	0.5 <sup>ab</sup>	8.0 <sup>ab</sup>	531.5 <sup>ab</sup>	79.6 <sup>ab</sup>	0.8 <sup>a</sup>	164.5 <sup>ab</sup>		1.8 <sup>a</sup>
	Boiled for dinner	884.0±	9.4±	73.5±	10160±	1041.3±	15.9±	4513.5±	nd	39.3±
		19.6 <sup>ab</sup>	0.7 <sup>b</sup>	0.6 <sup>ab</sup>	258.6 <sup>bc</sup>	26.1 <sup>bc</sup>	0.0 <sup>a</sup>	109.9 <sup>ab</sup>		2.1 <sup>a</sup>
	Boiled and Fried for lunch	896.1±	10.5±	72.8±	9384.6±	989.1±	16.2±	4219.8±	nd	38.6±
		28.1 <sup>ab</sup>	0.4 <sup>ab</sup>	3.1 <sup>ab</sup>	215.1 <sup>c</sup>	13.9 <sup>c</sup>	0.1 <sup>a</sup>	65.4 <sup>b</sup>		0.2 <sup>a</sup>
	Boiled and Fried for dinner	847.1±	7.1±	66.8±	9938.3±	993.4±	15.1±	4202.1±	nd	37.4±
		89.1 <sup>b</sup>	0.8 <sup>a</sup>	4.6 <sup>b</sup>	964.9 <sup>bc</sup>	113.2 <sup>c</sup>	1.6 <sup>a</sup>	488.3 <sup>b</sup>		4.7 <sup>a</sup>
Green-Biofortified	Raw	1036.6±	9.6±	85.0±	11998.6±	1265.6±	16.7±	5462.9±	0.2±	40.7±
		55.7 <sup>b</sup>	0.7 <sup>a</sup>	5.0 <sup>a</sup>	745.2 <sup>a</sup>	52.8 <sup>a</sup>	0.9 <sup>a</sup>	406.3 <sup>a</sup>	0.2	1.4 <sup>a</sup>
bean	Soaked	1027.1±	9.6±	80.2±	10846.5±	1246.7±	15.8±	4841.2±	nd	40.6±
		36.9 <sup>a</sup>	0.1 <sup>a</sup>	6.2 <sup>a</sup>	664.1 <sup>ab</sup>	99.0 <sup>a</sup>	1.2 <sup>a</sup>	339.2 <sup>a</sup>		1.9 <sup>a</sup>
	Boiled for lunch	1064.7±	8.7±	76.9±	9724.6±	1122.8±	15.8±	4552.9±	nd	40.3±
		142.2 <sup>a</sup>	1.0 <sup>ab</sup>	4.9 <sup>a</sup>	1039.2 <sup>ab</sup>	145.3 <sup>ab</sup>	1.6 <sup>a</sup>	557.3 <sup>a</sup>		4.2 <sup>a</sup>
	Boiled for dinner	1045.8±	9.7±	78.8±	10500.9±	1147.4±	16.6±	4983.1±	nd	42.2±
		48.3 <sup>a</sup>	0.4 <sup>a</sup>	5.7 <sup>a</sup>	259.4 <sup>ab</sup>	28.2 <sup>ab</sup>	0.9 <sup>a</sup>	239.7 <sup>a</sup>		2.3 <sup>a</sup>
	Boiled and Fried for lunch	923.9±	8.5±	73.9±	9861.8±	1056.3±	15.4±	4471.0±	nd	37.4±
		51.0 <sup>a</sup>	0.3 <sup>ab</sup>	3.3 <sup>b</sup>	295.0 <sup>ab</sup>	69.4 <sup>ab</sup>	0.8 <sup>a</sup>	282.2 <sup>a</sup>		2.1 <sup>a</sup>
	Boiled and Fried for dinner	848.6±	7.1±	67.6±	9019.3±	908.0±	14.0±	4060.2±	0.1±	34.5±
		52.0 <sup>a</sup>	0.3 <sup>b</sup>	3.0 <sup>a</sup>	123.7 <sup>ab</sup>	40.3 <sup>c</sup>	0.9 <sup>a</sup>	219.5 <sup>a</sup>	0.0	2.1 <sup>a</sup>

Values are given as mean±SD (n=3)

<sup>A</sup> Data marked by the same superscript letters were not significantly different ( $p < 0.05$ )

<sup>B</sup> nd = not detected

Bean batches coded pink and white had iron concentrations at 46.8 and 54.3 mg/Kg respectively while batches coded blue and green had iron concentration of 88.2 and 88.5 mg/Kg respectively. On the basis of our results bean batches coded pink and white can be considered as low iron varieties while batches coded blue and green can be considered as high iron varieties. According to Ugen *et al.*,<sup>3</sup>, bean varieties with Iron (above 70 mg/kg) and Zinc (above 30 mg/kg) above the average nutrient content for beans of 50 mg/kg and 20 mg/kg of Fe and Zn, respectively are considered high iron and zinc

containing bean varieties. Bean samples coded pink and white had zinc concentrations of 31.2 and 33.3 mg/kg respectively. Whereas samples coded blue and green had iron concentrations corresponding to 42.9 and 40.7 mg/kg respectively.

Boiling, re-boiling and frying did not significantly ( $p > 0.05$ ) reduce the concentrations of Ca, Cu, Fe, K, Mg, Mn, P, and Zn in beans cooked without prior soaking. Selenium was detected at concentrations at or below 0.2 mg/kg in raw beans. While in bean samples coded pink, white and blue Se was

completely not detected after boiling, re-boiling and frying. Soaking did not significantly ( $p > 0.05$ ) decrease the concentration of all minerals tested in this study. The bean cooking duration can be long due to the hard to cook phenomena therefore soaking of beans prior to cooking is done to reduce on the cooking time and save on energy required for bean cooking. Addition of onions, cooking oil, celery, green paper and seasoning agent during the frying process did not significantly ( $p > 0.05$ ) increase the mineral content of the beans mainly because of small quantities used and in case of cooking oil minerals are normally removed during the oil refining process.

The retention of iron during the cooking process was 78.1, 83.8 and 92.7% in the white, blue and green batches of beans, respectively indicating that very little loss of iron occurred during the cooking process. Although loss of iron during soaking process ranged 1.8 to 4.8 mg/Kg, statistical analysis showed that soaking of the beans prior to boiling did not significantly ( $p > 0.05$ ) decrease in the iron and zinc content of the beans. Moreover Sebastia *et al.*,<sup>20</sup> reported losses of calcium, iron and zinc at 32.9, 31.05 and 22.53% respectively during traditional cooking. Similarly, Meiners *et al.*,<sup>21</sup>

reported losses ranging from one third to one half of values of minerals in raw legumes. In this study the experiment was designed to depict the actual cooking practices used in Rwanda and many parts of East and Central Africa. However, in the traditional cooking process, heat applied to the cooking vessel is not controlled causing froth formation and overflow of broth which could also lead to loss of minerals. Bean broth was reported to contain between 1.24 to 3.35 mg/100g of iron and 0.300.71 mg/kg of zinc<sup>21</sup>. In preliminary study that we conducted the broth was found to contain 51.3 to 114.8 mg/kg of iron and 6.6 to 24 mg/kg of zinc. Therefore, control of overflow of cooking water and or broth may be a key method of preventing loss of minerals during cooking. In this study, the boiling process was controlled to prevent loss of cooking water and broth. On an overall basis, the cooking process used in this study resulted into greater retention of iron and zinc as well as other minerals.

#### Effect of Cooking Conditions on Total Polyphenols in Beans

The effect of cooking conditions such as soaking, boiling and frying on total polyphenol contents of beans are shown in Table 2. Total phenolic content ranged from 3.15-7.31 mg GAE/g sample. .

**Table 2: Total phenolic contents of raw, soaked, boiled and fried beans<sup>AB</sup>**

Treatment	Bean samples			
	Pink	Blue	White	Green
Raw	6.50 ± 0.29 <sup>a</sup>	7.31 ± 0.29 <sup>a</sup>	6.40 ± 0.29 <sup>a</sup>	6.29 ± 0.43 <sup>a</sup>
Soaked	ND	6.19 ± 0.43 <sup>a</sup>	ND	7.06 ± 0.43 <sup>a</sup>
Boiled for lunch	3.71 ± 0.50 <sup>bc</sup>	3.35 ± 0.14 <sup>b</sup>	3.15 ± 0.00 <sup>c</sup>	3.30 ± 0.07 <sup>b</sup>
Boiled for dinner	3.05 ± 0.29 <sup>c</sup>	3.25 ± 0.14 <sup>b</sup>	3.85 ± 0.00 <sup>bc</sup>	3.40 ± 0.93 <sup>b</sup>
Boiled and fried for lunch	4.87 ± 0.00 <sup>b</sup>	4.37 ± 0.29 <sup>b</sup>	4.62 ± 0.36 <sup>b</sup>	5.38 ± 1.01 <sup>ab</sup>
Boiled and fried for dinner	4.37 ± 0.43 <sup>bc</sup>	4.16 ± 0.43 <sup>b</sup>	4.82 ± 0.36 <sup>b</sup>	5.08 ± 0.14 <sup>ab</sup>

Values are given as mean ± SD (n = 3); ND = not done

<sup>A</sup> Data marked by the same superscript letters were not significantly different ( $p < 0.05$ )

<sup>B</sup> Results are expressed as milligrams gallic acid equivalent per sample

The results in the present study are in agreement with those reported in legumes by Amarowicz and Pegg<sup>22</sup>. The authors reviewed that the total phenolic

content of green and yellow peas, lentils, red kidney and black beans ranged from 1.07-7.53 mg GAE/g sample. In comparison to data obtained by Akond



*et al.*,<sup>23</sup> in which 29 bean varieties were studied; the total polyphenol concentration ranged from 5.87 to 14.14 mg g<sup>-1</sup> GAE which points out that the total polyphenol contents of beans used in this study was low. A lower amount of total polyphenol content in beans is beneficial from the perspective of enhancing mineral bioavailability. Soaking did not significantly ( $p > 0.05$ ) reduce the total polyphenol contents of the beans. Generally for all bean samples analysed, results indicated that both boiling and frying reduced total phenolic levels in beans. Boiling of the beans significantly reduced the total polyphenol contents by 42.9, 54.1, 50.8, and 47.5% for beans samples coded pink, blue, white and green respectively. [24] also found that boiling and steam cooking reduced the quantity of phenolic compounds in mung bean (*Vigna radiata*) by 73%. Similar results were reported by Rocha-Guzman *et al.*,<sup>25</sup> when phenolic content in common beans reduced drastically after pressure cooking. Heat generated during cooking may result in formation of heat induced insoluble protein-phenol complexes reducing phenolic compound extractability<sup>26</sup>. Siddhuraju and Becker<sup>27</sup> also reported a reduction in extractable total phenolics of autoclaved cowpea samples. Keeping the beans for a

further 8 hours did not have any significant ( $p > 0.05$ ) effect on the total polyphenol content of the beans. This treatment was conducted because beans take a long time to cook due to the hard to cook effect. To save on energy required for cooking, poor bean consumers within East African region tend to cook a large quantity of beans sufficient for two or more meals. Fried samples had higher but not significant ( $p > 0.05$ ) concentrations of total phenols compared with boiled samples probably because during frying other sources of phenolic compounds such as onions and spices were added to cooked beans to enhance flavor. Onions, tomatoes and spices are rich sources of phenolic compounds<sup>28,29,30</sup>. Therefore, high phenolic observed in frying samples might have been due to phenolic contribution from added onions and spices. Reductions in total polyphenols contents due to boiling are advantageous since it may lead to enhanced mineral bioavailability.

#### Degradation of Phytic Acid During the Cooking Process

The effects of soaking, boiling and frying on degradation of phytic acid content of beans are shown in Table 3.

**Table 3: Phytic acid contents of raw, soaked, boiled and fried beans ABC**

Treatment	Bean sample codes		
	Pink	Blue	White
Raw	10.3 ± 0.4 <sup>a</sup>	11.2 ± 0.1 <sup>b</sup>	12.5 ± 0.0 <sup>a</sup>
Soaked	ND	11.3 ± 0.1 <sup>b</sup>	ND
Boiled for lunch	9.7 ± 1.4 <sup>a</sup>	7.9 ± 0.1 <sup>d</sup>	6.1 ± 0.7 <sup>b</sup>
Boiled for dinner	9.7 ± 0.1 <sup>a</sup>	5.9 ± 0.1 <sup>e</sup>	6.3 ± 0.0 <sup>b</sup>
Boiled and fried for lunch	12.4 ± 0.8 <sup>a</sup>	12.0 ± 0.1 <sup>a</sup>	11.0 ± 1.3 <sup>ab</sup>
Boiled and fried for dinner	10.9 ± 2.7 <sup>a</sup>	11.1 ± 0.1 <sup>c</sup>	11.4 ± 0.0 <sup>ab</sup>

Values are given as mean ± SD (n = 3); ND = not done

<sup>A</sup> Data marked by the same superscript letters were not significantly different ( $p < 0.05$ )

<sup>B</sup> Results are expressed in mg/kg; C Not Done

The phytic acid contents of raw beans ranged from 8.2 to 12.5 mg/kg which is similar to values obtained in another study conducted in our laboratory on bean cultivars commonly grown in Rwanda. Soaking did not significantly ( $p > 0.05$ ) reduce the phytic acid contents

of the beans. Similarly, [31] found that soaking for 16 h had very little influence on phytic acid especially in legumes. However, boiling significantly ( $p < 0.05$ ) reduced the phytic acid content of bean samples coded white, blue and green. In bean sample coded

pink there was a reduction in phytic acid content but not significantly ( $p > 0.05$ ). Reductions in phytic acid contents after boiling were; 5.8, 29.2, 29.5 and 51.2% for bean batches coded pink, green, blue and white respectively. The decrease in phytic acids during boiling is in advantageous since it decreases mineral bioavailability.

Frying of the beans resulted into a significant ( $p < 0.05$ ) increase in the phytic acid content of the beans except in the sample coded pink in which there was an insignificant ( $p > 0.05$ ) increase in phytic acid content. Grains are a component of spices and may be the major cause for the increase in phytic acid after frying.

### Conclusion

Soaking of the beans did not significantly ( $p > 0.05$ ) decrease minerals, total polyphenols and phytic acid content of beans. While boiling of beans significantly ( $p < 0.05$ ) decreased the concentration of total polyphenols and phytic acid but not the mineral content of the beans. On the other hand bean frying process decreased the total polyphenol content but

increased the concentration of phytic acids in beans. The bean boiling method used in this study in which soaking water and bean broth were not discarded resulted into better retention of microminerals and degradation of total polyphenols. Since frying results in increase in phytic acid, there is need to come out with frying techniques that use low phytic acid containing ingredients.

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