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Heavy Metal Concentration and Human Health Risk Assessment of Selected Brands of Seasoning Powder Sold in Port Harcourt, Nigeria

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ABSTRACT

Seasoning powders are frequently used to enhance the taste, aroma, color and overall acceptability of food by consumers. Different brands of seasoning powder are frequently consumed without adequate human health risk assessment of the heavy metals constituents in them. This study was conducted to determine the heavy metal concentration in four (4) brands of seasoning powder (SP1, SP2, SP3 and SP4) sold in Port Harcourt, Nigeria. Ten (10) heavy metals (As, Cr, Ni, Pb, Cd, Fe, Co, Cu, Mn and V) were analyzed by standard methods using Flame Atomic Absorption Spectrophotometer (Model: GBC 908PBMT). Human Health Risk Assessment of heavy metals in the seasoning powders, taken in through ingestion route, was evaluated by computing the Estimated Daily Intake (EDI), Hazard Quotient (HQ), Hazard Index (HI) and Cancer Risk (CR) based on adult human model variables provided by United States Environmental Protection Agency (USEPA). The results showed that mean concentration of Pb and Cd were 6.35 ± 1.07 mg/kg and 0.53 ± 0.09 mg/kg respectively which are above the Food and Agricultural Organization (FAO) and World Health Organization (WHO) Maximum Permissible Limit (MPL) of 5.0 mg/kg and 0.3 mg/kg respectively. Concentration of Fe, Co, Cu, Mn, and V were below the MPL and As and Cr were not detected in all the samples assessed. EDI for the measured heavy metals were less than the tolerable limits specified by USEPA. The HQ and HI values were within acceptable range of <1 except for SP2 with HI value of 1.04. The results obtained for CR were above the specified acceptable range of 1.0 x10^{-6} to1.0 x 10^{-4}. These results suggest possible health concerns associated with the consumption of these seasoning powders in the long term. Strict and systematic surveillance of heavy metals such as Pb and Cd in commonly consumed seasoning powder should be done to ensure safety and quality of the seasoning powders sold in Nigeria.

Key words: Seasoning powder, Heavy metals, Bio-toxicity, Hazard index, Cancer risk.

1.0 INTRODUCTION

Seasoning powders are made of substances used to enhance taste, color and aroma of food. The use of these substances over time had increased in Nigeria owing to the fact that these substances contribute to overall acceptability of food by the ever-growing population. Consequently, several brands of seasoning powders are now constantly advertised and flooding in different markets in the country (Aigberua *et al*., 2018).

In recent times a number of Biochemists, Toxicologists, Food Scientists, Environmental Protection Agencies and the scientific community are getting concerned with the type and concentration of metals in these seasoning powders considering the possibility of toxic heavy metal contamination during processing (Asomugha *et al*., 2016). Heavy metals exert toxicity on important organs affecting

the general health of humans. Several health challenges like cancer, cardiovascular, respiratory and neurological diseases have been associated with heavy metal exposure over time (Gebeyehu *et al*., 2020).

Health Risk Assessment (HRA) for hazardous substances via several routes of exposure has been strongly encouraged by different regulatory authorities due to the bio-toxicity and to address potential human health risk (Fraken *et al*., 2020; Maddela *et al*., 2020; Noventa*et al*., 2021). Heavy metal contamination in foods is becoming an unavoidable problem ((Żukowska and Biziuk, 2008). Strict and periodic surveillance of the constituents in commonly consumed seasoning powder becomes necessary to address potential human health risk.The study was aimed at investigating the concentration of heavy metals in selected brands of seasoning powder commonly sold in Port Harcourt, Rivers State, Nigeria and to use the Human Health Risk Assessment Model to evaluate the potential health risk associated with exposure via ingestion.

2.0 MATERIALS AND METHODS

2.1 Sample collection

Four brands of the commonly consumed seasoning powder (labeled SP1, SP2, SP3 and SP4) were obtained from retail shops in different markets in Port Harcourt metropolis, Nigeria in February 2021. They were brought to the laboratory, sorted out according to brand names, and subjected to analyses.

Table 1: Identification Code for Different Brand of Selected Seasoning Powder

S/N	Brand Name	Code	Ingredients
1	*Spicity Stew and Jollof*	SP1	Salt, flavour enhancer, monosodium glutamate, starch, vegetable fat, vegetable extract, natural colours and spices.
2	*Lasor Chicken*	SP2	West African spices herbs, salt, sugar hydrogenated vegetable oil, flavor enhancer (E62P) and corn starch.
3	*Kitchen Glory*	SP3	Salt, monosodium glutamate, sugar, corn powder, beef seasoning powder, dextrin, vegetable protein powder, soy sauce powder, corn starch, black pepper powder, onion powder, garlic powder.
4	*Onga Classic*	SP4	Iodized salt, flavour enhancers (Glutamate innosinate, guanylate), corn flavour, cassava flour, soy protein powder, spices and spice extracts (cinnamon, rosemary), beef flavour, colorant (E150C).

2.2 Sample Preparation and Digestion

The method described by (Aigberua*et al*., 2018) was used with slight modification. The samples were carefully opened and dried to a constant weight. 2g of each sample was transferred into 100 ml digestive tube and 2 ml concentrated nitric acid (HNO_3), 10 ml of concentrated hydrochloric acid (HCl) and 2 ml H_2SO_4 were added and heated until the evolution of a clear solution. The mixture was left to cool for 30 minutes and filtered into a 50 ml standard volumetric flask which was eventually made up to the 50 ml mark using distilled water. The digested samples were further analyzed for the heavy metals content using Flame Atomic Absorption Spectrophotometer (FAAS) (Model: GBC 908 PB MT). Adequate quality assurance measures were carried out ensure reliability of results.

2.3 Human Health Risk Assessment

In order to estimate the degree of human health concern from ingestion of metals through consumption of seasoning powder, the Estimated Daily Intake (EDI), Hazard Quotient (HQ), Hazard Index (HI) and Cancer Risk (CR) were employed based on an adult human model toxicological variables provided by United States Environmental Protection Agency (USEPA).

2.3.1 Estimated Daily Intake

Estimated Daily Intake (EDI) is used to evaluate the oral exposure dosage for both the carcinogenic and non-carcinogenic metals during the exposure. The EDI of heavy metals via consumption of seasoning powder (SP) was assessed using Equation (1).

$EDIsp = (C \times Isp) / (B.W)$ ……(1)

Where:

EDI = Estimated Daily Intake (mg/kg/day)
C = Mean Concentration of Heavy Metals in Seasoning Powder
Isp = Seasoning Powder Ingestion Rate
B.W = Average Body Weight of an Adult

2.3.2 Hazard Assessment (Non-Cancer Effects)

The non-cancer risk for each metal via ingestion of seasoning powder was evaluated by computing the HQ and HI using equations (2) and (3)

$HQ = EDI / RfDo$ …… (2)

Where:
HQ = Hazard Quotient.
RfDo = Oral Reference Dose

HI is the potential Human Health Risk through exposure to more than one heavy metal. It is the summation of HQ of each heavy metal in this study.

Hazard index (HI) = $HQ_1 + HQ_2 + HQ_3 +$ …...HQn …….. (3)

Where:
n= 1, 2, 3, 4 ……..n for individual metal.

2.5 Cancer Risk Index

Cancer risk (CR) index, due to exposure to heavy metal via ingestion of seasoning powder, was calculated using equation (4).

CR= EDIsp X CSF …… (4)

Where:
CSF is ingestion cancer slope factor (mg/kg/day).

The Incremental Cancer Risk as a result of exposure to multiple carcinogenic heavy metals via ingestion of seasoning powder was calculated as the sum of the CR individual heavy metal using equation (5).

ILCR = $CR_1 + CR_2 + CR_3 + CR_4$+……CRn ……(5)

Where: n= 1, 2, 3, 4……n for individual carcinogenic metal.

Table 2.0 Human Model Toxicological Variables for Assessment

Variables	Unit	Values	Reference
Concentration	mg/kg	Data from the study	Data from the study
Oral Reference dose (RfD)	mg/kg/day	Table 3	USEPA (2011)
Average Intake of seasoning Powder Average life time for cancer	Kg	0.01 (54 x 365)	Ikechukwu *et al.*, (2016); Oladoye*et al*., (2016)
Life time	Years	54	Fasanmadeand Dagogo-Jack (2015).
Exposure duration (ED)	Years	54	Fasanmadeand Dagogo-Jack (2015).
Adult body weight (BW)	Kg	65	(Aigberua*et al*., (2018)
Average Life Time for Cancer Effects (ATn)		LT x EF (54x365)	USEPA (2011)
Exposure Frequency (EF)	Day/year	365 by	USEPA, 2011
Cancer Slope Factor (SF)		Table 3	(USEPA IRS,2011) (CALEPA, 1997) (USDOE, 2011)
Maximum acceptable cancer risk		$1.0x10^{-4}$ to $1.0x10^{-6}$	USEPA (2011)

Table 3.0 Cancer Slope Factor and Reference Dose (RfD) for Heavy Metals

Heavy Metal	Cancer Slope Factor (mg/kg/day)	Heavy Metal	Reference Dose (RfD) (mg/kg/day)
Cd	6.3 (USDOE, 2011)	Fe	0.7 (USEPA IRIS, 2011)
-	-	Cu	0.04 (USEPA IRIS, 2011)
-	-	Mn	0.14 (USEPA IRIS, 2011)
Pb	0.0085 (USDOE, 2011)	Co	0.03 (USEPA IRIS, 2011)
Ni	0.91 (CALEPA, 1997)	V	0.007 (USEPA IRIS, 2011)

2.6 Statistical Analysis

The data were statistically analyzed by SPSS software version 26. Values obtained were expressed as mean ± SD.

3.0 RESULTS

3.1 Concentration of Heavy Metals in the Seasoning Powder

The result obtained for total heavy metal concentration in four (4) different brands of selected seasoning powder is presented in Figure 1. The result showed that Fe recorded highest heavy metal concentration (16.31 ± 2.56 mg/kg) while V recorded the lowest concentration (0.16 ± 0.10 mg/kg). When compare to the FAO/WHO maximum permissible limit (MPL), the values obtained for Pb (6.35 ± 1.07 mg/kg) and Cd (0.53 ± 0.09 mg/kg) were above the set MPL of 5.0 mg/kg and 0.3 mg/kg respectively.

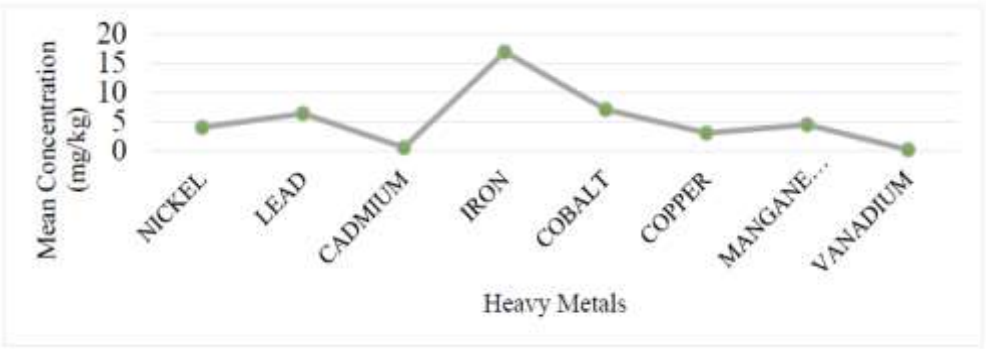

Figure 1: Total Heavy Metal Concentration in Seasoning Powder Sold in Port Harcourt.

The heavy metal concentration in individual seasoning powder is presented in Figure 2. In SP1, SP3, and SP4, mean concentration of Pb (5.94 ± 0.02 mg/kg, 5.72 ± 0.07 mg/kg, 12.61 ± 0.03 mg/kg respectively) and Co (3.86 ± 0.02 mg/kg, 3.67± 0.02 mg/kg, 7.66 ± 0.04 mg/kg respectively) were above the WHO MPL of 5mg/kg and 3.5mg/kg respectively. The mean concentration of Cd in SP1, SP2 and SP4 were 0.35± 0.02 mg/kg, 0.96 ± 0.03 mg/kg and 0.56 ± 0.01 mg/kg respectively which is above the WHO MPL value of 3.0 kg/mg. SP2 and SP4 had mean concentration of Mn as 10.45 ± 0.04 mg/kg and 2.78 ± 0.04 mg/kg respectively. These values are above the WHO MPL. The mean concentration of Ni (8.24 ± 0.01 mg/kg) in SP4 was also above the WHO MPL of 5.0 mg/kg. The concentration of Fe, Cu and V were below the WHO MPL. As and Cr were not detected in all samples.

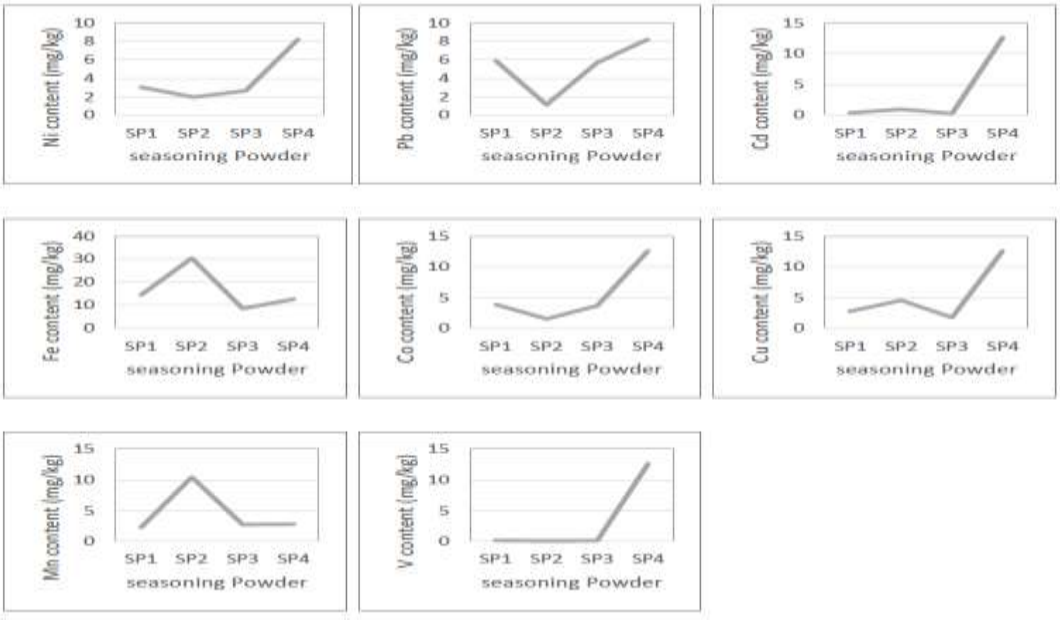

Figure 2: Heavy Metal Concentration in Different Seasoning Powder Sold in Port Harcourt.

3.2 Health Risk Assessment

Results obtained for the EDI of heavy metal in the seasoning powder for adult population, compared to the Maximum Tolerable Daily Intake (MTDI) is contained in Table 4.0. The values are relatively lower than the MTDI for the heavy metals under study. However, in SP1, SP3 and SP4, the EDI values obtained for Pb were above the tolerable limit of 0.5 µg/kg/day set by FAO/WHO.

Table 4: Estimated Daily Intake (EDI) of heavy metals (µg/kg/day) for Adults in Different Brand of Seasoning Powder Sold in Port Harcourt.

Heavy Metal	SP1	SP2	SP3	SP4	Maximum Tolerable Daily Intake (MTDI)
Ni	0.469	0.305	0.407	1.260	2.8 (EFSA, 2015)
Pb	0.912	0.171	0.881	1.940	0.5 (FAO/WHO, 2010)
Cd	0.0152	0.4630	0.0385	0.0801	49.5 (FAO/WHO, 2010)
Fe	2.21	4.70	1.32	2.20	4000 (IOM, 2003)
Co	0.595	0.242	0.565	1.090	23 (EFSA, 2003)
Cu	0.419	0.704	0.271	0.470	500 (SCF, 2003)
Mn	0.351	1.607	0.342	0.424	3000 (EFSA, 2013)
V	0.0206	0.0119	0.0174	0.0467	1800 (IOM, 2001; EFSA, 2004)
As	ND	ND	ND	ND	ND
Cr	ND	ND	ND	ND	ND

Results obtained for the non-cancer risk (Hazard Quotient) are contained in Table 5.0. All calculated values for Hazard Quotient (HQ) were below the threshold value of 1. The Hazard Index (HI), which is summation of individual heavy metals in the sample, also showed values <1 except for SP2 which had values >1 which is not within the safe limits.

Cancer Risk (CR) and Incremental Lifetime Cancer Risk (ILCR) for intake the heavy metals investigated are shown in Table 6. The CR ranged from 2.78 $x10^{-4}$ to 11.5 $x10^{-4}$, 1.45 $x10^{-6}$ to 16.5 $x10^{-6}$ and 2.43$x10^{-4}$ to 95.8$x10^{-4}$ for Ni, Pb and Cd respectively. Values obtained exceeded the acceptable limit of 1.0 $x10^{-6}$ to 1.0 x 10^{-4}. The value obtained for ILCR indicted that SP1, SP2, SP3 and SP4 was 5.31 times, 31.99 times, 6.01 and 16.71 times higher than the set limit. The ILCR for the carcinogenic heavy metals follows the order, SP2 > SP4 > SP3 > SP1.

Table 5: Hazard Quotient (HQ) of Heavy Metals (µg/kg/day) in Different Brand of Seasoning Powder Sold in Port Harcourt.

HEAVY METALS	SP1	SP2	SP3	SP4
Ni	0.023	0.015	0.021	0.063
Pb	0.261	0.050	0.251	0.554
Cd	0.0304	0.926	0.077	0.160
Fe	0.003	0.007	0.002	0.003
Co	0.014	0.006	0.014	0.027
Cu	0.011	0.018	0.007	0.012
Mn	0.003	0.012	0.002	0.003
V	0.003	0.002	0.003	0.007
As	ND	ND	ND	ND
Cr	ND	ND	ND	ND
Hazard Index (HI)	0.35	1.04	0.26	0.83

Table 6: Carcinogenic Risk (CR) of Heavy Metals in Different Brand of Seasoning Powder Sold in Port Harcourt.

HEAVY METALS	SP1	SP2	SP3	SP4
Ni	$4.27x10^{-4}$	$2.78x10^{-4}$	$3.70x10^{-4}$	$11.5x10^{-4}$
Pb	$7.75x10^{-6}$	$1.45x10^{-6}$	$7.5x10^{-6}$	$16.5x10^{-6}$
Cd	$95.8x10^{-4}$	$29.2x10^{-4}$	$2.43x10^{-4}$	$5.05x10^{-4}$
Fe	NA	NA	NA	NA
Co	NA	NA	NA	NA
Cu	NA	NA	NA	NA
Mn	NA	NA	NA	NA
V	NA	NA	NA	NA
As	ND	ND	ND	ND
Cr	ND	ND	ND	ND
Excess Cancer Risk (ECR)	$5.31x10^{-4}$	31.99 x 10^{-4}	$6.01x10^{-4}$	$16.7x10^{-4}$

NA= Not Available, ND= Not detected.

3.3 DISCUSSION

Heavy metals play critical role in health and diseases of plants, animals and human. Everyday exposure to these metals becomes important in addressing potential human health risk (Fraken *et al*., 2020). Seasoning powder most often contain small amounts of heavy metals which contribute to dietary intakes. These products are frequently added to food to improve the taste, aroma, color and overall acceptability (Asomugha *et al*., 2016; Mubeen *et al*., 2009).

The mean concentration of heavy metals in the seasoning powder samples used for this study were in the order; Fe > Co > Pb > Mn > Ni > Cu > Cd > V (Figure 1). Concentration of As and Cr were below detection limit of the instrument in all the seasoning powders investigated. The concentration of Fe, Cu, Co, Mn and V

recorded were below the WHO maximum permissible limit (WHO, 2010). While the concentration of Pb, Ni and Cd were above the WHO maximum permissible limit (MPL) hence may pose significant health challenge following prolonged consumption (WHO, 2010).

There were variations in the concentration of heavy metals in the individual seasoning powders. The mean concentration of Pb and Co was above the WHO MPL of 5mg/kg and 3.5mg/kg respectively, in SP1, SP3, and SP4. SP1, Also, SP2 and SP4 had mean concentration of Cd above the WHO MPL of 3.0 kg/mg. SP2 and SP4 had mean concentration of Mn above the WHO MPL and Ni in SP4 was also above WHO MPL of 5.0mg/kg. The results for Cd, Ni and Pb are in contrast with Makanjuola and Osinfade, (2016) who reported that the concentration of Pb and Cd were lower than the WHO/FAO MPL and Ni was not detected in the five widely consumed seasonings sold in Ogun State, South West, Nigeria.

Other reports in literature have highlighted higher levels of Pb, Ni and Cd in different spices exceeding the maximum allowable levels set by the WHO. Asomugha *et al.*,2016), reported that the range of heavy metal like Pb, Ni were high than WHO/FAO permissible limit. In a study conducted to determine Pb, Cd, Zn, and Cu contents in the most popular spices and herbs used in Polish cuisine, results indicated high amounts of Pb and Cd above standard permissible limits (Krejpcioand Sionkowski, 2007). Ziyaina*et al.,* (2014) also reported increased amounts of Cd and Pb in mixed spices purchased from markets in Libya. The concentration of Cd and Pb were also found to be high in spices from Malaysia (Nordin and Selamat, 2013).

Heavy metals like Pb, Ni and Cd are contaminants of public health concerns. These metals exert no known beneficial effect in biological systems (Udom *et al*., 2022) and are classified as potential carcinogens (Beyersmann and Hartwig, 2008; Olujimi *et al.*, 2015). They have been implicated in a number of neurological, gastrointestinal, reproductive, renal and systemic organ toxicity (Beyersmann and Hartwig, 2008). Pb induces renal tumors and impairs the normal function of the kidneys, joints, reproductive and nervous systems (Udom *et al*., 2022). Increased concentration of nickel (Ni) can cause metal displacement in many enzymes of metabolism. High concentration of Ni is associated with heart attacks, hemorrhages, and kidney impairment (Emejulu*et al*., 2021). Long term exposure to Cd can cause systemic toxicity. It induces oxidative stress by binding to sulfhydryl group, inhibit mitochondria function and elicit epigenetic changes in DNA expression (Bernhoft, 2013).

In biological systems, the bioavailability of essential metals, in trace amounts play important role in several metabolic processes. Fe and Cu form complexes which are essential in the electron transport system for the generation of cellular energy in form of ATP. Iron (Fe) is also involved in protein synthesis. However, at high concentrations the excess Fe can facilitate the formation of free radicals (reactive oxygen species) which can accumulate in body tissues and cause damage (Halliwell,2001).

Impairment in iron metabolism is among the numerous diseases of humans with diverse clinical manifestation ranging from anemia to iron overload (Abbaspour *et al*., 2014). Copper (Cu) function as a cofactor of many metalloenzymes. In combination with Fe, Cu enables the formation of erythrocytes, healthy bones, nerves, and immune function. However, impairment in Cu homeostasis can disrupt many physiological processes (Prashanth *et al*., 2015). Cobalt is a component of vitamin B_{12} (Cobalamin). It regulates the shuttle of homocysteine methyltransferase. Deficiency of cobalt causes cardiomyopathy, polycythemia, and thyroid enlargement (Prashanth *et al*., 2015). Manganese (Mn) plays a role in oxidative phosphorylation, fatty acids and cholesterol metabolism (Prashanth e*t al*, 2015). Vanadium (V) is one of newer trace

elements suggested to be necessary for the formation of bones, teeth, and cartilage. It is also suggested to function in growth and reproduction as well as cholesterol, glucose and insulin production in the body (Ali and Al-Zubaidi, 2012).

Estimated Daily Intake (EDI) and Hazard Quotient (HQ) were developed by USEPA for the evaluation of potential risk associate with long term exposure to hazardous substances (USEPA, 2011; Fuentes-Gandara *et a*l., 2018). Considering the fact that chronic exposure to heavy metals can contribute to high concentration in the biological system, EDI is used an index to evaluate the potential risk of exposure of heavy metals via seasoning powder ingestion. The EDI values were below the Maximum Tolerable Daily Intake (MTDI) and recommended oral Reference Dose. (RfD). The daily consumption of seasoning powder could serve as dietary intake of metals like Fe, Co, Cu, Mn and V to compensate for dietary deficiency or add up to synergistically facilitate body burden and toxicity.

The non- carcinogenic risk of heavy metals in the seasoning powder was calculated using HQ and HI. HQ and HI are used to characterize the health risk of non-cancer adverse effects (Soldatova *et al*., 2018). HI is the potential risk of adverse health effect from a mixture of heavy metal in the seasoning powder. According to the USEPA, if values obtained is less than unity (<1.0), there is no potential adverse health risk and if values obtained is greater than 1.0 there is significanthealth risk (USEPA, 2011). Data obtained from this study showed that all HQ values were < 1.0 which suggest that there is no significant health risk associated with consumption of seasoning powder, except for SP2 with HI >1.0 indicating apparent adverse effects of heavy metals in this sample.

The cancer risk (CR) represents the probability of developing cancer from exposure to heavy metals (Bamuwamye*et al*., 2015). If CR value is < 1.0 x 10^{-6}, risk is acceptable and if value obtained is > 1.0 x 10^{-4}, there is cancer risk (USEPA, 2011). The Cancer risk values obtained in this study exceeded the acceptable range of 1.0 x 10^{-6} to 1.0 x 10^{-4} in the order; SP2 > SP4 > SP3 > SP1. This implies that chronic exposure to heavy metals through consumption of these seasoning powders is associated with cancer risk. Therefore, heavy metal content in seasoning powders consumed in Nigeria should be reduced and continuously monitored and long-term exposure should be discouraged.

4.0 CONCLUSION

Heavy metals were detected in the different brands of seasoning powder investigated and their concentration varied in the order Fe > Co > Pb > Mn > Ni > Cu > Cd > V. The concentration of Pb and Cd were above the maximum permissible limits set by WHO/FAO while Fe, Cu, Co, Mn and V were below the recommended permissible limits. In all the samples, As and Cr were not detected.

Hazard Index values for SP1, SP3 and SP4 were below the maximum limit of 1.0 stipulated by USEPA. However SP2 had Hazard Index of 1.04 indicating an apparent adverse non-cancer effect following chronic use of the seasoning powder. The cancer risk calculated for Pb, Ni and Cd exceeded the acceptable limit of 1.0 x 10^{-6} to 1.0 x 10^{-4} in the order of SP2 > SP4 > SP3 > SP1. Thus, excessive and prolonged use of seasoning powder poses significant cancer risk.

Strict and systematic surveillance of heavy metals such as Cd, Pb and Ni in commonly used seasoning powder should be of utmost importance to ensure safety and quality of the seasoning powders sold in Nigerian.

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