Determination of metals in pepper by flame atomic absorption spectroscopy

Erepamowei YOUNG and Timi TARAWOU

Department of Chemical Sciences, Faculty of Science, Niger Delta University, Wilberforce Island, PMB 71, Yenagoa, Bayelsa State, Nigeria.

*Corresponding author; E-mail: ttarawou@gmail.com; Tel.: +234 (0) 803 67 26 459.

ABSTRACT

The health risk of dietary intake of metals by humans cannot be over-emphasized, especially metal-contaminated farm produce from an abandoned waste dump site. Metals could contaminate farm produce during pre- and post-harvest periods. The metal (zinc, copper, chromium, iron, manganese and cobalt) contents of Capsicum annum, Capsicum chinens and Capsicum frutescens pepper samples (harvested from an abandoned waste dump site in Gbarantoru in Bayelsa State) were determined and compared with WHO permissible levels. The pepper samples were crispy-dried, ground to fine powder, and digested with an acid mixture of sulphuric acid, perchloric acid and nitric acid in the ratio of 1:4:40. Prepared samples were analyzed using flame atomic absorption spectrophotometer. The results showed a wide range of concentrations: Capsicum annum (Zn = 40.28 mg/kg, Cu = 4.17 mg/kg, Cr = 3.32 mg/kg, Fe = 105.23 mg/kg, Mn = 0.54 mg/kg, Co = 0.69 mg/kg), capsicum chinens (Zn = 54.48 mg/kg, Cu = 4.22 mg/kg, Cr = 2.81 mg/kg, Fe = 158.33 mg/kg, Mn = 0.66 mg/kg, Co = 0.73 mg/kg), Capsicum frutescens (Zn = 47.45 mg/kg, Cu = 4.36 mg/kg, Cr = 3.20 mg/kg, Fe = 63.13 mg/kg, Mn = 0.59 mg/kg, Co = 0.77 mg/kg). The metal concentrations of the three hot pepper samples were found to be below WHO permissible limits except for zinc in Capsicum chinens, which was found to be higher than the permissible limit. These species of pepper grown in this abandoned waste dumpsite are safe for food as stipulated by WHO.

Keywords: Metals, pepper, flame, atomic absorption, waste, dumpsite.
loose-textured soil with a pH of 4.3 to 9.7 (Johnson, 2007).

Because of the burning sensation caused by capsaicin when it comes in contact with mucous membranes, pepper is commonly used in food products to give them added spice or heat (Stewart et al., 2007).

Farm produce can get contaminated through different anthropogenic sources and many researchers have found different interests in determining metals in farm produce. Antonious et al. (2010) determined the concentrations of heavy metals in plants, fruits and vegetables because of the potential health risks associated with eating heavy-metal-contaminated edible plants. Metals could be accumulated in agricultural soils through irrigation and subsequently bioaccumulated in pepper grown in them. In order to investigate the levels of accumulation, Adeyeye (2005) determined trace metals (Zinc, manganese, cobalt, selenium, copper, molybdenum, chromium, iron, aluminium, lead and cadmium) in buds, flowers, fruits, seeds, leaves, stems, roots, cobs, styles, shaft, grains and efflorescence of plants and in soils from Fadama farms in Ekiti State in Nigeria.

Another investigation was also carried out on Fadama Farms (Owodo et al., 2008) on the levels of some trace metals (lead, chromium and copper) in the Fadama soils and pepper (Capsicum annuum) along the bank of River Challawa in Nigeria. Investigations have also been carried out to determine the bioaccumulation of hazardous chemicals in plants, fruits and vegetables (Anhwange et al., 2009; Audu et al., 2005).

An abandoned waste dump site in Gbarantoru in Bayelsa State (Nigeria) is used for pepper-farming. This farm produce constitute some percent of pepper supply to local markets such as Swali Market in Yenagoa metropolis. The harvested pepper is also exposed to dust during transportation and while on sale in the open baskets and exposed tables in the market. The pepper could be contaminated with metals before harvest (via metal uptake and bioaccumulation) and after harvest. The uptake and bioaccumulation of metals by plant and the factors influencing such phenomena were reported by Yusul and Osibanjo (2006). The possible contamination of farm produce such as vegetables by air and aerosol was investigated and reported by De Nicola et al. (2008). However, the ultimate consequence of the contamination is the attendant health risk; Turkdogan et al. (2003) have reported the health risk of dietary intake of metals by humans and animals. The report established a connection between the prevalence of high gastrointestinal cancer and dietary intake of high levels of metals.

This research was aimed at comparing the levels of Zn, Cu, Cr, Fe, Mn, and Co in some species of pepper produced from an abandoned waste dump site with the WHO-permissible metal levels. This was done against the backdrop of the health risk associated with dietary intake of metals.

MATERIALS AND METHODS
Chemicals/reagents
Analytical grade chemicals and distilled water were used throughout the study.

Glassware and equipment
All glassware and plastic containers were washed with detergent, followed by 20% (v/v) concentrated trioxonitrate (IV) acid, and rinsed with distilled water.

Sample collection and preparation
The three species of hot pepper (Capsicum annuum, Capsicum chinensis, and Capsicum frutescens) were bought at Swali Central Market in Yenagoa, Bayelsa State of Nigeria; all samples were produce from farms in Gbarantoru waste dumpsite.

The pepper samples were washed with tap water, rinsed with distilled water, sliced in nearly uniform size to facilitate drying of the pieces at the same rate. The sliced pieces were then dried in an oven at 105 °C for 24 hours until they were brittle and crispy. At this stage, no microorganism can grow and care was taken to avoid any source of
contamination. The dried samples were ground into fine particles using clean mortar and pestle.

**Digestion**

The procedure adopted by Awofolu (2005) was used for the digestion of the samples. Oven-dried pepper (*Capsicum annuum*) was ground into fine powder in a porcelain mortar. 0.5 g of the ground pepper sample was weighed into 100 mL beaker followed by the addition of 20 mL of an acid mixture of sulphuric acid, perchloric acid and nitric acid in the ratio of 1:4:40. This was placed on hot plate (heater) and heated slowly until frosting stopped and the foaming ceased. Then the heat was raised until the mixture started boiling. Boiling was maintained until a clear solution was obtained. The heat was removed; samples were allowed to cool down, filtered and then diluted to mark with distilled water in a 100 mL-capacity volumetric flask. The samples were then analyzed for the concentrations of zinc, copper, chromium, iron, manganese, and cobalt using flame atomic absorption spectrometer (FAAS).

This was repeated for the other species of pepper, *Capsicum chinens* and *Capsicum frutescens*. Five replicate analyses were carried out for each of the samples under investigation.

**RESULTS**

The concentrations of the selected metals (zinc, copper, chromium, iron, manganese, and cobalt) were determined by flame atomic absorption spectrometer and the results are presented in Table 1. The concentrations of Zn, Cu, Cr, Fe, Mn, and Co as given in Table 1 were compared with the maximum permissible standard concentration of the metals as recommended by the World Health Organization (FAO and WHO, 2001); the WHO standards of the metals being studied are presented in Table 2. Calibration graphs were plotted and equations of straight line and regression coefficient ($R^2$) values were obtained for all the metals being determined and are presented in Table 3.

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**Table 1:** Concentration of metals in the various hot pepper samples.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Zn</th>
<th>Cu</th>
<th>Cr</th>
<th>Fe</th>
<th>Mn</th>
<th>Co</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Capsicum annuum</em></td>
<td>40.28 ± 0.03</td>
<td>4.17 ± 0.03</td>
<td>3.32 ± 0.03</td>
<td>105.23 ± 0.09</td>
<td>0.54 ± 0.02</td>
<td>0.69 ± 0.01</td>
</tr>
<tr>
<td><em>Capsicum chinens</em></td>
<td>54.48 ± 0.08</td>
<td>4.22 ± 0.01</td>
<td>2.81 ± 0.01</td>
<td>158.33 ± 0.06</td>
<td>0.66 ± 0.02</td>
<td>0.73 ± 0.03</td>
</tr>
<tr>
<td><em>Capsicum frutescens</em></td>
<td>47.45 ± 0.05</td>
<td>4.36 ± 0.02</td>
<td>3.20 ± 0.02</td>
<td>63.13 ± 0.08</td>
<td>0.59 ± 0.01</td>
<td>0.77 ± 0.02</td>
</tr>
</tbody>
</table>

**Table 2:** Codex permissible standards for metals in hot pepper (2001).

<table>
<thead>
<tr>
<th>Metal</th>
<th>Recommended standard (mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fe</td>
<td>300</td>
</tr>
<tr>
<td>Cu</td>
<td>20</td>
</tr>
<tr>
<td>Cr</td>
<td>30</td>
</tr>
<tr>
<td>Zn</td>
<td>50</td>
</tr>
<tr>
<td>Mn</td>
<td>100</td>
</tr>
<tr>
<td>Co</td>
<td>50</td>
</tr>
</tbody>
</table>
Table 3: Equation of straight line and regression coefficient ($R^2$) values for the metals.

<table>
<thead>
<tr>
<th>Metal</th>
<th>N</th>
<th>Equation</th>
<th>$R^2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fe</td>
<td>5</td>
<td>$y = 0.0571x + 0.0113$</td>
<td>0.996</td>
</tr>
<tr>
<td>Cu</td>
<td>5</td>
<td>$y = 0.0109x + 0.0027$</td>
<td>1.000</td>
</tr>
<tr>
<td>Cr</td>
<td>5</td>
<td>$y = 0.0535x + 0.0189$</td>
<td>0.999</td>
</tr>
<tr>
<td>Zn</td>
<td>5</td>
<td>$y = 0.0005x + 0.0130$</td>
<td>1.000</td>
</tr>
<tr>
<td>Mn</td>
<td>5</td>
<td>$y = 2.9477x + 0.0523$</td>
<td>1.000</td>
</tr>
<tr>
<td>Co</td>
<td>5</td>
<td>$y = 3.992x + 0.0040$</td>
<td>1.000</td>
</tr>
</tbody>
</table>

$N =$ Number of replicate analysis; $R^2 =$ regression coefficient

DISCUSSION

Comparing the WHO-recommended standards and the determined metal concentrations, it was found that the zinc concentration was tolerable for the hot pepper samples *Capsicum annum* (40.28 mg/kg) and *Capsicum frutescens* (47.45 mg/kg). While for the *Capsicum chinens* sample, the determined zinc concentration was 54.48 mg/kg, which was found to be higher than the permissible limit of 50 mg/kg. Therefore, consumption of *Capsicum chinens* from the same source where the sample was obtained on a continuous basis may be a health risk.

It was further shown in Tables 1 and 2 that for all the pepper samples being studied, the determined concentrations of the other metals (Cu, Cr, Fe, Mn and Co) were much lower than the permissible recommended levels. Thus, consumption of the three pepper samples would not pose any health risks to consumers. Although the concentrations of iron (Fe) in the three pepper samples were found to be relatively higher than the concentrations of other metals under investigation (105 mg/kg, 158 mg/kg and 63 mg/kg respectively for *Capsicum annum, Capsicum chinens* and *Capsicum frutescens*), the values are still within WHO-permissible level, which is 300 mg/kg. The importance of dietary intake of Fe cannot be over-emphasized because it forms part of the structural and functional components of hemoglobin in red blood cells (Alan, 2008), myoglobin in muscles cells (Brunori et al., 2004) and a constituent of all cysteinato-heme enzymes (Bernard et al., 2004).

Cobalt can be toxic at high levels, however the determined concentration values of cobalt for all the three samples of hot pepper being studied (*Capsicum annum, Capsicum chinens* and *Capsicum frutescens*) were very low (0.69 mg/kg, 0.73 mg/kg and 0.77 mg/kg respectively); much lower than the WHO standard of 50 mg/kg.

From Table 2, the high regression coefficient ($R^2$) values for Fe, 0.996; Cu, 1.000; Cr, 0.999; Zn, 1.000; Mn, 1.000; and Co, 1.000 showed that the calibration graphs were suitable for verifying unknown metal concentration in the various hot pepper samples.

Conclusion

All the three samples of hot pepper being studied contained various concentrations of zinc, copper, chromium, iron, manganese, and cobalt. The metal concentrations of the three hot pepper samples were found to be within tolerable limits except for zinc in *Capsicum chinens*, which was found to be higher than the permissible limit of 50 mg/kg. The concentration of iron (Fe) in the three pepper samples were found to be relatively high and was desirable since the permissible standard recommended for iron is 300 mg/kg. Therefore, farm produce (sold in Swali local market) from abandoned waste-dump site at Gbarantoru in Bayelsa State (Nigeria) is safe for consumption in respect of
the metals under investigation as stipulated by WHO.

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