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DETERMINATION OF HEAVY METALS AND POLYCYCLIC AROMATIC HYDROCARBONS IN SOME SPICES AND FOOD COLORANTS CONSUMED IN NIGERIA

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Abstract: **This study investigated the presence and concentration levels of heavy metals and polycyclic aromatic hydrocarbons in widely consumed spices and food colorants in Nigeria. For accurate results, most popular samples were collected from a major market in Port Harcourt, Nigeria. Spices are substances from organic or inorganic origins that add flavour to dishes, derived from different parts of plants and can also be synthetically produced. In this study, Nitric acid - Hydrochloric acid Digestion tests and advanced analytic techniques including Atomic Absorption Spectroscopy (AAS) and Gas chromatography- Mass Spectrometry (GC-MS) were carried out to determine the concentrations of different heavy metals in selected spice samples. The concentrations of some metals are Lead(Pb) - 3.4 ppm, 0.32 ppm, 1.22 ppm, 0.46 ppm and 3.01 ppm in samples A, B, C, D and E respectively. Copper (Cu) concentrations in these five samples were 8.12 ppm, 3.23 ppm, 5.43 ppm, 1.35 ppm and 8.00 ppm. Iron (Fe) concentrations are 55.21 ppm, 31.32 ppm, 38.11 ppm, 22.21zinc and chromium were also found in the samples in high concentrations when compared with the World Health Organization standards. Poly aromatic hydrocarbons including naphthalene, acenaphthylene, fluorine, chrysene were found to be in very high concentration of 82.41 ppm, 74.78 ppm, 140.28 ppm, 58.78 ppm and 132.93 ppm in these five samples. This research aims to assess the safety and quality of these common food spices and colorants given the potential health risks associated with these contaminants.**

Keywords: **Spices, Lead, Copper, Iron, Chromium, Naphthalene, AAS, and GC-MS.**

1. INTRODUCTION

Spices are substances from organic or inorganic origins that add flavor to dishes. They can be derived from different parts of plants including roots, rhizomes, stems, leaves, bark, flowers, fruits or seeds. (Abdullahi,2008). Today, spices are used around the world including Nigeria. Despite the flavoring effects of spices, some however are made with rather harmful and toxic chemicals and are sold around the nation with lots of consumers on daily basis (Singh et al,2015). This study gives a report of some spices consumed in Nigeria for the presence of some heavy metals and polycyclic aromatic hydrocarbons. Some spices are called flavour enhancers. A flavour enhancer increases the way food is perceived and desired without changing the actual flavour of the prepared dish. Flavor enhancers do this by affecting your taste buds. Monosodium glutamate (MSG) popularly known as Maggi, is an example of a flavour enhancer. It intensifies the flavour of savory foods (Susan and Anne, 1998). A flavouring, on the other hand, is an ingredient that actually changes the natural flavour of the foods to which it is added. This enhances the experience of tasting the dish. Both spices and flavorings improve or strengthen the natural flavour of foods ((Singh et al, 2015). Apart from adding flavour to foods, some spices also contain medical and health benefits such as lowering of cholesterol levels, removal of scalp itching and peeling caused by candidiasis, relieve arthritis and back pain, healing of colds, sinus infections and sore throats, burn calories, speed up metabolism, cancer-fighting and anti-inflammatory properties, help to fight communicable diseases as well as lower the levels of bad cholesterol and triglycerides in the blood (Singh et al,2015 and Jashankar et al, 2014). According to Susan and Anne,(1998), seasonings, which are also ingredients used in soup making are composed of notable list of phyto-nutrients, essential oils, antioxidants, minerals and vitamins that are essential for good health. The use of spices in food preparation has become an integral part of life over the centuries, in many parts of the world (Otunola *et al*., 2010).Other examples of spices common in Nigeria include cinnamon, curry, spice city, Lasor, Onga, rosemary, knorr, etc (Brown et al, 2003). However, wide variations in concentrations of trace metals have been reported in some spices (Akpanyung, et al, 2005). Heavy metal contamination in the food chain is caused by environmental pollution and lead (Pb) and copper (Cu) have aroused considerable concern (Nnorom et al,2007). In Nigeria spices are manufactured as bouillon cubes and other well-packaged powdery forms with different brand names. They contain salt, monosodium glutamate (MSG), and some spices as indicated by the manufacturers and are used extensively in food preparation in most homes and restaurants (Oladele et al,2015). MSG is the sodium salt of the amino acid glutamic acid and has long been recognized as a flavour enhancer. It is sold as a fine white crystalline substance similar in appearance to salt or sugar. It is considered a primary taste, umami. Pure MSG does not have a pleasant taste until it is combined with a consonant savory smell (Scatter et al,1989). Glutamate is found in most foods especially foods that are high in protein such as meat, fish, and dairy products. Other sources include mushrooms and tomatoes. In Africa, the current quest for easy or fast way to prepare foods has brought with it a progressive loss of important component of the African food culture. The African richly enormous variety of food spices and condiments are today gradually being replaced by the largenumber of bouillon cubes and spices in the market (Stadman,1990). Bouillon cubes are taste enhancers and are added to foods to augment the taste properties of food (Akpanyung,2005). The major active ingredients in bouillon cubes are salt (NaCl) and monosodium glutamate (MSG). Other raw material used include soybeans, locust beans, onion, tomato, hydrogenated palm oil (HPO), caramel, hydrolyzed plant/vegetable protein and natural spices (RMRDC, 2003). Foods spices are known to contain calcium, magnesium, potassium, sodium and cadmium, lead (Muhammad, 2011). This study was carried out to identify the levels of macro minerals and heavy metals content andthe safety assessment of some seasonings and bouillon cubes readily consumed in Nigeria. Fourtoxic metals (Pb, Cu, Cd and Zn) and four essential micro minerals (Mg, Ca Na and K) were determined to assess human ingestion of these metals from the consumption of foods prepared with these seasonings.In Nigeria, spices are commonly used in the production of dishes and drinks such as: pepper soup, jollof rice, yam pottage, all types of soups and stews, local drinks such as Zobo, ginger and other fruit drinks. Though, spices are used in small quantities, they contribute to the nutrient content of the food (Stadman,1990). These spices are manufactured as bouillon cubes and in powdered form which are packaged with different brand names and are used extensively in food preparation (Otunola et al, 2010; Singh et al,2015). They are added to soups, stews, puddings and sometimes as stimulants which are mixed along with other beverages and used as pepper soup ingredients (Mudgil and Barak,2017; Hudson et al,1984). A bouillon cube is a dehydrated stock formed into a small cube or dried granules. It is formed into a small cube of about 15mm wide. It is typically made by dehydrating chicken stock, fish, meat stock, a small portion of fat, salt, seasonings that are dried or dehydrated and shaping them into a small cube. Bouillon cube is also available in a granular form. When placed in water, the cubes dissolve and its flavors are released. (Ekpo and Jimmy, 2005).Spices and Food colorants which are contaminated by heavy metals from the atmosphere, soil and water pose health risks to humans and animals (Kabata-Pendias, 2001). This study investigated the presence of heavy metals and Poly aromatic hydrocarbons and also the health risks. The growing use of these spices as flavouring, colouring and preservative agent gotten/prepared from variety of plant seed, fruit, root and vegetables which could have been contaminated by heavy metals during agricultural cultivation, industrial or human activities and from leaded gasoline (Khairiah, 2009) or as deliberate ingredients by producers interested only in profit making from the business.

2. MATERIALS AND METHODS

Sample Collection

Five different commercially available brands of spices were purchased at random from local shops around the Port Harcourt Metropolis, Nigeria. The samples were registered by NAFDAC at the time of this study with appropriate batch number and expiry date. The samples were hermetically sealed, and purchased after preliminary investigations had indicated that they were the most consumed seasoning in the metropolis. The spices were: Larsor chicken spice, Mr. Chef peppersoup spice, Onga, spicity jollof rice spice and Mr. Chef Jollof rice spice.

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SAMPLE DIGESTION (wet digestion)

Method: APHA 3030F (Nitric acid-Hydrochloric acid digestion)

A total volume of 100 ml hydrochloric acid and nitric acid in the ratio of 3:1 was mixed to form an aqua regia solution. A 1g of the sample was weighed into a conical flask. About 20 ml of the mixed acid was added to the sample in a conical flask. This was digested using a fume cupboard with hot plate until the appearance of white fumes. It was allowed to cool and filtered into a 100 ml volumetric flask and made up to the mark with distilled water. Portions of the solution were used for atomic absorption spectrophotometer analysis. All the metals were analyzed using the above method but at every point of reading a particular metal with Atomic Absorption spectrophotometer, the cathode lamp for that particular metal is used.

POLYCYCLIC AROMATIC HYDROCARBON

Method No: US EPA 8015 (Extraction for sediment samples)

A beaker was washed and rinsed with acetone. About 10 g of well mixed wet sample was weighed into it. About 25 ml of 1 + 1dichloromethane and acetone were added into it. The sample was sonicated for about 20 mins and centrifuged for 10 mins. The solvent was extracted through a filter paper containing 5g of anhydrous sodium sulphate and preconditioned with 1+ 1 DCM and acetone. Then the second 50 ml of 1+1 DCM and acetone was added to the conical flask. The extraction was repeated combining extracts in the Erlenmeyer flask; this was concentrated into 5 ml using a rotary evaporator. For the aromatics and in the same column 10 ml of DCM was added and eluted. This was used for the GC analysis.

3. RESULTS AND DISCUSSION

Table 1: Metals and Polycyclic Aromatic Hydrocarbon Levels in Samples

S/N0.	PARAMETERS	SAMPLE A	SAMPLE B	SAMPLE C	SAMPLE D	SAMPLE E
1	Pb (ppm)	3.42	0.32	1.22	0.46	3.01
2	Cu (ppm)	8.12	3.23	5.43	1.35	8.00
3	Cr (ppm)	0.45	0.12	0.23	0.08	0.22
4	Fe (ppm)	55.21	31.32	38.11	22.21	47.32
5	Zn (ppm)	11.23	8.22	6.45	3.55	7.23
6	PAHs (%)	82.43	57.78	100.27	57.78	129.09

Table 2: Levels of Polycyclic Aromatic Hydrocarbons in Sample A

S/NO.	Target Compounds	R.T.	Qlon	Response	Conc.Units (ppm)	Dev(Min) Qvalue
1	Naphthalene	4.009	128	592	0.00	61
2	Acenaphthylene	5.434	152	156	2.10	1
3	Acenaphthene	5.584	153	242	17.02	1
4	Fluorene	6.105	166	413	12.02	75
5	Phenanthrene	7.102	178	15969	7.01	63
6	Anthracene	7.102	178	15969	11.00	61
7	O-Terphenyl	7.379	230	4121994	0.66	99
8	Fluoranthene	8.272	202	54053	2.03	91
9	Pyrene	8.489	202	72217	3.00	96
10	Benz(a)anthracene	9.887	228	24638	11.07	63
11	Chrysene	9.887	228	24638	0.01	58
12	Benzo(b)fluoranthene	12.046	252	118256	0.30	59
13	Benzo(k)fluoranthene	12.046	252	118256	5.00	59
14	Benzo(a)pyrene	12.785	252	15383	4.11	59
15	Dibenzo(a,h)anthrancene	15.458	276	307	1.00	36
16	Indeno(1,2,3-cd)pyrene	16.140	276	498	3.00	56
17	Benzo(g,h,i)perylene	16.181	276	160	3.10	1
	Total				82.43	

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Table 3: Levels of Polycyclic Aromatic Hydrocarbon in Sample B

S/NO.	Target Compounds	R.T.	QIon	Response	Conc.Units (ppm)	Dev(Min) Qvalue
1	Naphthalene	4.021	128	259	1.00	68
2	Acenaphthylene	5.441	152	96	10.00	1
3	Acenaphthene	5.588	153	156	3.00	1
4	Fluorene	6.135	166	455	0.00	15
5	Phenathrene	7.098	178	6016	0.00	72
6	Anthrancene	7.098	230	2693965	0.00	65
7	O-Terphenyl	7.376	202	17315	0.77	99
8	Fluoranthene	8.279	202	12119	0.01	85
9	Pyrene	8.482	228	220	10.00	59
10	Benz (a) anthrancene	9.816	228	10197	3.00	1
11	Chrysene	9.880	252	149	0.00	65
12	Benzo (b) Fluoranthene	11.852	252	155	10.00	26
13	Benzo (k) Fluoranthene	11.945	252	149	2.00	6
14	Benzo (e) Pyrene	12.680	276	146	1.00	1
15	Dibenzo (a,h) anthracene	15.724	276	310	5.00	1
16	Indeno (1,2,3-cd) Pyrene	16.076	276	310	5.00	1
17	Benzo (ghi) Perylene	16.361	276	290	7.00	61
	Total				57.78	

Table 4: Levels of Polycyclic Aromatic Hydrocarbons in Sample C

S/NO.	Target Compound	R.T.	QIon	Response	Conc. Units(ppm)	Dev(Min) Qvalue
1	Naphthalene	4.01	128	568	0.00	72
2	Acenaphthylene	5.456	152	3203	0.01	73
3	Acenaphthene	5.621	153	374	20.00	1
4	Fluorene	6.109	166	6035	0.03	72
5	Phenanthrene	7.057	178	51628	0.02	73
6	Anthrancene	7.106	178	62704	10.01	69
7	O-Terphenyl	7.368	230	3864508	0.07	98
8	Fluoranthene	8.230	202	91095	0.03	93
9	Pyrene	8.455	202	113198	0.01	90
10	Benz(a) anthrancene	9.850	228	15441	20.01	90
11	Chrysene	9.880	228	8891	10.00	62
12	Benzo (b) Fluoranthene	11.983	252	242023	0.02	75
13	Benzo (k) Fluoranthene	11.983	252	242023	10.00	75
14	Benzo (e) Pyrene	12.736	252	94804	0.04	66
15	Dibenzo (a,h) anthracene	15.728	276	2715	20.01	27
16	Indeno (1,2,2-cd) Pyrene	15.983	276	455	10.00	75
17	Benzo ghi) Perylene	16.384	276	12508	0.01	78
	Total				100.27	

Table 5: Levels of Polycyclic Aromatic Hydrocarbons in Sample D

S/NO.	Target Compounds	R.T.	QIon	Response	Conc.Units (ppm)	Dev.(Mins) Qvalue
1	Naphthalene	4.021	128	259	1.00	68
2	Acenaphthylene	5.441	152	96	10.00	1
3	Acenaphthene	5.588	153	156	3.00	1
4	Fluorene	6.135	166	455	0.00	15
5	Phenanthrene	7.098	178	6016	0.00	72
6	Anthracene	7.098	178	6061	0.00	65
7	O-Terphenyl	7.376	230	26939650	0.77	99
8	Fluoranthene	8.279	202	17315	0.01	85

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9	Pyrene	8.482	202	12119	10.00	59
10	Benz (a) anthracene	9.816	228	220	3.00	1
11	Chrysene	9.880	228	10197	0.00	65
12	Benzo (b) fluoranthene	11.852	252	149	10.00	26
13	Benzo (k) fluoranthene	11.942	252	155	2.00	6
14	Benzo (e) Pyrene	12.680	252	149	8.00	1
15	Dibenzo (a, h) anthrancene	15.724	276	146	5.00	1
16	Indeno(1,2,3-cd) pyrene	16.076	276	310	5.00	1
17	Benzo (ghi)perylene	16.361	276	290	1.00	61
	Total				58.78	

Table 6: Levels of Polycyclic Aromatic Hydrocarbons in Sample E

S/NO.	Target Compound	R.T.	QIon	Response	Conc.Units (ppm)	Dev(Min) Qvalue
1	Naphthalene	4.009	128	216	30.30	1
2	Acenaphthylene	5.449	152	242	0.30	1
3	Acenaphthene	5.595	153	222	30.00	1
4	Fluorene	6.161	166	1237	3.03	32
5	Phenanthrene	7.113	178	7627	0.40	70
6	Anthrancene	7.113	178	7627	2.10	70
7	O-Terphenyl	7.379	230	3013623	1.10	99
8	Fluoranthene	8.290	202	21445	0.02	86
9	Pyrene	8.515	202	23081	0.01	62
10	Benz (a) anthrancene	9.973	228	166681	10.05	62
11	Chrysene	9.973	228	166681	11.30	55
12	Benzo (b) fluoranthene	12.163	252	2252	5.20	90
13	Benzo (k) fluoranthene	11.805	252	326	10.10	11
14	Benzo (e) pyrene	12.609	252	214	0.00	1
15	Dibenzo (a,h) anthrancene	15.506	276	200	0.02	1
16	Indeno (1,2,3-cd) pyrene	16.140	276	318	25.08	41
17	Benzo (ghi) perylene	16.140	276	318	0.08	27
	Total				129.09	

Figure 1: Quantitation Report Abundance-Time graph.

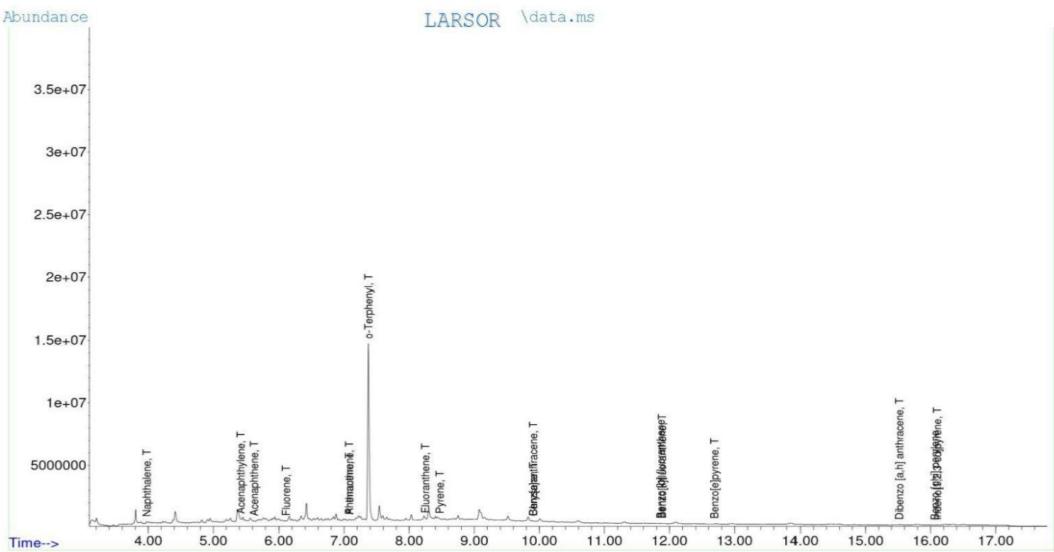

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Figure 2: Quantitation Report Abundance-Time graph.

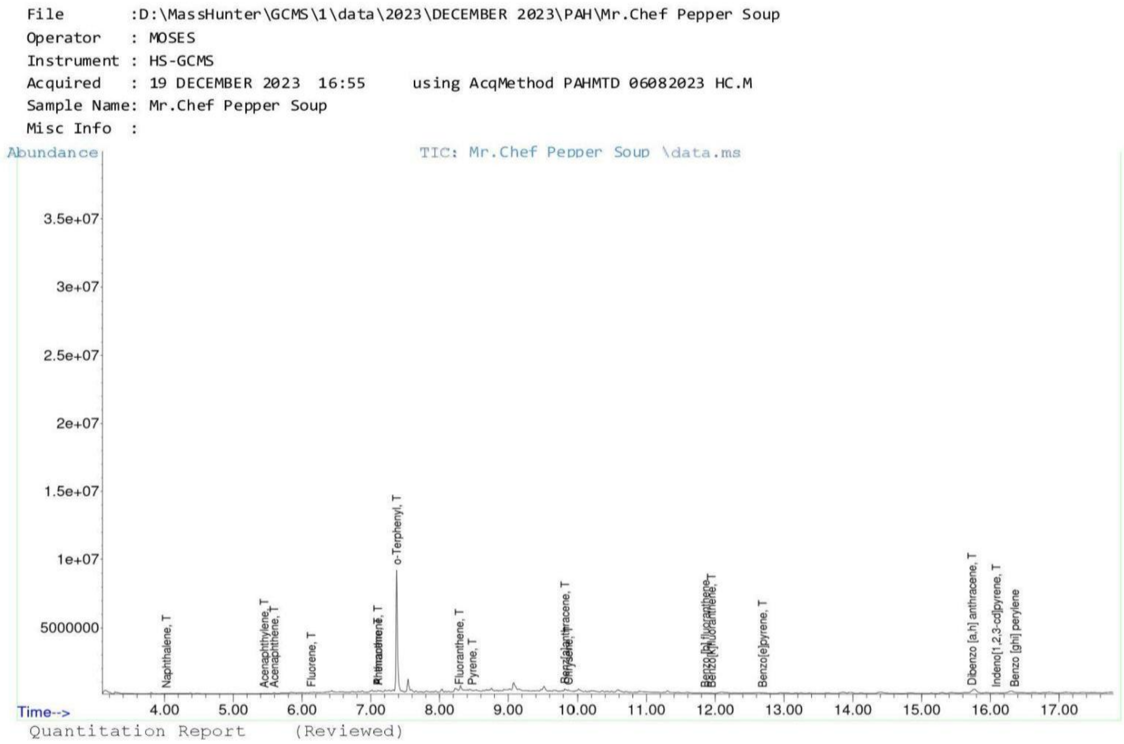

Figure 3: Quantitation Report Abundance-Time graph.

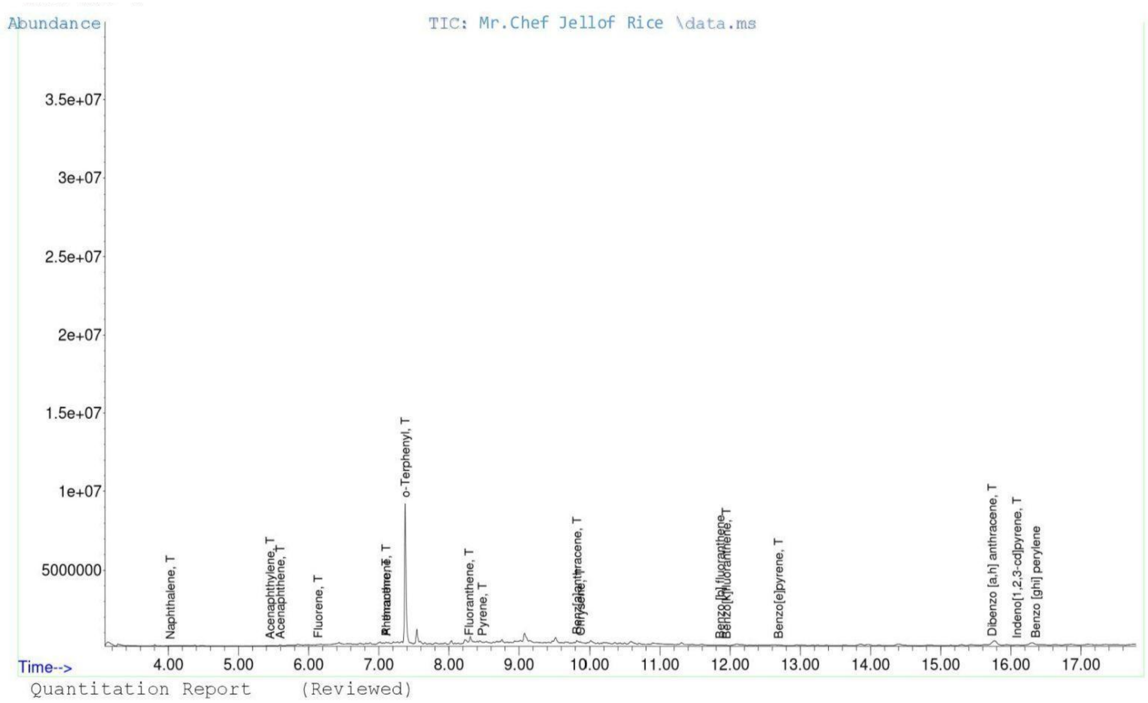

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Figure 4: Quantitation Report Abundance-Time graph.

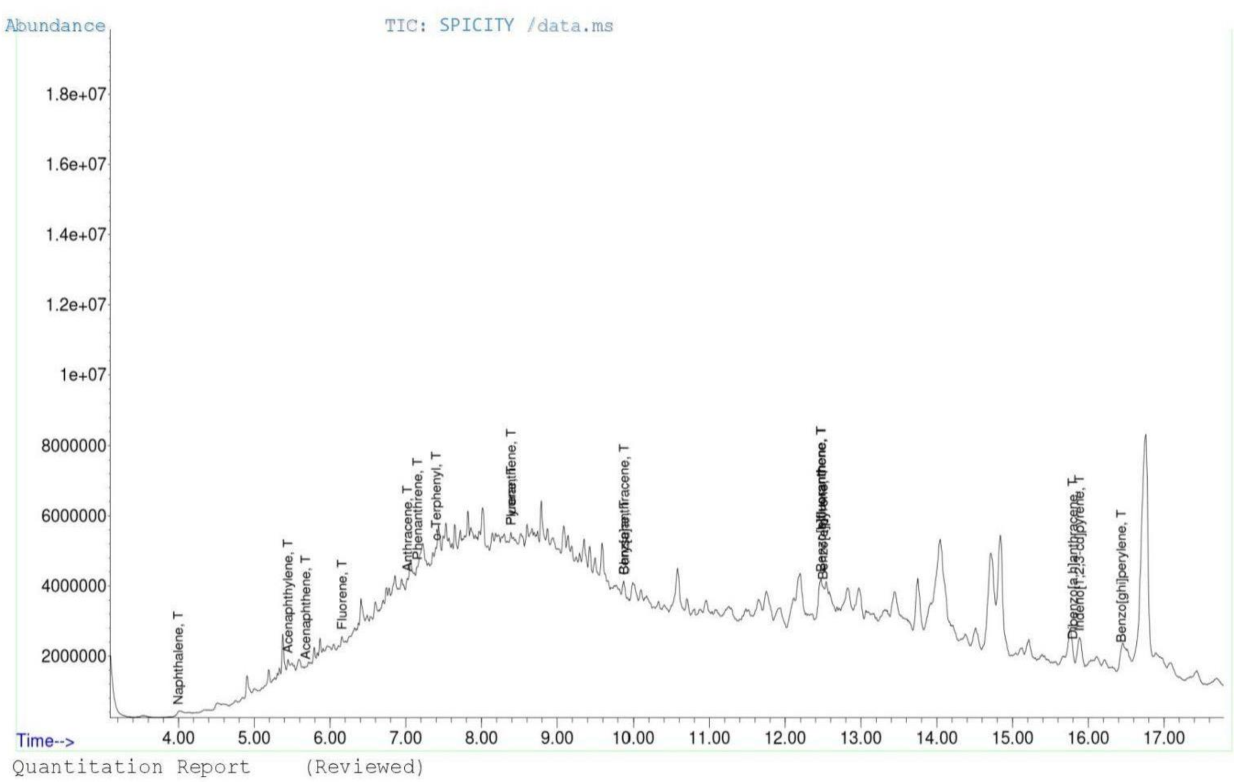
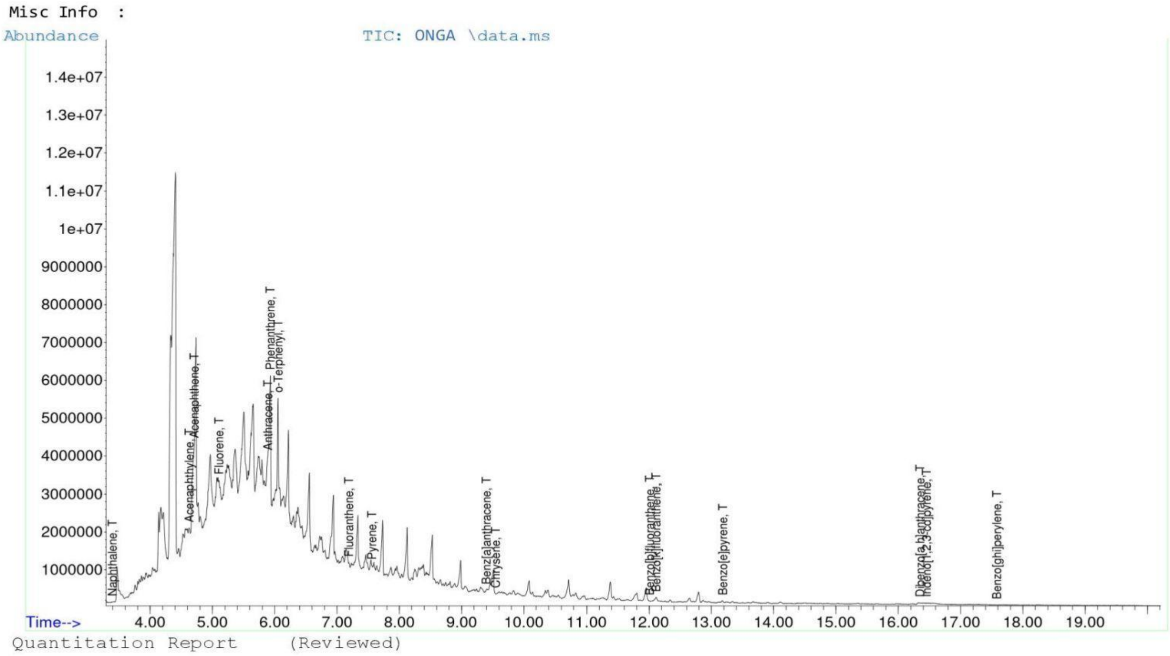


Figure 5: Quantitation report Abundance-Time graph.



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4. DISCUSSION

Lead: This heavy metal was detected in all five spice samples as can be seen in table 1. Lead is a toxic metal of public health concern with no known biological function and reported to induce toxicity at concentrations (Jaishankar, 2014). The lead content in spices and food colorings could be attributed to the addition of lead during processing to impart colour, sweet taste or increase the weight of these products (Kakosy et al, 1996). Some studies reported that traffic density also increases the lead burden in the environment thereby increasing the lead content in vegetation (Nabulo, 2004). Lead pollution has been shown to be commensurate with population/vehicular density (Ansari et al,2004). Generally, lead contaminations occur in vegetables grown on contaminated soils, through air deposition or through sewage sludge/wastewater application (Opaluwa et al, 2012). Lead toxicity is known to inhibit the action of these enzymes because they have free sulfhydryl groups. This is particularly noted with the precursors of haem, and leads to a decrease in haem synthesis, and hence to anemia. Irrespective of the way a lead compound enters the body, it first penetrates the initial cellular barrier before reaching the intracellular fluid. The compound then penetrates the capillary blood vessels and thus enters the circulatory system which distributes it throughout the body (Petersdorf, 1999). Lead poisoning is a global reality, and fortunately is not a very common clinical diagnosis yet in Nigeria except for a few occupational exposures (Anetor et al,1999). In this study, the concentrations of Pb in all five samples are generally higher than the permissible levels by FAO/WHO of 0.001ppm and hence pose health risks to consumers.

Copper: This is an essential trace mineral in the human body, functioning in enzymatic reactions, connective tissue formation, nervous system health, immune function, antioxidant defense, etc. However at excessive concentration levels, copper tends to become toxic with neurological symptoms including Alzheimer's disease. It also interferes with absorption and metabolism of other minerals and causes Wilson's disease. In this study, the concentration levels of copper in all selected spice samples are above the FAO/WHO permissible standard of 0.02 ppm, hence posing health risks to consumers.

Chromium: It helps in glucose and lipid metabolism in the body and also in weight management but becomes toxic in high concentrations leading to environmental pollution, kidney and liver damage due to prolonged exposure. In this study, chromiun concentration levels are slightly higher than FAO/WHO standard of 0.005ppm. Sample D is the closest to this standard value and thus poses the least health risk to consumers.

Iron: It is a key component in haemoglobin, a protein in red blood cells responsible for transporting oxygen from the lungs to the rest of the body. It also supports energy production as it involves electron transport. Iron is important for immune functions and health and plays cognitive and muscle functions. Iron deficiency can lead to anemia characterized by fatigue, weakness, impaired cognitive function and decreased immunity. Whereas excessive iron accumulation can damage organs particularly the liver, heart, pancreas, and increase the risk of diabetes, liver cancer and heart disease. In our current study, the levels of Fe in samples B and D are relatively close to standard requirements of 24ppm while the others exceed by large margins and are thus harmful to health.

Zinc: This metal plays a crucial role in supporting the immune system, promoting wound healing and aiding in cell growth and division. It also helps in taste and smell sensations. However it is important to note that excessive zinc intake can have risks such as the interference of mineral absorption and effectiveness of drugs, neurological crisis, diarrhea, severe headaches, vomiting, loss of appetite, etc. In our study, only sampleD meets the WHO standard level of 3 ppm. All four other samples therefore pose health risks as they are higher than this standard.

Polycyclic Aromatic Hydrocarbons in Spices and Food Colorants:

In this work, seventeen different polycyclic aromatic hydrocarbon target compounds were tested in all five(5) samples. In Nigeria the Standard Organization (SON), Federal Ministry of Environment, National Agency For Food and Drug Administration and Control (NAFDAC) and the National Environmental Standards and Regulations Enforcement Agency (NESREA) are bodies which play roles in regulating, monitoring and enforcing standards related to PAHs content in various contexts within Nigeria to ensure that their levels do not pose a significant risk to public health safety.

Naphthalene concentration in samples A,B,C, and D is below 2 ppm and is considered safe by WHO and OSHA(Occupational Safety and Health Administration) but in sample E it is extremely high and pose health risks to consumers.

Acenaphthylene grossly exceeded maximum 8 ppm concentration in samples B and D.

Pyrene, fluorene and other PAHs in the samples all exceeded recommended levels and thus pose serious health risks to consumers.

5. CONCLUSION

This study on the determination of heavy metals and polycyclic aromatic hydrocarbons(PAHs) in commonly used spices and food colorants in Nigeria has provided significant insights into the current state of food safety and potential health risks associated with these products. This extensive analysis revealed varying levels of heavy metals and PAHs, some of which exceeded the recommended safety limits set by the regulatory bodies. The presence of heavy metals such as lead, cadmium, copper, zinc, etc in some spices and food colorants highlights a concerning issue, given their known health risks, including neurological damage, renal failure and increased risk of cancer. Similarly, the detection of PAHs, particularly those known for their carcinogenic properties, necessitates immediate attention due to their potential to accumulate in the human body over time, leading to long-term health consequences. This study advocates for increased public awareness regarding the potential health risks of contaminated spices and colorants. Educating consumers about safe handling practices and the importance of choosing products from reputable sources can play a significant role in minimizing exposure to these harmful substances. In the light of this study, further and continuous research is strongly recommended to monitor the levels of these heavy metals and PAHs in food products, complemented by efforts from government agencies and food safety authorities to develop comprehensive risk management strategies as this is of paramount importance.

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