1	<u>Original Research Paper</u>
2 3 4	EVALUATION OF CONCENTRATION OF COPPER, LEAD, AND ZINC IN DIFFERENT BRANDS OF CIGARETTE SOLD IN KEFFI MAIN MARKET OF NASARAWA STATE, NIGERIA
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6	ABSTRACT
7 8 9 10	<b>Background:</b> Tobacco smoking is one source of heavy metals exposure in the general population. Tobacco smoke has toxic, genotoxic, and carcinogenic properties and has been linked to fatal pregnancy outcomes.
11 12 13	<b>Aim:</b> This research was carried out to investigate the concentration of Copper (Cu), lead (Pb) and Zinc (Zn) in different cigarette brands sold in Keffi main market of Nasarawa state and compare with recommended standard.
14 15 16	<b>Study Design:</b> To determine the presence of heavy metals in different brands of cigarette sold in Keffi main Market, Nasarawa State, Nigeria and compare them with relevant authorities' standard.
17 18	Place and Duration: The study was carried out in the department of physics, Nasarawa State University, Keffi, Nasarawa State, Nigeria between March 2018 and November, 2018.
19 20 21 22 23 24	Materials and Methods: Ten brands of cigarette were purchased from Keffi main central market using random sampling. The cigarettes were expose to sun light for 3 hours until they became dry. The filters and papers of the cigarettes were removed after drying and the tobacco content were pulverized using agate mortar and pestle. The processed samples were packaged in a well labelled polyethylene bag. A total of ten samples (one sample per brand) were analyzed using XRF spectrometry.
25 26 27 28 29	<b>Results:</b> The range of Cu, Pd, and Zn concentration in ten brands of cigarette were (14.30 to 64.30 mg/kg), (5.43 to 10.55mg/kg), and (12.05 to 160.45 mg/kg) respectively. The brand with the highest concentration of Cu, Pb, and Zn are St. moriz (160.45 mg/kg), London (10.55 mg/kg) and Benson (64.30 mg/kg) respectively, while the lowest concentration found in Bohem (12.05 mg/kg), Excel (5.43 mg/kg), and Yes (14.30 mg/kg) respectively.
30 31 32 33	Conclusion: The concentration of Cu, Pd, and Zn are high in most of the cigarette brands. Concentration of Pb are above the recommended limit reported in the United Kingdom (1.35mg/kg) and Korea (0.74 mg/kg). Concentration Cu and Zn are also high in the cigarette brands.
34	Key words: Heavy metals, Cigarette, Tobacco and Smoking.
35	INTRODUCTION
36 37	Several heavy metals are found in tobacco smoke such as Cd, Cr, Pb and Ni also accumulate in tissues and fluids through smoking [1, 2, 3, 4]. Tobacco smoking is one source of Cd exposure in

the general population. According to AI-Bader *et al.* [4], the most common sources of Cd in humans are through smoking and food. Cadmium can enter the body through tobacco smoking, diet, drinking water, and inhaling it from the air. Small amounts of Cd taken over many years may cause kidney damage and fragile bones, since Cd is mainly stored in bone, liver and kidneys [5,6]. Furthermore, Cd causes stomach irritation, vomiting, and diarrhea. Cadmium and lead, present in tobacco smoke, contribute substantially to cancer risk [7]. Cadmium is a group I carcinogen and lead has recently been elevated from a group IIB to a Group IIA carcinogen [8]. Cigarette smoking leads to an estimated 443,000 deaths per year in the United States [9]. Smoking-related diseases are ultimately the result of nicotine addiction [10], which leads to the repeated inhalation of a variety of toxicants in cigarette smoke, including nitrosamines, polycyclic aromatic hydrocarbons, volatile organic compounds, and several toxic heavy metals [11]. Among these toxicants, there have been comparatively fewer studies conducted on the role of heavy metals as causes of smoking-related diseases and there is a need for basic studies on the levels of heavy metals in cigarettes and other tobacco products.

The Cu content in tobacco leaves was reported to be 156 mg/g [34]. Serum concentrations in youngsters are about 0.95 mg/l [29, 31], in adults 1.10 mg/l [29, 30], and 1.31 mg/l in smokers (more than 10 cigarettes/day) [31], although the differences did not reach significance. However, Lapenna et al. described a significant correlation between plasma levels of Cu and lipid peroxidation in smokers [32]. Since free Cu2b is a potent catalyst in the so-called Fenton reaction in which radicals are produced, these observations might indicate that Cu is increased and 'functional' in smokers.

The Pb content of a cigarette is around 1.2 mg, and about 6% passes over to mainstream smoke, which is inhaled by smokers [31]. Mortada et al. [27] report blood Pb levels (mean+SD) in non-smokers of 101.6 + 30.9 mg/l and in smokers of 143.7 + 33.8, whereas Satarug et al. [28] report Pb serum levels of 4.2 + 5.4 mg/l in non-smokers and 9.0 +12 in smokers. The dramatic difference between blood and serum Pb concentrations is due to the fact that Pb in the circulation is mainly bound to erythrocytes [34]. Pb is eliminated from the body via urine, but this occurs slowly and accumulation in the skeleton is observed. Although the blood brain barrier is relatively impermeable for Pb, especially children are at a high risk to accumulate the neurotoxic Pb in the brain and central nervous system, resulting in mental retardation and other neurological disorders [33]. In addition, children from parents who smoke accumulate high levels of blood lead via passive smoking. These facts are just one further example that stresses the urgent need to protect children form active as well as passive smoking.

Although zinc is present in cigarettes (average 24 mg/g) and about 70% is transferred to the smoke [31], Zn serum concentrations in the average population have not been found to be affected by the smoking status (e.g., a study by Galan et al. [33] with 3128 participants; 13.3% smokers). However, there is evidence that under special circumstances, like pregnancy, mononuclear cells and maybe also other cell types have a reduced Zn content [31]. Reduced Zn levels have been shown to constitute a pro-carcinogenic factor [34] as well as affecting immune system function [34]. The mechanism by which cigarette smoking reduces Zn levels is thought to result from an increased blood Cd concentration, which induces the expression of metallothioneins, which bind Cd but also Zn. Although zinc is present in cigarettes (average 24 mg/g) and about 70% is transferred to the smoke [31], Zn serum concentrations in the average population have not been found to be affected by the smoking status (e.g., a study by Galan et al. [33] with 3128 participants; 13.3% smokers). However, there is evidence that under special circumstances, like pregnancy, mononuclear cells and maybe also other cell types have a reduced

- Zn content [31]. Reduced Zn levels have been shown to constitute a pro-carcinogenic factor [45]
- as well as affecting immune system function [34]. The mechanism by which cigarette smoking
- reduces Zn levels is thought to result from an increased blood Cd concentration, which induces
- the expression of metallothioneins, which bind Cd but also Zn [34].
- The level of exposure to metals in the smoke drawn from a single cigarette is small and likely not
- 89 acutely toxic, but the accumulation of metals in the body over months, years, and decades of
- exposure is, depending on clearance rates, a health concern [12, 13]. Several heavy metals found
- 91 in tobacco smoke, such as Cd, Cr, Pb, and Ni, accumulate in tissues and fluids after smoking [14,
- 92 15, 16]. This is a particular issue for cadmium (Cd) and lead (Pb), which have long (10–12 year)
- 93 half-lives in the human body. Cigarette smoking is a major exposure route for Cd (and to a lesser
- extent Pb) in the general population [16, 17]. This study was carried out to ascertain the levels of
- 95 these heavy metals in different brand of cigarette that are sold in Keffi main Market, Nasarawa
- 96 State, Nigeria.

## MATERIAL AND METHOD

- Ten brands of cigarette were purchased using random sampling from Keffi central market in
- 99 Keffi, Nasarawa state, Nigeria. Keffi lies in Latitude 8°50'47.44"N and
- Longitude 7°52'24.74"E. It has an area of 138 km² and a population of 92,664 at the 2006
- census. The postal code of the area is 961. The major occupation of the people is subsistence
- farming and artisanal mining.
- The brands of cigarettes were expose to sun light for 3 hours until they became dry. The filters
- and papers of the cigarettes were removed after drying and the tobacco content were pulverized
- using agate mortar and pestle. Thus, care was taken so that the samples were not directly
- influenced by dust during drying. The pulverized tobacco content was passed through 2.00 mm
- sieve. The mortar and pestle were thoroughly clean using tissue paper and methylated spirit
- before the next sample is applied for pulverization to avoid mixture in the sample preparation.
- The processed samples were stored in polyethylene bags and labeled properly and was taken to
- the Center for Energy Research and Development (CERD) at Obafemi Awolowo University Ile-
- 111 Ife Osun State, Nigeria. At CERD, elemental analysis was carried out.
- The elemental analysis of all the eleven plant parts were carried out using the Energy Dispersive
- X-ray Fluorescence (EDXRF) spectrometry at the Centre for Energy Research and Development
- 114 (CERD), Obafemi Awolowo University, Ile-Ife, Nigeria. The spectrometer brand name is
- ECLIPSE III supplied by AMTEK INC. MA; USA. It is a self-contained miniature X ray tube
- system. The detection system for all the measurements is a Model XR-100CR which is a high
- performance X-ray Detector with preamplifier and a cooler system which uses a
- thermoelectrically cooled Si-PIN photodiode as an X-ray detector. The power to the XR-100CR
- is provided by a PX2CR power supply. The detector is coupled to the pocket MCA 8000A
- Multichannel Analyzer. The resolution of the detector for the 5.9 keV peak of 55Fe is 220 eV
- FWHM with 12µs shaping time constant for the standard setting and 186 eV FWHM with 20µs
- time constant for the optional setting.
- The samples to be irradiated are placed in the sample chamber. The sample chamber has
- connections to it, which are at angle 45° to it respectively, the source X-ray tube and the Si- PIN

- photodiode detector. The source X-ray tube is maintained at a voltage of 25kV and a current of 50µA and each of the samples is irradiated for 1000sec.
- Quality control measure was taken to ensure reliability of results. Samples were handled
- carefully to avoid contamination. Recovery test was carried out on the XRF machine by spiking
- 129 analyzes.

# RESULTS AND DISCUSSION

- The results of heavy metal concentrations in tobacco products sold in Keffi main Market of
- Nasarawa State, Nigeria, are given in Tables 1. The range of heavy metals concentration in 10
- brands of cigarette (Benson, Bohem, Excel, London, Pallmall, Rothmas, Royal S, St moriz, Yes,
- and Tiger) were Cu (14.30 to 64.30 mg/kg), Pb (5.43 to 10.55mg/kg), and Zn (12.05 to 160.45
- 135 mg/kg).

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Table 1: Concentrations of heavy metals in cigarette samples (mg/kg)

			138
Samples	Cu	Pb	Zn 138
	(mg/kg)	(mg/kg)	(mg/ <u>llgg</u> )
Benson	64.30	7.01	150.01
Bohem	44.04	8.00	12.0 <sup>540</sup>
Excel	30.54	5.43	78.45 <sub>41</sub>
London	44.66	10.55	111.23
Pall mall	26.34	8.40	77.5842
Rothmas	54.12	8.79	134.823
Royal S.	23.00	7.35	64.98
St.moriz	62.60	8.02	160.4454
Yes	14.30	6.33	47.3045
Tiger	32.11	7.03	82.40
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Copper was detected in all the cigarette brands with 'Benson' having the highest concentration of 64.30 mg/kg. The order of decreasing concentration Cu in the cigarette brands are 'St. moriz'(62.60 mg/kg), 'Rothmas' (54.12 mg/kg), 'London' (44.66 mg/kg), 'Bohem' (44.04 mg/kg), 'Tiger' (32.11 mg/kg), 'Excel' (30.54 mg/kg), 'Pall mall' (26.34 mg/kg), 'Royal S' (23.00 mg/kg) and 'Yes' (14.30 mg/kg) with the lowest concentration (Fig.1). Copper in high doses can cause anemia, liver and kidney damage, and stomach and intestinal irritation in human [18].

The concentration of lead in all the cigarette brands was high with 'London' having the highest concentration of 10.55mg/kg. Rothmas, Pall mall, St. moriz, and Bohem also have high concentration of 8.79 mg/kg, 8.40 mg/kg, 8.02 mg/kg, and 8.00 mg/kg respectively. The brand with the lowest concentration of Pb is 'Excel' with 5.43 mg/kg of Pb (Fig.1). These results

obtained for Pb are above those reported by Muhammad [19] and Watanabe *et al.* [20] that Pb content in cigarettes sampled from various countries ranged from 0.46 to 3.66 mg/kg. When compared with the Pb contents reported in the United Kingdom (1.35mg/kg) and Korea (0.74 mg/kg), the Pb contents in cigarette brands studied are far higher. According to a Reuters report, a recent tobacco study conducted by researchers from the Buffalo-based Roswell Park Cancer Institute found that cigarettes produced in China contain three times the amount of heavy metals found in Canadian-manufactured brands. It has been documented in the literature that an average of 5.8% of Pb contained in cigarettes is passed to mainstream smoke [22]. Smoking of 20 cigarettes per day has been estimated to result in the inhalation of 1-5µg Pb, or even more [23, 24, 25].

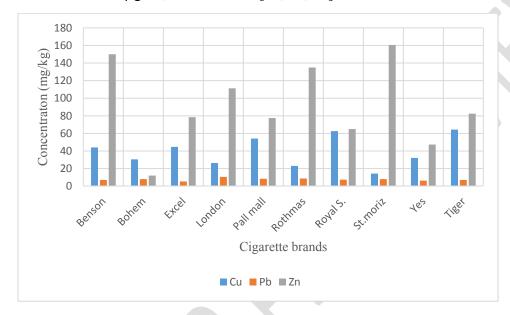


Fig. 1. Mean concentrations of heavy metals in cigarette samples (mg/kg)

The concentration of Zn in the cigarette sample are high with St. moriz having the highest concentration of 160.45 mg/kg. Benson, Rothmas, London, Tiger, Excel, Pall mall, Royal S, Yes and Bohem are having the concentration of 159.01 mg/kg, 134.89 mg/kg, 111.23 mg/kg, 82.40 mg/kg, 78.45 mg/kg, 77.56 mg/kg, 64.98 mg/kg, 47.30 mg/kg, and 12.05 mg/kg respectively (Fig.1). Zinc is a trace element that is essential for human health [26]. Inhalation of Cigarette which has high concentration of Zn can lead to adverse health effect. However, there is no literature that suggests or estimates health effect of Zn in cigarettes.

# **CONCLUSION**

This study confirms that tobacco is a notable source of many heavy metal pollutants particularly Pb. The amount of Pb inhaled from smoking one pack of 20 cigarettes of the brands studied is estimated to be 1.98-3.37 mg/kg and this value is nearly 4 times higher compared with the United Kingdom cigarettes (0.22-0.65 mg/kg) and 3.5 times that of Korean cigarettes (0.4-1.19 mg/kg). The concentration of Cu (14.3-64.3 mg/kg) and Zn (12.05-160.45 mg/kg) in the cigarette brand sold in Keffi are high, but however there is no much research done in this area to study the effect of some of this heavy metals.

- 186 There is no sufficient data about the heavy metal concentrations in cigarette brands in Nigeria
- including Cu, Pb, and Zn. This study will contribute and assist health authorities such as the
- Ministry of Health, Ministry of Environment and other world health authorities such as the
- 189 UNICEF and WHO. Moreover, the results obtained gives very important information for the
- smokers in Nigeria to know that Pb are toxic pollutants that affect adversely on their health
- besides to the other toxic chemicals present in cigarettes such as nicotine. It is recommended that
- the people in Keffi should be sensitized on the effect of heavy metal in cigarettes by the relevant
- 193 authorities.

## **COMPETING INTERESTS DISCLAIMER:**

 Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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