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*Full Length Research Paper*

## **Raised Iron Levels in Wet- ground *Vigna unguiculata* and *Capsicum frutescens* using Domestic Grinding Techniques**

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### **ABSTRACT**

The biosafety of commonly used domestic grinding techniques was investigated; the effects of attrition mills using new (attrition mill 1) and old (attrition mill 2) plates, wooden mortar and pestle, grinding stone and electric blender on iron content of wet-ground staple foods, *Vigna unguiculata* (cowpea) and *Capsicum frutescens* (pepper) were examined in this study. Attrition mill 1 was in use 4 weeks prior to this study while the attrition mill 2 had newly installed grinding plate. The wet-ground pepper and cowpea were analyzed using Atomic Absorption Spectrophotometer (AAS). The iron contents of wet- ground foods (pepper and cowpea) from both attrition mills were significantly higher ( $p>0.05$ ) than iron content of ground food using other methods of grinding. A 30-40 folds increases in the iron content of ground food samples were detected using attrition mills. Ground pepper from attrition mill 1 showed higher iron contents ( $4300\pm474.35\text{mg}$ ) than pepper ground in attrition mill 2 ( $3199\pm281.68\text{mg}$ ). These values are higher than recommended dietary allowances for iron intake. The increased iron content in ground pepper and cowpea observed in the present study confirmed the high risk of iron overloading using attrition milling. The level of contamination of ground food increased with use in attrition mills as a result of wear and tear of grinding plates.

**Key words:** Attrition mill, iron contamination, staple food, dietary intake

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### **INTRODUCTION**

Iron is the second most abundant metal found in the earth's crust after aluminum and transition elements with high melting and boiling point (World Steel Association, 2011). Iron is an integral part of many proteins and enzymes that maintain good health. In human, iron is an essential component of proteins involved in oxygen transport (National Institutes of Health, 2010). A deficiency of iron limits oxygen delivery to cells, resulting in fatigue, poor work performance, and

decreased immunity (Robert *et al.*, 2010; Institute of Medicine 2001,). On the other hand, excess amounts of iron can result in toxic effects and may be lethal (Bacon *et al.*, 2011).

Excess iron stores in the body tissues and organs where it causes injures since iron is not excreted once absorbed. Blood loss and desquamation of epithelia are the only ways stored iron levels are lowered (Anderson *et al.*, 2009). Iron overload is association with several genetic diseases including hemochromatosis, which affects approximately 1 in 250 individuals of Northern

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European descent (Burke *et al.*, 2000). Individuals with hemochromatosis absorb iron efficiently, which can result in a build-up of excess iron and can cause organ damage such as cirrhosis of the liver and heart failure (Pietrangelo, 2010; Burke, 2000; Institute of Medicine, 2001).

Removal of metal from the surfaces of grinding plates occurs in both dry and wet grinding. The factors affecting wear are governed by the distribution and characteristics of micro constituents in the metal or alloy. The degree of contamination depends on pH of the environment, chemical composition, microstructure, and hardness of the mill plate materials (Andrews and Kwofie, 2010). Earlier research carried out on the wearing ability of corn-mill grinding plates had shown that the corn-mill plates that were used in wet grinding tend to wear faster than corn-mill plates used in dry grinding (Ampadu-Mintah, 2008). Moisture plays an active role in the corrosion of iron. This study was designed to investigate status of wet-ground common staple foods; pepper and cowpea with a focus on probable added iron in raw food during milling processing.

## MATERIALS AND METHODS

Samples of two staple foods (cowpea- *Vigna unguiculata* from legumes and pepper-*Capsicum frutescens*, a stimulant from plant were purchased from Bodija market Ibadan, Oyo state, Nigeria. One hundred (100) grams of fresh pepper was weighed and washed with distilled water into white plastic container. Three replicates (unprocessed) served as control, while three other replicates, each for the different methods of

grinding (Attrition mills, mortar and pestle, grinding stone and electric blender) were milled. This was repeated six times consecutively at three days (72 hours) interval. Same procedure was repeated for cowpea using new pairs of grinding plate, except that the cowpea was peeled before milling.

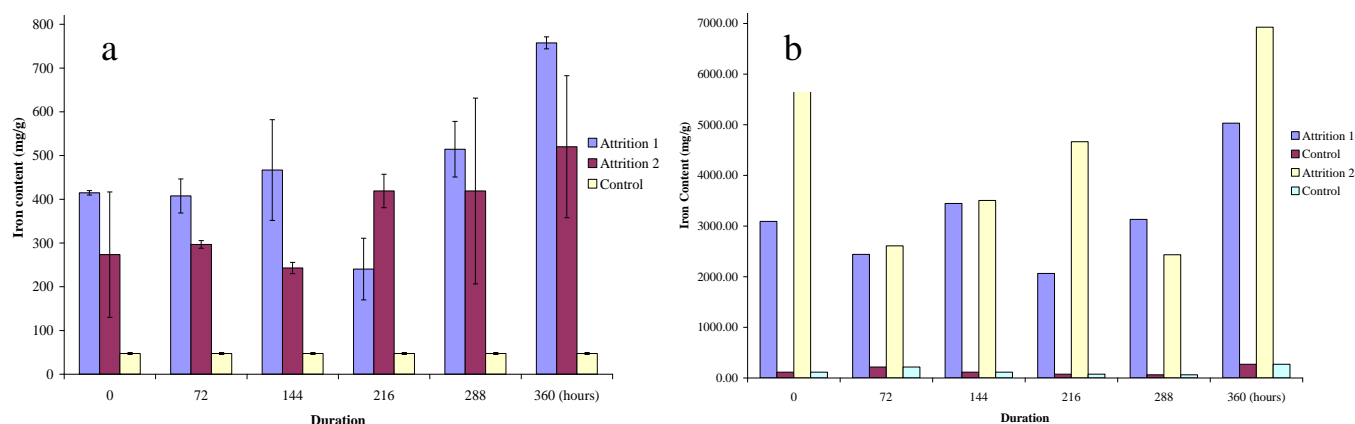
Samples were digested using modified Zheljzkov and Nielson's (1996) method, as outlined by Middleton (1973) and Anderson *et al.* (1974). Iron concentration was determined spectrophotometrically (Essien *et al.*, 1992) using Atomic Absorption Spectrophotometer, AAS Buck scientific equipment by Perkin-Elmer, model 210 VGP at a wavelength of 248 nm and slit setting of 7Å. The accurate functioning of the machines was ascertained by calibration with absorption standards of this mineral.

SPSS 16.0 for windows was used to determine level of significant differences in iron contents of ground and control samples.

## RESULTS

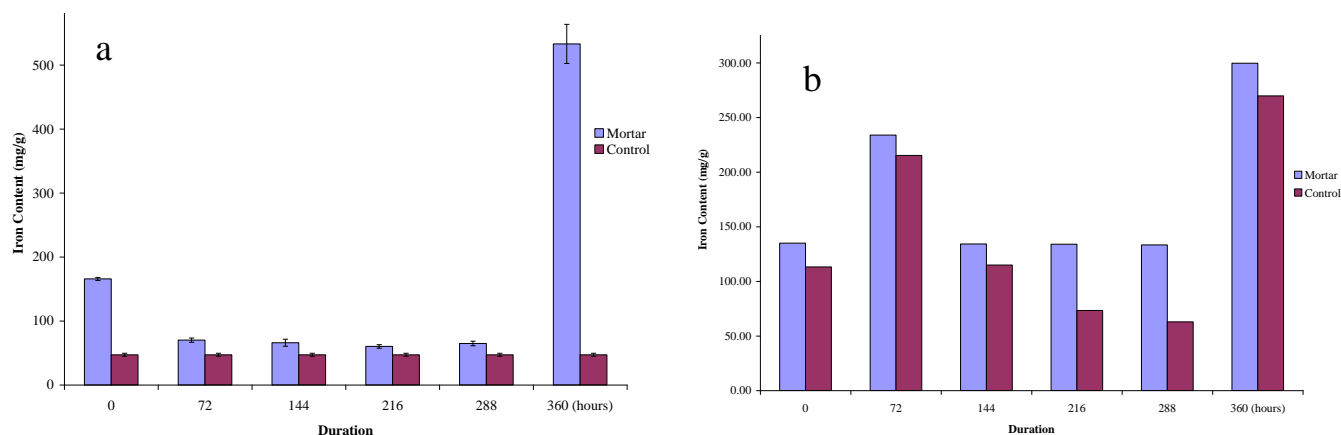
### Wet ground cowpea and pepper in attrition mills

Iron concentrations of ground cowpea samples obtained at 72 hours for 360 hours as shown in Figure 1a. The concentrations of iron obtained in cowpea ground using attrition mill 1 were  $414.50 \pm 5.11 \text{ mg/g}$ ,  $407.50 \pm 38.92 \text{ mg/g}$ ,  $466.67 \pm 115.09 \text{ mg/g}$ ,  $240.33 \pm 70.40 \text{ mg/g}$ ,  $514.33 \pm 63.59 \text{ mg/g}$  and  $757.50 \pm 13.77 \text{ mg/g}$  while control sample was  $47.25 \pm 2.38 \text{ mg/g}$ . Differences in iron contents with increasing duration of use compared with the control using attrition mill 1 were significant ( $F = 14.36$ ,  $p < 0.05$ ,  $df 6, 14$ ).



**Fig. 1**

Iron content of (a) wet- ground cowpea and (b) wet- ground pepper in attrition mills 1 and 2

**Fig. 2**

Iron content of (a) wet- ground cowpea and (b) wet- ground pepper in mortar and pestle

The concentrations of iron obtained in cowpea ground in attrition mill 2 were  $273.33 \pm 143.36$ mg/g,  $296.67 \pm 8.82$ mg/g,  $242.75 \pm 12.95$ mg/g,  $418.92 \pm 162.35$ mg/g,  $418.75 \pm 212.47$ mg/g and  $520.00 \pm 38.19$ mg/g while control sample was  $47.25 \pm 2.38$ mg/g. The difference in 72 hourly were iron content was significant ( $F = 1.77$ ,  $p > 0.05$ ,  $df = 6, 14$ ). Iron contamination was higher in the first attrition mill; the new mill plate.

Concentrations of iron obtained in pepper ground in attrition mill 1 were  $3090.33 \pm 90.22$ mg/g,  $2439.83 \pm 436.60$ mg/g,  $3445.83 \pm 609.23$ mg/g,  $2062.83 \pm 18.99$ mg/g,  $3130.83 \pm 619.34$ mg/g and  $5029.17 \pm 667.57$ mg/g at 72 hour intervals respectively as shown in Figure 1b. Differences in iron content by the hour compared with the control was significant ( $F = 7.22$ ,  $p < 0.05$ ,  $df = 6, 14$ ). Iron content was highest at 360 hours ( $5029.17 \pm 667$ mg/g).

Concentrations of iron obtained in pepper ground in attrition mill 2 were  $5666.50 \pm 624.41$ mg/g,  $2609.17 \pm 54.21$ mg/g,  $3504.17 \pm 167.29$ mg/g,  $4665.00 \pm 1477.30$ mg/g,  $2432.50 \pm 25.66$ mg/g and  $6925.00 \pm 967.47$ mg/g at 72 hour intervals, between 0 and 360 hours respectively. There was significant difference in iron content with increasing hours using attrition mill 2 ( $F = 0.74$ ,  $p < 0.05$ ,  $df = 6, 14$ ). However, iron contamination was higher in the attrition mill 2, which was an old mill plate.

#### Wet-ground cowpea and pepper in mortar and pestle

Figure 2a shows the result of iron concentrations obtained in wet- ground cowpea using mortar and pestle. The concentrations of iron were  $165.77 \pm 2.40$ mg/g,  $70.10 \pm 3.24$ mg/g,  $66.03 \pm 5.55$ mg/g,  $60.35 \pm 2.74$ mg/g,  $64.92 \pm 3.61$ mg/g, and  $532.93 \pm 30.56$ mg/g, corresponding to 0-360 hours at 72 hours intervals respectively, while control sample had  $47.25 \pm 2.38$ mg/g iron content. Differences in iron content with increasing hour compared with the control was significant ( $F =$

$215.36$ ,  $p > 0.05$ ,  $df = 6, 14$ ) while 360 hours was the highest ( $532.93 \pm 30.56$  mg/g).

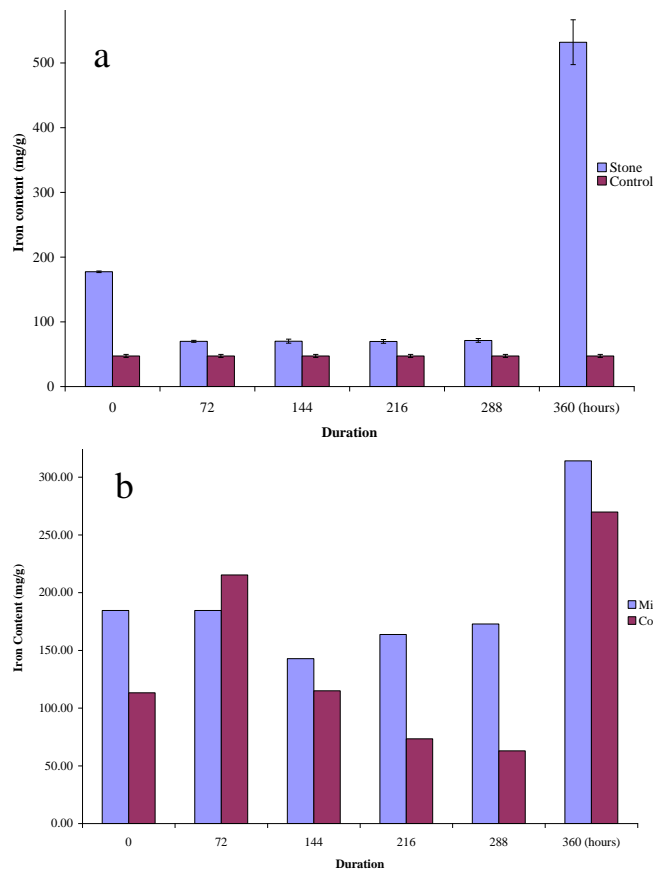
Figure 2b shows the result of iron concentrations obtained in wet- ground pepper using mortar and pestle. The concentrations of iron were  $135.00 \pm 0.14$ mg/g,  $233.92 \pm 0.58$ mg/g,  $134.30 \pm 1.16$ mg/g,  $134.08 \pm 0.93$ mg/g,  $133.42 \pm 1.59$ mg/g and  $299.75 \pm 33.75$ mg, at 72 hour intervals respectively. Iron content of control samples were  $133.25 \pm 38.09$ mg/g,  $215.33 \pm 64.00$ mg/g,  $115.00 \pm 4.51$ mg/g,  $73.33 \pm 8.85$ mg/g,  $63.00 \pm 13.79$ mg/g and  $269.85 \pm 47.50$ mg/g, also at 72 hour intervals, corresponding to 0- 360 hours respectively. Differences in iron contents were significant ( $F = 26.87$ ,  $df = 6, 14$ ,  $p < 0.05$ ), while control samples also recorded relatively high iron load (mg/g).

#### Wet-ground cowpea and pepper using grinding stone

Figure 3a shows iron concentrations obtained in wet-ground cowpea using traditional grinding stone. The concentrations were  $177.33 \pm 1.23$  mg/g,  $69.83 \pm 1.45$  mg/g,  $70.08 \pm 3.14$  mg/g,  $69.67 \pm 2.96$  mg,  $71.17 \pm 2.92$  mg/g and  $531.92 \pm 34.61$  mg/g, corresponding to 72 hourly samples respectively, while control sample was  $47.25 \pm 2.38$  mg. Millstone ground cowpea sample followed the same pattern as mortar and pestle. Iron content was significantly higher in samples obtained at 360 hours ( $F = 172.59$ ,  $df = 6, 14$ ,  $P < 0.05$ ). Least iron load was recorded in the control sample, while 216, 72, 144, 216 and 288 hours show no significant variation ( $p > 0.05$ ).

Figure 3b shows the result of iron concentrations obtained in wet- ground pepper using grinding stone. The concentrations of iron were  $184.50 \pm 21.63$ mg,  $184.50 \pm 18.67$ mg/g,  $142.83 \pm 10.64$ mg/g,  $163.75 \pm 29.91$ mg/g,  $172.83 \pm 39.38$ mg/g and  $314.17 \pm 11.03$ mg, at 72 hour intervals respectively. Iron content of control samples were  $133.25 \pm 38.09$ mg/g,  $215.33 \pm 64.00$ mg/g,

115.00  $\pm$  4.51mg/g, 73.33  $\pm$  8.85mg/g, 63.00  $\pm$  13.79mg/g and 269.85  $\pm$  47.50mg/g at 72 hour intervals respectively. Iron content of pepper ground on stone also follows the same pattern as mortar and pestle. Iron content difference was significantly highest ( $F = 6.38$ ,  $df = 6, 14$ ,  $p < 0.05$ ) in 360 hours sample, while the control also recorded high iron load (mg/g).



**Fig. 3**

Iron content of (a) wet-ground cowpea and (b) wet-ground pepper using grinding stone

### Wet-ground cowpea and pepper using electric blender

Figure 4a shows the result of iron concentrations obtained in wet-ground cowpea using electric blender. The concentrations of iron were 152.58  $\pm$  11.69 mg/g, 52.25  $\pm$  6.14 mg/g, 93.50  $\pm$  20.26 mg/g, 53.42  $\pm$  7.83 mg/g, 54.42  $\pm$  8.96 mg/g and 580.08  $\pm$  27.00 mg/g, at 72 hour intervals respectively, while control sample was 47.25  $\pm$  2.38mg/g. Iron content difference was significantly highest ( $F = 181.27$ ,  $df = 6, 14$ ,  $p < 0.05$ ) in 360 hours sample, while the control recorded the least iron load (47.25mg/g).

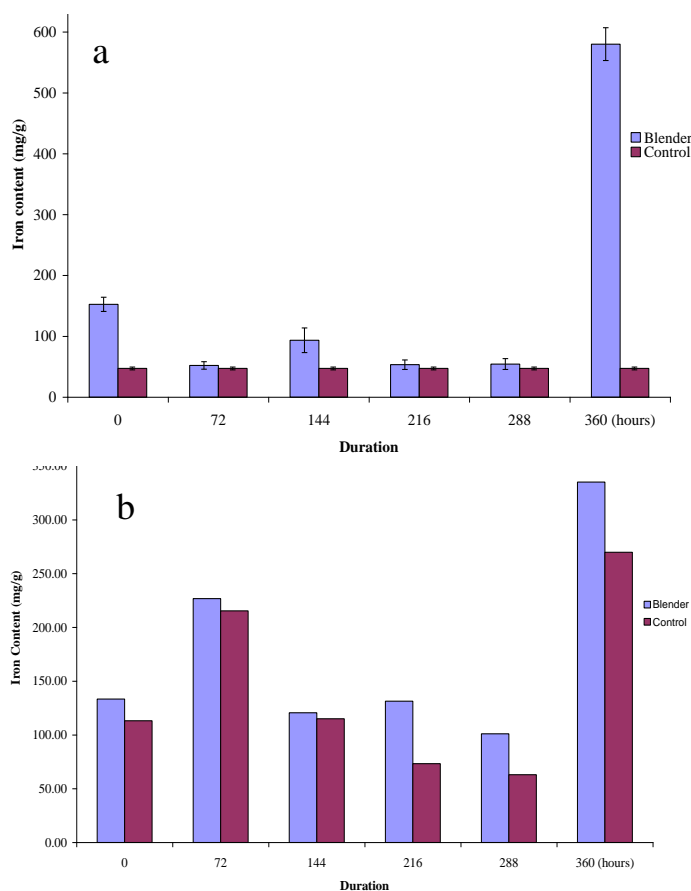
Figure 4b shows the result of iron concentrations obtained in wet-ground pepper using electric blender. The concentrations of iron were 133.33  $\pm$  23.97mg/g, 226.75  $\pm$  21.63mg/g, 120.67  $\pm$  10.95mg/g, 131.42  $\pm$

29.05mg/g, 101.08  $\pm$  13.41mg/g and 335.08  $\pm$  30.67mg/g, also corresponding 0- 360 hour respectively. Iron content of control sample between the periods were 133.25  $\pm$  38.09mg/g, 215.33  $\pm$  64.00mg/g, 115.00  $\pm$  4.51mg/g, 73.33  $\pm$  8.85mg/g, 63.00  $\pm$  13.79mg/g and 269.85  $\pm$  47.50mg/g. The result was significantly higher ( $F = 15.49$ ,  $df = 6$ ,  $p < 0.5$ ) in 360 hours sample, both in the control and test experiments.

### Comparative iron content in different mills

Table 1 shows that iron content is significantly higher in pepper ( $p < 0.05$ ) than in cowpea irrespective of the type of grinding. The attrition mill 2 with aged grinding plate showed higher iron concentration in pepper than ground pepper in attrition mill 1. However, cowpea deviated from this trend as the new attrition mill imparted more iron.

The attrition mills generally showed higher iron content (3199.81  $\pm$  281.68, 4300.39  $\pm$  474.35 and 406.87  $\pm$  43.42, 316.81  $\pm$  49.04mg/g) for pepper and cowpea respectively



**Fig. 4**

Iron content of (a) wet-ground cowpea and (b) wet-ground pepper using using electric blender

**Table 1:**

Iron concentrations of pepper and cowpea in relation to mill types

Food type	Iron concentrations (mg/g)				
	AM1	AM2	EB	GS	MP
Pepper	3199±281.68	4300.39±474.35	174.72±21.38	193.76±15.84	178.41±16.54
Cowpea	406.87±43.42	316.81±49.04	164.38±46.22	165.00±41.21	160.02±41.66

Key: AM1 = 1st Attrition mill, AM2 = 2nd Attrition mill, B = Electric blender, S = Grinding stone and M = Mortar and pestle

## DISCUSSION

A 30-40 fold increase in the iron content was noted when attrition mill was used for the two staple foods sample used in the present study, while, lower iron content was observed in other methods of grinding confirming that iron contamination occurs during grinding. It was also observed that rate of increased iron content varied significantly depending on mill type. Yahaya *et al.* (2012) reported an increase in iron metal concentration from 7.00ppm on pounded tomatoes to 26.33ppm on ground tomatoes.

Recommended dietary allowances for iron intakes according to the Institute of Medicine (2001) are 11mg/day for 7 – 12 months old, 7mg/day for 1- 3 years old, 10mg/day for 4- 8 years and 8mg/day from 9 – 13 years old for both sexes. Moreover, 11mg/day for 14 – 18 years for male while 15mg/day for female between these ages (14 – 18 years) is also within safe limit. Furthermore, 8mg/day for male from 19 years above and 18mg/day for female at 19 – 50 years of age while 8mg/day is adjudged safe for female from 51 years above (menopause age). Intakes below these recommendations can result in iron deficiency while iron overload is toxic to living tissue. Iron content obtained in the present study for ground samples from attrition mill was very high compare to recommend dietary allowances iron intakes, although all is not expected to be absorbed by duodenum and upper jejunum. Healthy adults have been reported to absorb about 10-15% of dietary iron, while individual absorption is influenced by several factors such as iron status, nutritional deficiencies, infection/inflammation, genetic disorders among others (Miret *et al.*, 2003; Hurrell and Egli, 2010) Iron content for all grinding methods fluctuated within the period of grinding and this was observed more in the attrition mills showing that iron corrosion and abrasion continues unabated (Andrew and Kwofie, 2010). This may be due to the state of the attrition mills used. According to Normanyo *et al.* (2009), poor alignment will cause vibration and lead to wear of bearings, seals and other rotating elements, which is the most probable reason for the difference observed in iron content between the grinding methods.

Iron content of cowpea ground in attrition mills 1 and 2 is disparate compared to other methods of grinding and control. Iron content recorded for this study in ground cowpea using attrition mills over the period of the time of sample collection was between  $407.50 \pm 38.92$  and  $757.50 \pm 13.77$ mg for the first machine while the second machine range between  $242.75 \pm 12.95$  to  $520.00 \pm 38.19$ . The difference in iron content of the cowpea paste obtained was significantly higher ( $p < 0.05$ ) in the two mills compared to the control and other methods. Mustapha and Ogundahunsi (2010) in a related work using cast iron pots in cooking obtained  $413 \pm 0.02$ mg of iron imparted after the cooking.

From the present study, it can be inferred that cast iron grinding plates would wear faster in wet grinding due to the tendency to form oxide layer, which would result in corrosive wear in addition to the abrasive wear as explained by Andrew and Kwofie, (2010). Wear is assumed to be affected by corrosion only in regions where protective oxide layer forms. The grinding mechanism of food in attrition mill machine causes surface interaction between the two milling plates and the food being processed. This surface interaction increased the corrosion products which subsequently contributed to increased corrosion rate as it exposes fresh surfaces for corrosion to occur (Normanyo, *et al.* 2009). Subsequent contamination of the milled food is inevitable. Consumption of the corrosion products could impact on the health of consumers as the form of the oxide consumed could be insoluble and therefore bio-accumulate in the liver (Normanya *et al.*, 2009).

An experiment carried out by Normanyo *et al.* (2009) showed that there were iron fillings in the food produced by dry grinding machine and the contact made by the grinding discs is what produces the iron fillings. Francis, (2004) also conducted two experiments using wheat flour or cereal, water, zip-lock plastic bag or bowl and a magnet to discover presence of iron particles in ground food material. Studies by Geerligs *et al.* (2003) used iron pot to improve haemoglobin concentration in adults and reduce iron deficiency in children living under malarious endemic conditions. These authors further reported high content of iron obtained from food samples cooked in cast iron pot. Iron content reported in all ground food



differed from reference values available in food composition tables presented by FAO (1968); Platt (1975) and Yahaya *et al.* (2012).

There was higher moisture content both in control and ground pepper samples compared to the raw as well as ground cowpea samples. According to Nielsen (1998), foods are heterogeneous materials that contain different proportions of chemically and physically bound, capillary, trapped or bulk water. Despite the same chemical formula ( $H_2O$ ) water molecules in food may be present in varying state depending on interaction with the surrounding molecules. The water in different environments normally have different physicochemical properties. This might explain the variations in iron content observed in raw and processed sample foods. Moisture plays an active role in the corrosion of iron (Normanyo *et al.*, 2009). Usually, the corrosion product does not adhere to the iron surface and therefore corrosion continues unabated resulting in material degradation. Cast iron is strongly susceptible to corrosion attack in food product environments due to their acidic nature. It was observed that sample with high moisture value has high content of iron and water used during grinding in attrition mills and could also increased amount of corrosion of grinding plates, thus increasing concentration of Iron in wet ground foods. The fluctuation between the samples from various treatments could be due to the continuous production of hydroxide ions from corroded cast iron plate in use. Observations from earlier works suggested that corrosion rate decreased steadily to pH values greater than 6 (Roberge, 1999). From Fe- $H_2O$  potential-pH diagram at pH above 6, protective scale forms in the form of  $Fe(OH)_2/Fe(OH)_3$  which hinder mass transport of oxygen and ionic species in the solution. The pH of the corrosive environment increases with time from acidic to alkaline environments, where the tendencies to form any protective layer exist.

## CONCLUSION

The use of the grinding mill to grind pepper, cowpea and other foodstuffs produced iron as part of the food consumed giving rise to iron overload or hemochromatosis which is the cause of myriads of health disorders. The present study has shed more light on the fact that eating habit can results in increased self inflicted harm. Food processed with attrition mill may contribute to increment in iron requirements of consumed staple foods. There is need for in-depth research to further probe into this area of study so as to throw more light on this micronutrient which is of high importance to human health. Suitable material for food milling with combination of wear and corrosion resistances needs to be put in place. Suitable materials

and designs for food milling with combination of wear and corrosion resistance needs to be developed to prevent iron requirements from exceeding recommended limits.

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