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QUANTIFICATION AND HEALTH RISK ASSESSMENT OF HEAVY METALS IN FRESH AND GRILLED SAUSAGES SOLD IN THE TAMALE METROPOLIS, GHANA

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Abstract
Heavy metals concentrations are in greater levels in major cities or industrial areas than they would exist in the natural environment. They pose a serious threat to humans due to their characteristic toxicity, bioaccumulation and bio-magnifications in the food chain. Street foods like meat products such as grilled sausages pose a potential health risk to regular consumers as a result of their exposure to the open surroundings. This study reports on the presence and levels as well as the potential associated health risks of Lead (Pb), Manganese (Mn), Iron (Fe) and Copper (Cu) in fresh and grilled sausages sold in the Tamale metropolis. The samples were analyzed using the Atomic Absorption Spectrophotometer (AAS). Lead was not detected in either of the samples. The average levels of Copper, Manganese and Iron were 0.3057 ± 0.0918 mg/kg, 0.1657 ± 0.0054 mg/kg and 1.2436 ± 0.1664 mg/kg, respectively for the fresh samples. The average levels of Copper, Manganese and Iron recorded in the grilled samples were 0.0470 ± 0.0096 mg/kg, 0.1512 ± 0.0048 mg/kg and 1.3242 ± 0.1881 mg/kg, respectively. The health risks assessment conducted indicated that there were no possible risks of cancer posed towards the consumers.

Keywords: Heavy metals, Sausages, bioaccumulate, risk assessment, carcinogenic risk, grill, contamination

Introduction

Metals are natural components of the environment. However, rapid growth of industrialization has accounted for their widespread diffusion into the environment (Rajaganapathy et al., 2011). Numerous researches have indicated that heavy metal concentrations are in greater levels in major cities or industrial areas than would exist in the natural environments (Breward, 2003; Rawat et al., 2003; Marcuvechio, 2004).

Metals including Fe, Cu, Zn and others are of great importance for human development, but prolonged metabolic disorders may occur due to the deficiency or excess of these metals in the human body. However, non-essential elements such as Pb, Cd, Cr, Ni and several others are regarded as harmful and toxic and their presence in the body even in lower concentrations can cause severe biochemical and neurological changes (Schroeder, 1991).

Heavy metals have specific weight above 5 g/cm^3 (Gonzalez-Waller et al., 2006) and can be called trace elements due to their presence in trace (10 mg/kg) or in ultra-trace (1µg/kg) quantities in the environmental media (Raikwar et al., 2008). Some heavy metals are neither vital nor have beneficial effects but possess severe health effects at trace levels (Goyer, 2004).

The relevant sources of environmental exposure to heavy metals include but are not limited to the following; fossil fuels, mining industries, waste disposals and municipal sewage, and farming due to the uses of fertilizers and pesticides (Jayasekara et al., 1992).

The toxicity of heavy metals is further enhanced substantially due to their synergistic relationships within the natural ecosystem, the average long life of these elements, their persistence and their potential transformation to more toxic compounds (Nollet, and De Gelder, 2000).

Elements like vanadium, tungsten and cadmium are known to be toxic for some organisms, however, they can be beneficial under certain conditions (Gupta, 2012). In higher concentrations, they can result in metabolic abnormalities. It is therefore problematic when it comes to establishing the differences in their beneficial and toxic levels (Auroville Innovative Urban Management, 2013). Ingestion of contaminated foods and food products including meats and meat products constitute one of the major and basic sources through which higher levels of heavy metal concentration can get into the body of humans.

Meat constitutes an essential component of the food we eat and it is mainly comprised of protein, fat and some quantities of essential elements (Akan et al., 2010). It also forms a good source of niacin, vitamins B6 and B12, phosphorous, zinc, and iron (Williams, 2007). Aside meat and meat products contributing significantly to human diet and as an important source of a variety of nutrients, it can also be a vehicle of toxic substances (Fathy et al., 2011).

Meat can be exposed to a variety of toxic substances through various sources such as animal drugs, pesticides, feed, agricultural or industrial chemicals substances and many more (Fathy et al., 2011). Animal proteins are of high biological value (Dabuo, 2011), and the availability of essential amino acids in them makes a complete protein (Bastin, 2007). However, studies have reported heavy metal contamination in meat and meat products during processing through handling of the raw material, spices, water and packaging (Brito et al., 2005; Santhi, et al., 2008). Also, inhalation of contaminated air, penetration through the skin (Raikwar et al., 2008; Santhi et al., 2008), contaminated animal feed and many others have been reported as sources of heavy metal contamination in meat (Sabir et al., 2003; Miranda et al., 2005). The ingestion of these contaminants by animals also result in traces of heavy metals residues in meat (Sabir et al., 2003).

Sausages constitute part of the meat products that are mostly consumed globally. In Ghana for instance, sausages are consumed in grilled forms at kebab joints or cooked forms at homes. This research therefore sought to examine the levels and the possible health risks associated with heavy metal contamination of fresh and grilled sausages.

Methods

Study Area

This study was conducted in the Tamale metropolis of the northern region of Ghana. The region has a total land size of about 646.9 km^2 and a geographical location between latitude 09°16 and 09°34 North and longitude 00°57 West (Ghana Statistical Service, 2014).

According to the 2010 Population and Housing Census, the population of the Metropolis was 233,252 (49.7% males and 50.3% females) representing 9.4% of the regional population with majority of the population being youth. About 63.3% of the population ages 15 years and above in the metropolis and were economically active (Ghana Statistical Service, 2014).

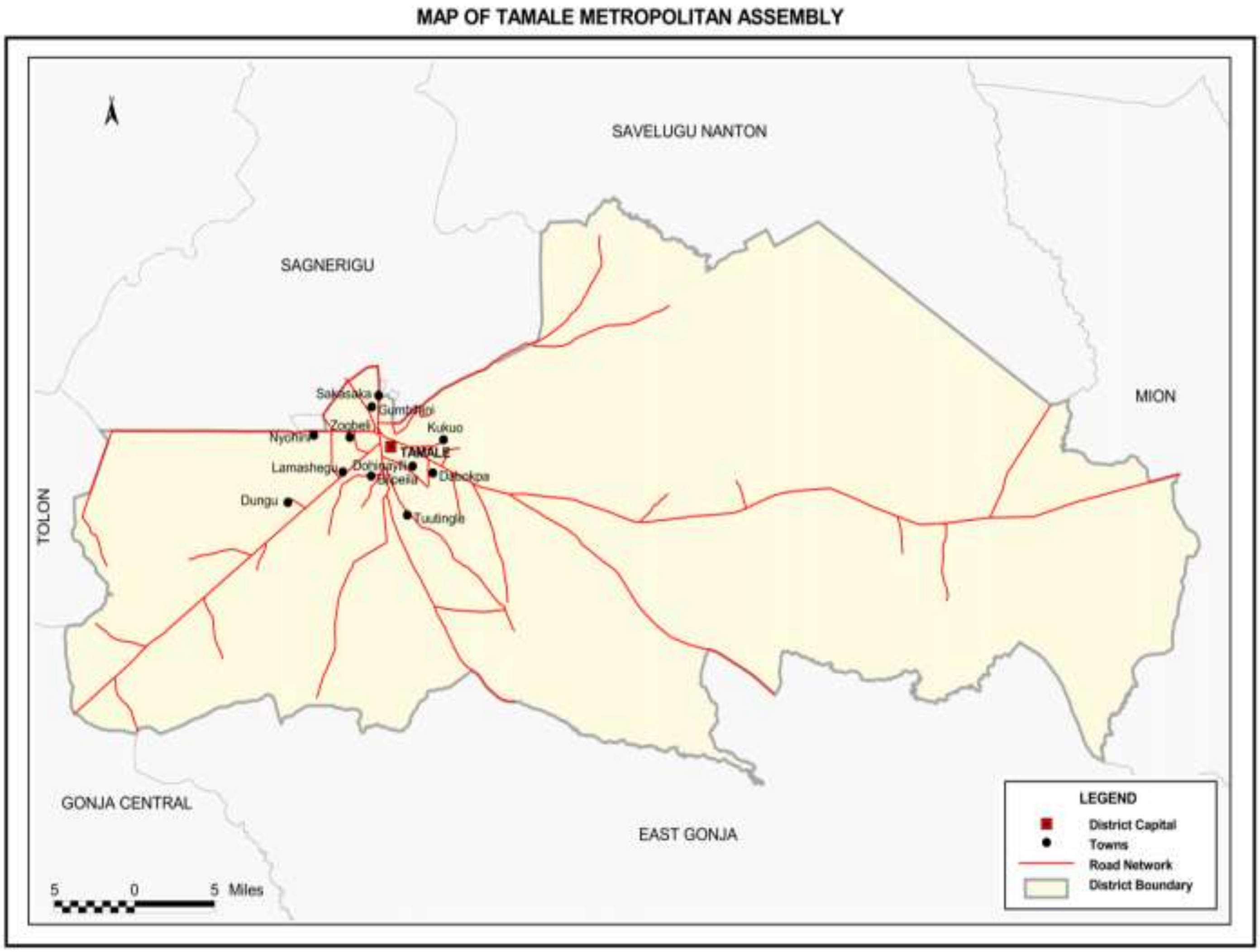

Fig 1: Map of the Study Area
Source: *Ghana Statistical Service (2014)*

Sample Collection

Ten (10) fresh and grilled sausages samples each were collected from ten (10) popular sausage joints in the business district of Tamale. Two samples (one fresh and one grilled) were collected from each joint. Samples were collected from sausage joints around Ghana Commercial Bank, Biege Bank and around the Jubilee Park in the Tamale metropolis. The samples were collected into zip bags, preserved in an ice chest at 6 ^{o}C and then transported to the central laboratory of Kwame Nkrumah University of Science and Technology (KNUST) for heavy metals analysis in March 2019.

Sample Preparation and Analysis

The collected samples were oven dried at 60 °C for twenty- four hours and then grinded into finer particles. One gramme of each grinded sample was kept in a 250 ml conical flask and 20 ml of di-acid mixture (HCL and $HClO_4$, 3:1 v/v) added to the mixture.

The content was then stirred to moisten the sample and heated over a hot plate at a temperature range of 180 °C to 200 °C in a fume chamber till there was the formation of white fumes. The flask was then taken off the hot plate and allowed to cool. It was filtered and topped up to 50 ml volumetric flask with a filter paper (whatman number one). The conical flask was rinsed severally and added to the mixture. The filtrate was transferred into sample containers and clearly labelled. They were then analyzed for the desired metals using Analytikjena model novAA400P Atomic Absorption Spectrophotometer (AAS) manufactured in Germany. Using a pneumatic nebulizer, a small volume of the sample was aspirated into a flame where the ions are reduced to elements and vaporized. The elements present in the sample then absorb light (generated from the HCL) at specific wavelengths (see table above) in the visible or the ultraviolet spectrum. This is dependent on the wavelength of maximum absorption of the analyte. After absorption, the transmitted light is detected with a detector after going through a monochromator (Lab Unlimited, 2013).

Exposure Assessment

The health risk assessment was done using the U.S. Environmental Protection Agency's exposure variables and guidelines manual (USEPA, 2002, 2003). The average daily intake (ADD_{ing}) for children and adults was assessed using the following equation:

$$ADD_{ing} C_{food} \times \frac{IngR \times EF \times ED}{BW \times AT} \times 10^{-6} \ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

$IngR$ is the Ingestion Rate

EF is the Exposure Frequency

The hazard quotient (HQ), carcinogenic risk (RI) and hazard index (HI) models were used to evaluate the non-carcinogenic effects (Ying et al., 2016). According to USEPA (1989), the hazard quotient refers to the ratio of the average daily dose of heavy metals/metalloids to its reference dose (RfD):

$$HQ = \frac{ADD}{RfD} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2)$$

HQ < 1, implies there is no likelihood of adverse effects, whereas HQ > 1, signifies the possibility of adverse effects (USEPA, 2011).

The hazard index (HI) method can be used to determine the general negative adverse effects of non-carcinogenic risk. To calculate for the HI of each of the samples, the HQ of the individual metals were summed up. The HI is defined as the combined effect of the HQ through the three routes of exposure for heavy metals/metalloids (USEPA, 1986).

$$HI = \sum HQ_i \ldots\ldots\ldots\ldots\ldots\ldots (3)$$

HI>1 means an unacceptable risk of carcinogenic effects and HI<1 means an acceptable level of risk of non-carcinogenic effects on health.

Findings

From the study, fresh samples recorded Copper concentrations ranging from 0.0282 to 0.9484 mg/kg and a mean of 0.3057 ± 0.0918 mg/kg. Manganese concentrations were from 0.1312 to 0.1904 mg/kg with an average of 0.1657 ± 0.0054 mg/kg. Iron concentrations were from 0.7960 to 2.222 mg/kg with a mean of 1.2436 ± 0.1664 mg/kg.

Table 1: Concentration of Mn, Cu and Fe in fresh sausage samples

Samples	Concentration in mg/kg							
	Cu	SD	Mn	SD	Pb	SD	Fe	SD
Blank	0.0024	0.0176	0.1238	0.0033	BDL	N/A	0.5937	0.0024
JF1	0.0395	0.0030	0.1312	0.0031	BDL	N/A	0.8943	0.0145
JF2	0.2732	0.0041	0.1757	0.0030	BDL	N/A	1.230	0.0158
JF3	0.1001	0.1495	0.1904	0.0020	BDL	N/A	0.9229	0.0059
JF4	0.0668	0.0009	0.1612	0.0017	BDL	N/A	1.057	0.0092
JF5	0.3537	0.0024	0.1656	0.0027	BDL	N/A	0.8862	0.0041
JF6	0.5894	0.0049	0.1894	0.0010	BDL	N/A	2.222	0.0185
JF7	0.9484	0.0181	0.1579	0.0042	BDL	N/A	0.7960	0.0044
JF8	0.2486	0.0048	0.1568	0.0026	BDL	N/A	0.8878	0.0103
JF9	0.4087	0.0039	0.1676	0.0032	BDL	N/A	2.135	0.0073
JF10	0.0282	0.0059	0.1611	0.0058	BDL	N/A	1.405	0.0185

The study recorded Copper concentration of the grilled samples to range from 0.0135 to 0.1042 mg/kg with a mean of 0.0470 ± 0.0096 mg/kg. Manganese concentration was from 0.1334 to 0.1841 mg/kg with a mean of 0.1512 ± 0.0048 mg/kg. The samples obtained Iron concentrations ranging from 0.6712 to 2.7210 mg/kg with a mean of 1.3242 ± 0.1881 mg/kg. There was no significant difference existing between the levels of Iron and Manganese in both fresh and grilled sausage samples (p =0.752). However, the Copper levels in both freshly produced and grilled sausages varied significantly (p= 0.020).

Table 2: Manganese, Copper and Iron concentrations in grilled sausage samples

Samples	Concentration in mg/kg							
	Cu	SD	Mn	SD	Pb	SD	Fe	SD
JG1	0.0209	0.0041	0.1444	0.0044	BDL	N/A	0.9427	0.0099
JG2	0.0861	0.0022	0.1656	0.0037	BDL	N/A	1.255	0.0178
JG3	0.0175	0.0055	0.1841	0.0028	BDL	N/A	1.023	0.0052
JG4	0.0135	0.0078	0.1507	0.0029	BDL	N/A	1.072	0.0068
JG5	0.0541	0.0044	0.1521	0.0037	BDL	N/A	1.597	0.0133
JG6	0.0224	0.0052	0.1601	0.0024	BDL	N/A	1.756	0.0037
JG7	0.0472	0.0013	0.1334	0.0066	BDL	N/A	0.8360	0.0057
JG8	0.0463	0.0038	0.1436	0.0017	BDL	N/A	0.6712	0.0132
JG9	0.1042	0.0055	0.1371	0.0042	BDL	N/A	1.369	0.0162
JG10	0.0573	0.0026	0.1410	0.0007	BDL	N/A	2.721	0.0143

BDL= Below Detectable Limits N/A= Not Applicable

The hazard quotients (HQ) recorded from the fresh samples for Copper concentration in children ranged from 9.0135×10^{-6} to 3.3305×10^{-5} and from 1.2094×10^{-6} and 4.0673×10^{-5} for adults. Hazard quotient for Manganese concentration ranged from 1.1981×10^{-5} to 1.7388×10^{-5} for children and from 1.6076×10^{-6} to 2.3329×10^{-6} for adults. Also, Iron concentrations recorded hazard quotients ranging from 1.4539×10^{-5} to 4.0583×10^{-5} for children and from 1.9507×10^{-6} to 5.4451×10^{-6} for adults.

Table 3: Hazard quotient n, Cu aof Mnd Fe in fresh samples collected

Samples	Hazard Quotient (HQ)					
	Cu		Mn		Fe	
	Children	Adults	Children	Adults	Children	Adults
JF1	1.2625×10^{-5}	1.6940×10^{-6}	1.1981×10^{-5}	1.6076×10^{-6}	1.6334×10^{-5}	2.1916×10^{-6}
JF2	8.7320×10^{-5}	1.1729×10^{-5}	1.6045×10^{-5}	2.1529×10^{-6}	2.2466×10^{-5}	3.1041×10^{-6}
JF3	3.1995×10^{-5}	4.2928×10^{-6}	1.7388×10^{-5}	2.3329×10^{-6}	1.6856×10^{-5}	2.2616×10^{-6}
JF4	2.1351×10^{-5}	2.8645×10^{-6}	1.4721×10^{-5}	1.9751×10^{-6}	1.9306×10^{-5}	2.5903×10^{-6}
JF5	1.1305×10^{-4}	1.5169×10^{-5}	1.5123×10^{-5}	2.0291×10^{-6}	1.6186×10^{-5}	2.1717×10^{-6}
JF6	1.8839×10^{-4}	2.5278×10^{-5}	1.7296×10^{-5}	2.3207×10^{-6}	4.0583×10^{-5}	5.4451×10^{-6}
JF7	3.0313×10^{-4}	4.0673×10^{-5}	1.442×10^{-5}	1.9347×10^{-6}	1.4539×10^{-5}	1.9507×10^{-6}
JF8	7.946×10^{-5}	1.0661×10^{-5}	1.4219×10^{-5}	1.9212×10^{-6}	1.6261×10^{-5}	2.1756×10^{-6}
JF9	1.3063×10^{-4}	1.7527×10^{-5}	1.5306×10^{-5}	2.0536×10^{-6}	3.8994×10^{-5}	5.2026×10^{-6}
JF10	9.0135×10^{-6}	1.2094×10^{-6}	1.4712×10^{-5}	1.9739×10^{-6}	2.5661×10^{-5}	3.443×10^{-6}

Hazard quotient for Copper concentration in grilled sausage samples ranged from 4.315×10^{-4} to 3.3305×10^{-5} for children and from 5.7895×10^{-7} to 4.4685×10^{-6} for adults. Manganese recorded a hazard quotient ranging from 1.2182×10^{-5} to 1.6812×10^{-5} for children and from 1.6345×10^{-6} to 2.2558×10^{-6} for adults.

Hazard quotient for Iron concentration in children ranged from 1.2259×10^{-5} to 4.9697×10^{-5} for children and from 1.6449×10^{-6} to 3.9136×10^{-5} for adults. There is no possibility of any adverse effect as the hazard quotient is less than one.

Table 4: Hazard quotient of Mn, Cu and Fe in grilled samples collected

Samples	Hazard Quotient (HQ)					
	Cu		Mn		Fe	
	Children	Adults	Children	Adults	Children	Adults
JG1	6.6803×10^{-6}	8.963×10^{-7}	1.3187×10^{-5}	1.7693×10^{-6}	1.4646×10^{-5}	2.4506×10^{-6}
JG2	2.752×10^{-5}	3.6925×10^{-6}	1.5123×10^{-5}	2.0291×10^{-6}	2.2921×10^{-5}	3.0754×10^{-6}
JG3	5.5935×10^{-6}	7.505×10^{-7}	1.6812×10^{-5}	2.2558×10^{-6}	1.8684×10^{-5}	2.507×10^{-6}
JG4	4.315×10^{-6}	5.7895×10^{-7}	1.3762×10^{-5}	1.8465×10^{-6}	1.958×10^{-5}	2.627×10^{-6}
JG5	1.7292×10^{-5}	2.3201×10^{-6}	1.389×10^{-5}	1.8636×10^{-6}	2.9169×10^{-5}	3.9136×10^{-5}
JG6	7.1595×10^{-6}	9.6063×10^{-7}	1.4621×10^{-5}	1.9671×10^{-6}	3.2071×10^{-5}	4.3031×10^{-6}
JG7	1.5086×10^{-5}	2.0242×10^{-6}	1.2182×10^{-5}	1.6345×10^{-6}	1.5269×10^{-5}	2.0487×10^{-6}
JG8	1.4799×10^{-5}	1.9856×10^{-6}	1.3114×10^{-5}	1.7595×10^{-6}	1.2259×10^{-5}	1.6449×10^{-6}
JG9	3.3305×10^{-5}	4.4685×10^{-6}	1.252×10^{-5}	1.6799×10^{-6}	2.5004×10^{-5}	3.3549×10^{-6}
JG10	1.8315×10^{-5}	2.4573×10^{-6}	1.2876×10^{-5}	1.7276×10^{-6}	4.9697×10^{-5}	6.668×10^{-6}

Fresh samples recorded a HI ranging from 4.094×10^{-5} to 3.3209×10^{-4} for children and from 5.4932×10^{-6} to 4.5558×10^{-5} for adults while the HI of the grilled samples ranged from 3.4513×10^{-5} to 8.0888×10^{-5} mg/kg for children and from 5.0525×10^{-6} to 4.3319×10^{-5} mg/kg for adults. This implies no cancer risk as hazard quotient is less than one.

Table 5: Hazard index (HI) for fresh sausage samples

	Hazard Index (HI)	
Samples	Children	Adults
JF1	4.094×10^{-5}	5.4932×10^{-6}
JF2	1.2583×10^{-4}	1.6986×10^{-5}
JF3	6.6239×10^{-5}	8.8873×10^{-6}
JF4	5.5378×10^{-5}	7.4299×10^{-6}
JF5	1.4436×10^{-4}	1.9369×10^{-5}
JF6	2.4627×10^{-4}	3.3044×10^{-5}
JF7	3.3209×10^{-4}	4.4558×10^{-5}
JF8	1.0994×10^{-4}	1.4758×10^{-5}
JF9	1.8493×10^{-4}	2.4783×10^{-5}
JF10	4.9387×10^{-5}	6.6263×10^{-6}

Table 6: Hazard index (HI) fresh samples collected

	Hazard Index (HI)	
Samples	Children	Adults
JG1	3.4513×10^{-5}	5.1162×10^{-6}
JG2	6.5564×10^{-5}	8.797×10^{-6}
JG3	4.1089×10^{-5}	5.5133×10^{-6}
JG4	3.7657×10^{-5}	5.0525×10^{-6}
JG5	6.0351×10^{-5}	4.3319×10^{-5}
JG6	5.3852×10^{-5}	7.2308×10^{-6}
JG7	4.2537×10^{-5}	5.7074×10^{-6}
JG8	4.0172×10^{-5}	5.3900×10^{-6}
JG9	7.0829×10^{-5}	9.5033×10^{-6}
JG10	8.0888×10^{-5}	1.0853×10^{-5}

Discussion of Findings

Lead (Pb) is toxic when taken into the body as it leads to lower cognitive and intellectual growth in kids, raises blood pressure and cardiovascular disease in adults (Lanphear et al., 2018). The levels of Lead in raw and grilled sausages from the central business district of the Tamale metropolis analyzed were below detectable limits. This may be as a result of the animals used for the sausage production not being exposed to water, feed and air contaminated by Lead prior to the production process. However, Gabriel et al. (2014) found levels of Lead in sausage (0.82 mg/kg) sold in Romania.

Nkansah and Ansah (2014) also found levels of Lead in all meat samples analyzed to exceed permissible limits ascribing this to the use of Lead containing products during meat processing. Muller and Anke (1995) also found out that Lead levels were higher in sausage samples compared to the raw meat used in their manufacture. They associated the spices used to make sausages as a possible cause for this increment. Copper levels were higher in the raw samples than in the grilled samples which can be as a result of the pre-exposure of the animals to heavy metals contaminated feed, water or even air prior to the production process. In most cases, when the milling machine is faulty, leaching of metals occurs into the meat during the production process, indicating a possible source of Copper into the sausage. The addition of spices into the meat during the production process could also account for the high levels of Copper in the raw samples than in the grilled sausages. However, the grilled samples recording reduced Copper levels may be as a result of the Copper volatizing on heating or binding to other elements present in the sausage sample to form new compounds. About 70% of the samples recorded Copper levels to be below the maximum permissible limits. However, the remaining 30% of the samples obtained Copper levels above the recommended levels. Zahran and Hendy (2015) recorded Cu levels in meat and sausage samples ranging from 1.44 to 6.92 mg/kg with a mean of 3.45 ± 0.38 mg/kg and from 19.83 to 65.21 mg/kg with a mean of 38.59 ± 2.96 mg/kg, respectively which were very high as compared to the permissible level of 0.10 mg/kg as indicated by the FAO/WHO (2000).

This study recorded reduced Manganese (Mn) levels for grilled samples. This may be due to the heat treatment and water association of Mn (Kobia et al., 2016). Inobeme et al. (2018) also recorded a decrease in concentration of Mn and Cu after smoking and grilling of goat meat and beef. Manganese concentrations in all the samples used for this study were within the permissible limits of 0.3 mg/kg set by FAO/WHO (2000). Contrarily, Zahran and Hendy (2015) recorded a mean Mn level of 1.95 ± 0.17 mg/kg (within the range of 0.80 - 3.27 mg/kg) and 8.40 ± 0.97 mg/kg (within the range of 3.32 - 18.38 mg/kg) for meat and sausages respectively, which were above the permissible level indicated by FAO/WHO (2000).

Greater levels of Iron (Fe) were recorded in grilled sausages than in the raw sausages used in this study. This can be as a result of the interactions between the sausage and the ungalvanised metal grid used in the grilling process which is often made of Iron. Adzitey et al. (2018) also recorded higher concentrations of Iron in grilled beef than the raw meat collected from Otano, Ghana. This rise in Fe levels in the grilled meat samples was attributed to the seasonings added to the meat during grilling. This was affirmed by Nkansah and Cosmos (2010) who reported the presence of some heavy metals in local spices used in kebab preparation. In this study, Iron levels were higher than the maximum permissible level of 0.3 mg/kg (FAO/WHO, 2000). Zahran and Hendy (2015) recorded Fe levels ranging from 82.95 to 352.9 mg/kg and a mean of 190.5 ± 19.43 mg/kg in meat and from 82.9 to 270 mg/kg with a mean of 135.0 ± 10.48 mg/kg in sausage sampled from Egyptian markets. These levels were also greater than the acceptable levels by FAO/WHO (2000). Alturiqi and Albedair (2012) also recorded high levels in sausage (242.44± 12.09 mg/kg) in Saudi Arabia.

The calculated hazard quotient (HQ) and hazard index (HI) were less than one (1) indicating no possibilities of adverse effects and non-carcinogenic effects associated with consuming the sausages (Table 3 and 4).

Conclusion

Lead concentration was below detectable limits in all the samples analyzed. Manganese concentration for all the samples were within the maximum permissible level. For Copper concentration, more than half of the samples collected recorded levels within the recommended levels. However, a few samples obtained levels more than the acceptable levels set by the World Health Organization and the Food and Agriculture Organization. Copper is vital for energy production in cells and required for women's fertility in relation to estrogen metabolism. It is also required for Iron transport in the body. Iron levels in all the sampled sausages recorded higher levels above the maximum allowable levels. Iron overload increases the risk for liver diseases, heart attack and many others. Though some of the metals recorded levels exceeding the recommended limits, the concentrations of all the metals from the study possessed no adverse and non-carcinogenic effects. This indicates that the sausages are wholesome for consumption.

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