Determination of polycyclic aromatic hydrocarbons and potentially toxic metals in commonly consumed beef sausage roll products in Nigeria

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ARTICLE INFO

Keywords:
Analytical chemistry
Food analysis
Food safety
Polycyclic aromatic hydrocarbons
Toxic metals
Sausage rolls
Health
Nigeria

ABSTRACT

Levels of polycyclic aromatic hydrocarbons (PAHs) and potentially toxic metals (PTMs) were determined in the commonly consumed beef sausage roll products (coded BS1 – BS6) in Nigeria. This was done in order to assess the safety of regular consumption of these products with respect to the substances determined. Three batches of six samples of beef sausage roll products were collected from Ile-Ife, Osun state, Nigeria. A part of the pretreated sample was Soxhlet extracted using n-hexane and analyzed with Gas Chromatography coupled with Flame Ionization Detector (GC-FID) to identify and quantify each of the PAHs in the sample, while Atomic Absorption Spectrometer (AAS) was used to profile the concentrations of As, Cd, Co, Cu, Pd, Mn and Zn in the digested sausage roll samples. Levels of PAHs in the samples ranged from 1.84 μg/g of Acenaphthylene in BS5 to 282.83 μg/g of Benzo[k]fluoranthene in BS1. Concentrations of benzo[a]pyrene in all the samples were higher than the guideline value of 0.003 mg/kg/day. For PTMs, a range of 0.075 μg/g As in both BS1 and BS6 to 2.950 μg/g Cu in BS3 was obtained. The study concluded that both PAHs and PTMs occurred in the samples at levels that called for caution on the part of consumers to prevent health infarctions that might be associated with prolonged regular and large consumption of beef sausage roll products.

1. Introduction

Packaged beef sausage roll is a popular snack commonly eaten in Nigeria and it comes with various trademarked names. Generally, beef sausage roll is made from pastry dough of plain flour formed into tubes around rolls of spiced minced meat and then baked.

Polycyclic aromatic hydrocarbons (PAHs) are widespread environmental contaminants that may originate from a variety of incomplete combustion and pyrolysis processes resulting from anthropogenic and natural sources (Lee and Shin, 2007; Varlet et al., 2007; Kim and Kim, 2015). A high amount of PAHs is emitted from processing coal and during the incomplete combustion of organic matter such as fuel oils (Kishikawa et al., 2003). Many of the PAHs, such as benzo[a]pyrene, benzo[a]anthracene, dibenzo[a,h]anthracene and chrysene, have been reported to possess carcinogenic and genotoxic properties (Poster et al., 2006; Rengarajan et al., 2015; Yebra-Pimentel et al., 2015). It is well documented that sources of PAHs in food items include contamination by air pollutants, uptake from soil and carbonization of carbohydrates, fats and proteins during food processing that involves smoking or high-temperature cooking (European Commission, 2002). Existing evidence has indicated that there is a strong relationship between exposure to environmental PAHs and incidence of cancer (Shen et al., 2017).

Alomirah et al. (2011) maintained that oils and fats are the main entrance sources of PAHs in food as fat is known to have played a major role in the enhancement of PAH formation via processes that involve thermal applications such as baking of bread and other flour products in which case high temperatures and addition of fat are employed for enhanced palatability. High molecular weight PAHs, such as benzo[a]pyrene, indeno[1,2,3-c,d]pyrene, dibenzo[a,h]anthracene, and benzo[g,h,i]perylene, are more stable and toxic than the low molecular weight PAHs (Ekhator et al., 2018).

Changes in socioeconomic status, tendency towards modernization, the need to meet up with appointments and general convenience are some of the factors that have made people to go for fast foods. Consequently, consumption of such fast foods as baked and packaged flour products, sausage rolls containing grilled meat, among others, has
increased dramatically in most towns and cities of Nigeria. Whereas grilled and smoked foods are capable of contributing significantly to the intake of PAHs if food items containing PAHs form a large part of the usual diet, there is a knowledge gap between what people consume and the inherent health hazards. For example, there is paucity of data on the levels of PAHs and similar xenobiotics in baked flour products and sausage rolls that are being increasingly consumed in Nigeria. Hence, enforcing regulations that guide the maximum allowable levels of such xenobiotics remains a major challenge.

Of equal health concern globally is the increasing presence of potentially toxic metals (PTMs) in the environment and as contaminants in food items. The exponential use of potentially toxic metals in several industrial, agricultural, domestic and technological applications has led to increased human exposure (Bradl, 2002). Metal ions have been found to interact with cell components such as DNA and nuclear proteins, causing DNA damage and conformational changes that may lead to cell cycle modulation, carcinogenesis or apoptosis (Beyersmann and Hartwig, 2008). Several laboratory studies have demonstrated that reactive oxygen species production and oxidative stress play a key role in the toxicity and carcinogenicity of arsenic, cadmium, chromium, lead and mercury (Tchounwou et al., 2001, 2004; Sutton et al., 2002; Yedjou and Tchounwou, 2008; Patellola et al., 2009).

This study investigated the levels of PAHs and PTMs in beef sausage rolls commonly consumed in Ile-Ife and its environs across various age groups. This was with a view to evaluating the extent to which the beef sausage rolls conformed to safety guidelines with respect to their PAHs and PTMs contents.

2. Methodology

2.1. Sample preparation and pretreatment

The samples collected were three batches of six types of beef sausage rolls which added up to eighteen samples. The samples were purchased from shops at Obafemi Awolowo University, Ile-Ife and other outlets within the town. The samples were kept in zip lock bags and stored at a temperature of about 4 °C in a deep freezer. The beef sausage samples were ground using mortar and pestle into powdered form. Samples for extraction and digestion were selected by coning and quartering method. Further analysis was carried out before the expiry dates of the analysed samples.

2.2. Sterilization of apparatus

All standard laboratory glassware (volumetric flask, beakers, watch glass, stirring rod, sample bottles) were cleaned by scrubbing with a nylon brush in a detergent solution in a wash basin, rinsed with tap water until no more soap was observed. They were then rinsed with distilled water and acetone. Glass vials to be used for PAHs analysis were soaked in 10% nitric acid for 72 h and then were rinsed with distilled water and acetone and dried dry.

2.3. Extraction of PAHs and clean-up procedure

Components of the Soxhlet extraction setup (condenser, flask, soxhlet extractor) were properly washed and then rinsed with acetone and n-hexane. For extraction, the flask was half-filled with the solvent (n-hexane) and 20 g of each sample was weighed into a pre-extracted thimble and fitted into the Soxhlet extractor. This was extracted until the solvent in the soxhlet extractor was permanently colourless.

For the clean-up procedure, silica gel was used as the stationary phase and n-hexane as the mobile phase. Little quantity of glass wool was used as a plug to prevent the loss of the stationary phase at the bottom of the column before the addition of silica gel. Anhydrous sodium sulphate was added on top of the silica gel. The packed column was added on top of the silica gel. The packed column was

2.4. Determination of PAHs

The qualitative identification and quantification of the PAHs were carried out using GC-FID available at the Nigerian Institute of Oceanography and Marine Research, Victoria Island, Lagos, Nigeria. The identification of PAHs was based on the comparison of the retention times of the peaks with those obtained from the mixture of PAHs standards purchased and provided by the analyzing laboratory. Quantification was based on external calibration curves prepared from the standard samples of each of the PAHs.

2.5. Sample digestion for PTMs analysis

One gram (1 g) of each sample was weighed into a Teflon beaker, 10 mL of concentrated HNO3 was added and the beaker was covered with a watch glass. The sample was heated over a thermostated hot plate for 2 h at a temperature of about 130 °C, while HNO3 was added in little quantities at intervals to prevent the sample from total drying. The sample was then brought down to simmer after which 1 mL of HClO4 was added and the sample was digested further for an additional period of 30 min at a temperature of 150 °C. The content of the beaker was quantitatively transferred into a 25 mL volumetric flask and made up with distilled water to the mark. It was then transferred into a plastic vial in readiness for analysis using Atomic Absorption Spectrophotometer (AAS) at the Centre for Energy Research and Development, Obafemi Awolowo University, Ile-Ife, Nigeria.

2.6. Quality control measures adopted

2.6.1. Blank determination

Ten millilitres (10 mL) of concentrated HNO3 was added into a beaker and the beaker was covered with a watch glass. The content was heated over a thermostated hot plate for 2 h then the beaker was brought down and 1 mL HClO4 was added. The content was heated further for about 30 min and cooled. The content of the beaker was quantitatively transferred into a 25 mL volumetric flask and made up distilled water to the mark. It was then poured into a plastic vial and stored prior to analysis using AAS.

2.6.2. Recovery experiment for PAHs

The percentage recovery (%R) was carried out by introducing 10 µg/mL of the available PAHs, namely: fluorene, anthracene, phenanthrene and chrysene into a known amount (A) of the pulverized beef sausage sample while an equally known amount of the beef sausage sample (B) was left unspiked. Both samples were taken through the extraction and clean-up stages. The extracts were analyzed for their PAHs content. The %R was evaluated from the relationship:

\[
% R = \frac{A - B}{10} \times 100 \tag{2.1}
\]

2.7. Human health risk assessment

2.7.1. The dietary daily intake (DDI) of PAHs

The Dietary Daily Intake (DDI) of PAHs in the beef sausage products was evaluated using the relationship adopted by Halek et al. (2007). The dietary intake of PAHs from beef sausage samples was evaluated by multiplying the respective PAH concentration in each beef sausage sample by the beef sausage ingestion rate (IR). Evaluation of Dietary Daily Intake (DDI) was calculated for individual PAHs, the sum of the 16 PAHs analyzed (total PAHs) and also for the sum of those PAHs
For As, it is 3 and 10.03 mg/kg/day for Co (Finley et al., 2012). When the index is more than 3 mg/kg/day respectively (USEPA, 2002), for Pb it is 3.5 μg/g (NIOSH, 1997). Three of the samples (BS1, BS2 and BS3) had naphthalene values above this range. Symptoms of exposure to elevated levels of naphthalene include nausea, vomiting, diarrhea, blood in the urine, jaundice and prolonged exposure to large amounts of naphthalene may destroy red blood cells as commonly observed in people with an underlying glucose-6-phosphate dehydrogenase (G6PD) deficiency (Santucci and Shah, 2000). By implication, regular consumptions of BS1, BS2 and BS3 over a long period of time could pose health challenges for humans, while BS4, BS5 and BS6 could be consumed without developing health challenges related to elevated naphthalene levels.

acenaphthylene levels ranged from 1.84 μg/g in BS5 to 9.18 μg/g in BS1. The exposure limit for naphthalene as regulated by the National Institute for Occupational Safety and Health (NIOSH) is 10–15 μg/g (NIOSH, 1997). Three of the samples (BS1, BS2 and BS3) had naphthalene values above this range. Symptoms of exposure to elevated levels of naphthalene include nausea, vomiting, diarrhea, blood in the urine, jaundice and prolonged exposure to large amounts of naphthalene may destroy red blood cells as commonly observed in people with an underlying glucose-6-phosphate dehydrogenase (G6PD) deficiency (Santucci and Shah, 2000). By implication, regular consumptions of BS1, BS2 and BS3 over a long period of time could pose health challenges for humans, while BS4, BS5 and BS6 could be consumed without developing health challenges related to elevated naphthalene levels.

acenaphthylene in the beef sausage roll samples were higher than the recommended amount. Hence, depending on the regularity of consumption and the amount consumed per time, irritation of the eyes, skin irritation, respiratory system discomfort, wheezing, shortness of breath, bronchitis, vomiting, kidney and liver damage could manifest as symptoms of systemic acenaphthylene overload in the consumers.

3.2. Levels (μg/g) of polycyclic aromatic hydrocarbons in sausage samples

A summary of the total concentrations of various PAHs (μg/g) present in the sausage roll samples (Table 2) ranged between 228.18 in BS6 and 967.88 in BS1 and occurred in the order BS6 (228.18) < BS3 (255.22) < BS2 (297.63) < BS4 (299.80) < BS5 (331.59) < BS1 (967.88). These values were higher than the 0.098–0.102 μg/g and 3.237–1.702 μg/g in sausage products respectively obtained by Lorenzo et al. (2011) and Roseiro et al. (2012).

Specifically, the concentration of naphthalene in the beef sausage samples ranged from 5.00 in BS5 to 55.68 μg/g in BS1. The exposure limit for naphthalene as regulated by the National Institute for Occupational Safety and Health (NIOSH) is 10–15 μg/g (NIOSH, 1997). Three of the samples (BS1, BS2 and BS3) had naphthalene values above this range. Symptoms of exposure to elevated levels of naphthalene include nausea, vomiting, diarrhea, blood in the urine, jaundice and prolonged exposure to large amounts of naphthalene may destroy red blood cells as commonly observed in people with an underlying glucose-6-phosphate dehydrogenase (G6PD) deficiency (Santucci and Shah, 2000). By implication, regular consumptions of BS1, BS2 and BS3 over a long period of time could pose health challenges for humans, while BS4, BS5 and BS6 could be consumed without developing health challenges related to elevated naphthalene levels.

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Levels of acenaphthylene ranged from “below detection limit (BDL)” in BS3, BS4, BS5 and BS6 to 19.34 μg/g in BS1. The daily oral exposure likely to be without an appreciable risk of deleterious effects during a lifetime for acenaphthene is 0.06 μg/g/day (or mg/kg/day) as the recommended daily oral exposure to acenaphthylene for humans. The concentrations of acenaphthene in the beef sausage roll samples were higher than the recommended amount. Hence, depending on the regularity of consumption and the amount consumed per time, irritation of the eyes, skin irritation, respiratory system discomfort, wheezing, shortness of breath, bronchitis, vomiting, kidney and liver damage could manifest as symptoms of systemic acenaphthylene overload in the consumers.

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levels ranged from 15.67 μg/g in BS4 to 55.68 μg/g in BS1.

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animal bioassays, USEPA (1987, 1993) to have placed Phenanthrene in weight-of-evidence, not classifiable as to human carcinogenicity.

Anthracene levels ranged from 3.73 to 26.85 μg/g, BS5 had the lowest concentration while BS2 had the highest concentration. Exposure to high doses of Anthracene at a short time causes damage to the skin, headaches, nausea, loss of appetite, inflammation or swelling of the stomach and intestines. Levels of Anthracene in the samples exceeded the daily oral exposure limit of 0.3 mg/kg/day for Anthracene (USEPA, 1990). In the case of Fluoranthene, its levels ranged from 1.90 to 6.69 μg/g. The daily oral exposure limit of Fluoranthene is 0.04 mg/kg/day (USEPA). All the samples analyzed were above the exposure limit except BS2 in which fluoranthene was not found. For Pyrene, detected levels ranged from 4.08 to 15.67 μg/g. Sample BS2 had the lowest concentration while BS1 had the highest concentration. Pyrene is a skin irritant, a suspected mutagen, and an equivocal tumour-causing agent. Workers exposed to 3 μg/g and 18.71 μg/g in BS6 to 34.44 μg/g in BS5, while its level was BDL (below detection limit) in BS1. Benzo[a]anthracene can cause irritation of eyes, nose, throat and skin. Also, levels of Anthracene in the samples exceeded the daily oral exposure (0.3 mg/kg/day) for anthracene as recommended by the USEPA (1990). Chrysene levels ranged from 5.85 μg/g in BS6 to 31.64 μg/g in BS1. Chrysene has been classified in weight-of-evidence Group B2, that is, it is a probable human carcinogen. For Chrysene, the recommended human exposure limit is 0.007 mg/kg (USEPA). Obviously, the samples had concentrations far above the recommended limit, which by implication, could portend health challenges for long-term reliance on frequent consumption of the sausage rolls. Benzo[b]fluoranthene levels in the samples ranged from 22.26 to 151.59 μg/g. The National Institute for Occupational Safety and Health Administration (NIOSH, 1997) has set a recommended exposure limit of 0.1 mg/kg for Benzo[b]fluoranthene, a limit that was significantly exceeded in all the samples. Benzo[b]fluoranthene has been classified to be genotoxic and carcinogenic by World Health Organization and Environmental Protection Agency. Amount of Benzo[k]fluoranthene detected in the samples fell between 19.71 and 282.83 μg/g. Recommended exposure limit for Benzo[k]fluoranthene, a carcinogenic substance, is 0.1 μg/g (NIOSH, 1997).

TEF values for the PAHs was adopted from (Nisbet and LaGoy, 1992).

**Dietary Daily Intake - DDI, The carcinogenic potencies – BaPTEQ.**

| Carcinogenic Risk Index | BS1 | BS2 | BS3 | BS4 | BS5 | BS6 |
|-------------------------|-----|-----|-----|-----|-----|-----|
| DDI (mg/day)            | 569.82 | 23.81 | 20.42 | 23.98 | 26.56 | 18.25 |
| DDI for carcinogenic PAHs (mg/day) | 524.95 | 9.89 | 70.42 | 13.09 | 19.61 | 9.96 |
| TEQ (μg/g)              | 322.64 | 57.85 | 28.48 | 152.04 | 232.13 | 131.50 |

### Table 3
Estimated dietary daily intake (DDI) and carcinogenic potencies of PAHs.

| PAH             | TEF | BS1 | BS2 | BS3 | BS4 | BS5 | BS6 |
|-----------------|-----|-----|-----|-----|-----|-----|-----|
|                 |     | DDI | BaPTEQ | DDI | BaPTEQ | DDI | BaPTEQ | DDI | BaPTEQ | DDI | BaPTEQ | DDI | BaPTEQ |
| Naphthalene     | 0.001 | 4.45 | 0.056 | 1.52 | 0.019 | 1.46 | 0.018 | 0.84 | 0.011 | 0.40 | 0.005 | 0.96 | 0.012 |
| Acenaphthylene  | 0.001 | 0.73 | 0.009 | 0.22 | 0.003 | 0.35 | 0.004 | 0.21 | 0.003 | 0.15 | 0.002 | 0.18 | 0.002 |
| Acenaphthene    | 0.001 | 9.47 | 0.019 | 0.91 | 0.010 | 0.00 | 0.000 | 0.000 | 0.000 | 0.00 | 0.000 | 0.00 | 0.000 |
| Phenanthrene    | 0.001 | 12.34 | 0.154 | 5.38 | 0.070 | 5.52 | 0.069 | 4.66 | 0.058 | 3.41 | 0.043 | 3.29 | 0.041 |
| Anthracene      | 0.100 | 2.15 | 2.685 | 0.91 | 1.139 | 0.75 | 0.939 | 0.43 | 0.536 | 0.30 | 0.373 | 0.30 | 0.376 |
| Fluoranthene    | 0.001 | 0.54 | 0.001 | 0.00 | 0.000 | 0.31 | 0.004 | 0.29 | 0.004 | 0.20 | 0.002 | 0.15 | 0.002 |
| Pyrene          | 0.001 | 1.25 | 0.016 | 0.39 | 0.005 | 0.92 | 0.011 | 0.50 | 0.006 | 0.33 | 0.004 | 0.46 | 0.006 |
| Benzo[a]anthracene | 0.100 | 1.70 | 2.122 | 0.86 | 1.074 | 0.15 | 1.817 | 0.98 | 1.227 | 0.48 | 0.602 | 0.43 | 0.054 |
| Benzo[k]floranthene | 0.100 | 506.23 | 28.283 | 2.64 | 3.304 | 1.58 | 1.971 | 4.06 | 5.072 | 5.25 | 6.568 | 2.34 | 2.925 |
| Benzo[a]pyrene  | 0.100 | 3.85 | 4.816 | 1.55 | 1.937 | 0.94 | 1.173 | 1.49 | 1.861 | 2.38 | 2.974 | 1.33 | 1.665 |
| Dibenzo[a,h]anthracene | 5.000 | 2.12 | 132.450 | 4.01 | 50.130 | 1.78 | 22.620 | 3.31 | 41.410 | 3.70 | 46.210 | 3.19 | 39.830 |
| Indeno[1,2,3-cd]pyrene | 0.100 | 0.00 | 0.000 | 0.00 | 0.000 | 0.00 | 0.000 | 0.00 | 0.000 | 2.21 | 2.765 | 0.00 | 0.000 |
| Benzo[g,h,i]perylene | 0.010 | 0.00 | 0.000 | 0.00 | 0.000 | 0.00 | 0.000 | 0.00 | 0.000 | 1.50 | 0.187 | 0.83 | 0.104 |

The estimated dietary daily intake (DDI) values of selected PAHs in the beef sausage samples are presented in Table 3.

3.3. Health risk assessment of polycyclic aromatic hydrocarbons in beef sausage

3.3.1. Dietary daily intake (DDI) of PAHs from consumption of beef sausage

Table 3 summarizes the dietary daily intake (DDI) of PAHs in the analyzed beef sausage roll samples. Dietary daily intake values (mg/day)
estimated from individual PAH concentrations in beef sausage rolls ranged from 0 – 502.63 (BS1), 0 – 5.58 (BS2), 0 – 5.52 (BS3), 0 – 4.66 (BS4), 0 – 5.25 (BS5), and 0 – 3.29 (BS6). The DDI (mg/day) of Benzo[a]pyrene (BaP), the most extensively studied carcinogenic PAH, is classified by IARC as a Group 1 or known human carcinogen (IARC, 1990) were 3.85, 3.3.2. Individual PAH carcinogenic potencies (BaPTEQ) consumption of BS1 in preference to other beef sausage products might carcinogenic PAHs was highest for BS1. The results indicate that the 524.95, 9.89, 70.42, 12.09, 19.61 and 9.96 forBS1, BS2, BS3, BS4, BS5 523 (Table 4), while the DDI (mg/day) for the total carcinogenic PAHs were 23.98, 26.56 and 18.25 for BS1, BS2, BS3, BS4, BS5 and BS6 respectively. PAHs in the assessed beef sausage samples were 569.82, 23.81, 20.42, respectively.

1.55, 0.94, 1.49, 2.38 and 1.33 for BS1, BS2, BS3, BS4, BS5 and BS6, (Table 4). The DDIs for BaPTEQ were 322.64, 57.85, 28.48, 152.04, 232.13 and 131.50 (BaP), the most extensively studied carcinogenic PAH, is classi (85.9). It has been suggested that compounds that have high carcinogenic the highest carcinogenic potency in BS1 (151.59), BS2 (50.13) and BS3 clinical signs of which include burning lips, painful swallowing, thirst, skin hyperpigmentation, hyperkeratosis, nasal congestion, and abdom-

inal pain. The major source of human exposure is food. Epidemiologic studies have linked chronic arsenic exposure to various cancers, including skin, lungs, and lymph glands (USEPA, 1997). Arsenic may also replace phosphorus in bone tissue and be stored for years. Some of the effects of arsenic acute poisoning are severe vomiting, watery and bloody diarrhea, severe abdominal pain, burning esophageal pain (which may occur within 30 min to 2 h), vasodilatation, myocardial depression, cerebral edema, and distal peripheral neuropathy. Later stages of poisoning include jaundice, renal failure, gangrene of the extremities, anemia, cancer of the skin, lung, and nasal tissue and death (resulting from cir-

cural failure within 24 h to 4 days).

For cobalt (Co), the levels detected were in the range 0.275 ± 0.013 μg/g in BS1 to 1.100 ± 0.028 μg/g in BS5. Exposure to excessive levels of Co causes allergic dermatitis, rhinitis and asthma. Levels of cobalt in the sample also exceeded the recommended oral exposure limit of 0.03 mg/ (Finley et al., 2012). The amount of cadmium determined in the beef sausage samples ranged from 0.200 ± 0.028 μg/g in BS6 to 0.650 ± 0.023 μg/g in BS4. Again, levels of cadmium in the samples exceeded the USEPA oral exposure limit of cadmium which is 0.001 mg/kg/day (USEPA, 1997). Cadmium has been linked to skeletal damage (Järup, 2003). Cadmium is also known to harm the reproductive system and embryonic development (Thompson and Bannigan, 2008). Acute effects of exposure to cadmium include nausea, vomiting, and abdominal pain (Hodgson, 2010). The main organ damaged following long-term exposure is the kidney, with the proximal tubules being the primary site of action. In the case of copper, the amount detected was in the range of 0.500 ± 0.010 μg/g in BS6 to 2.950 ± 0.023 μg/g in BS3. From the results obtained, these concentrations were below the permissible level of Cu (10 μg/g in foods (Salama and Radwan, 2005). The samples therefore can be considered free of copper contamination. Copper serves as an antioxidant and helps the body to remove free radicals and prevent cell structure damage (Salama and Ridwan, 2005) if it occurs at moderate levels in the body, but at higher levels, it could result in liver disease and severe neurological defects (Uriu-Adams and Keen, 2005). Manganese levels in the beef sausage samples ranged from 0.125 ± 0.005 (μg/g) in BS6 to 1.000 ± 0.025 (μg/g) in BS5. The levels of Mn found in the beef sausage samples in this study were below the daily intake of 2–3 mg/day recommended by WHO (WHO, 1993). Manganese plays a role in bone mineralization, protein and energy metabolism, metabolism regulation, cellular protection from damaging free radical species, and the formation of glycosaminoglycans (ATSDR, 2012). A diet deficient in Mn could lead to poor growth and impaired reproduction (Hidiroglou, 1979).

In the case of lead, its levels were in the range 0.075 ± 0.008 (μg/g) in BS6 to 0.35 ± 0.028 (μg/g) in BS5. k limit for Pb is 3.5 × 10−3 mg/kg/day (USEPA, 1997). The concentration of lead in the samples clearly exceeded the recommended limit by 2–100 folds. Lead in the body affects the nervous system, kidneys and blood (Aast et al., 2016). Exposure of lead causes tumors in the body and USEPA has classified lead as a probable human carcinogen (Group 2 B xenobiotics). Inorganic and organic lead may be absorbed through the gastrointestinal tract, the respiratory sys-

tem, and the skin; ingested inorganic lead is absorbed more efficiently from the gastrointestinal tract of children than that of adults (Hodgson, 2010). The main targets of lead toxicity are the reproductive system, hematopoietic system and the nervous system, especially in infants and young children in whom the nervous system is still developing. Lead interferes in the biosynthesis of porphyrins and heme (Nuwaybid et al., 2003; Hodgson, 2010). Even at low levels of exposure, children, and
sometime, adults may show hyperactivity, decreased attention span, mental deficiencies, impaired vision, fatigue, sleep disturbances, anemia, colic, neuritis. At higher levels, encephalopathy may occur in addition to delirium, hallucinations, ataxia, stupor, convulsions, coma, and even death in both children and adults (Goyer and Clarkson, 2001; Hodgson, 2010). Lead exposure can cause male and female reproductive toxicity, miscarriages, and degenerate offspring (Hodgson, 2010). Lead exposure can cause male and female reproductive toxicity, miscarriages, and degenerate offspring (Hodgson, 2010). Lead exposure can cause male and female reproductive toxicity, miscarriages, and degenerate offspring (Hodgson, 2010).

Zinc levels in the beef sausage samples ranged from 0.825 ± 0.023 (μg/g) in BS6 to 2.800 ± 0.025 (μg/g) in BS5. The required daily intake is 8 mg/day (WHO, 1993). Zinc helps to speed up the healing process after injury (Cohen and David, 2007), supports normal growth and development during pregnancy, childhood, and adolescence and is required for proper sense of taste and smell (Elinder, 1986). However, low levels or excess of zinc in the body has been linked to development of prostate enlargement and cancer in humans (Gonzalez et al., 2009).

#### Table 6

| Metals RfD (μg/kg/day) | BS1 | BS2 | BS3 | BS4 | BS5 | BS6 |
|-----------------------|-----|-----|-----|-----|-----|-----|
| As                    | 0.30| 0.13| 0.43| 0.44| 1.47| 0.40| 1.33|
| Co                    | 300 | 0.80| 0.03| 1.47| 0.05| 1.33| 0.04| 1.73| 0.06| 1.96| 0.07| 0.49| 0.02|
| Cd                    | 0.50| 0.40| 0.80| 0.84| 1.68| 0.80| 1.60| 1.16| 2.32| 1.07| 2.14| 0.36| 0.72|
| Cu                    | 400 | 1.47| 0.04| 4.49| 0.11| 5.24| 0.13| 4.44| 0.11| 4.22| 0.11| 0.89| 0.02|
| Mn                    | 140 | 0.36| 0.01| 1.24| 0.00| 0.98| 0.01| 1.38| 0.01| 1.78| 0.01| 0.22| 0.00|
| Pb                    | 3.50| 0.18| 0.05| 0.58| 0.17| 0.58| 0.17| 0.44| 0.13| 0.62| 0.18| 0.13| 0.04|
| Zn                    | 300 | 2.27| 0.01| 3.91| 0.01| 3.60| 0.01| 4.44| 0.02| 4.98| 0.00| 1.47| 0.01|

3.5. Health risk assessment of toxic metals in beef sausage samples

To assess the health risk associated with toxic metal contamination of beef sausage products, health risk index (HRI) was calculated. The results showed that Arsenic and Cadmium contamination in beef sausage had greatest potential to pose health risk to regular and heavy consumers of some of the sausage rolls (Table 6) as their health risk index was more than 1 in BS2, BS3, BS4 and BS5.

4. Conclusion

Varying concentrations of polycyclic aromatic hydrocarbons were detected in the beef sausage roll samples with the highest concentrations of PAHs in BS1. Sample BS1 had the highest observed values of estimated Dietary Daily Intake (DDI) for the total PAHs and carcinogenic toxic equivalents (TEQ) indicating that there were higher risk of exposure that consumed the samples at levels that suggested the sausage rolls as a major source of exposure to regular and long-term consumers. The total metal load for the sausage rolls (Table 5) shows a range of 2.100 (μg/g) in BS6 to 8.425 (μg/g) in BS6. Possible exposure to metals through the consumption of the sausage rolls was of the order BS6 (2.100) < BS1 (3.150 < BS3 (7.275) < BS2 (7.300) < BS4 (7.950) < BS5 (8.425).

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