Research Article



Concentrations of Polychlorinated Biphenyl in Mainstream Cigarette Smoke and its Risk Assessments

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Abstract

Cigarette smoking is the most abused tobacco product globally. Hence, level of Polychlorinated biphenyls (PCBs) in the mainstream smoke of cigarettes was determined in this study. Risk assessment of inhaling PCBs in the cigarette smoke was carried out using some health indexes. Mainstream smokes of popular brands of cigarette were extracted using smoke extractor. PCBs quantification was carried out using GC-MS operated in selected ion monitoring mode (SIM). The results showed the dioxin like PCBs ranges of: 1.2-3.37 ng, 1.22-3.61 ng, 0.13-4.21 ng, and 0.29-3.85 ng/ cigarette for PCB 77, PCB 105, PCB 114 and PCB 118, respectively. The Toxicity equivalent (TEQs) of the cigarettes ranged 8.7×10⁽⁻⁶⁾-3.21×10⁽⁻⁴⁾ ng WHO-TEQ. The calculated daily inhalation exposure values ranged, 0.0288-0.0655 ng TEQ kg-1 day-1. The calculated Incremental Life Cancer Risk (ILCR) values are within the limit stipulated by WHO, while the values of Hazard Quotients (HQ) were all above 1. The study revealed high risk of non-carcinogenic illness is associated with inhaling PCBs in the mainstream smoke of cigarette.

Keywords: Cancer risk; Hazard quotient; Mainstream smoke; PCBs; Tobacco

Introduction

The tobacco epidemic is the biggest global public health threat the world is currently facing despite the adoption of the Framework Convention on Tobacco Control (FCTC) in 2003 by several countries [1]. While there has been a decline in the demand for tobacco in developed countries, the growing and consumption rate of tobacco in developing countries is increasingly alarming due to rise in population, consumer purchasing power, and poor tobacco control policies, which has made these countries particularly those in Africa an attractive market for tobacco multinationals [2,3]. According to World Health Organization, Tobacco control plays a major role in the reduction of premature deaths from Noncommunicable diseases (NCDs), which is one of the Sustainable Development Goals (SDGs) targeted by 2030 [4,5]. In a year, the tobacco industry produces six trillion tobacco products consumed by one billion smokers worldwide, resulting in about seven million deaths, 75% of which occur in developing and underdeveloped countries, where more than 80% of smokers in the world lives [6,7]. A large percentage of this number dwells in extreme poverty with a lack of governmental or social support causing heavy health, economic and environmental burdens [8]. Nigeria, the seventh most populous country in the world, is a key tobacco industry market in Africa, with two multinational tobacco companies situated within the country [8-10]. The Global Youth Tobacco Survey of Nigeria indicated that about 4.7 million adults aged 15 years or older comprising 10% men and 1.1% women are active smokers [11-13], the most popular tobacco product smoked in Nigeria are cigarettes and cigars of different flavors and sizes, some of which are manufactured by the two multinational tobacco companies in Nigeria while others are imported into the country and is easily accessible to all at a very cheap rate, sold at retail outlets all over the country [10,14]. Tobacco product manufacturers make use of over 600 substances and ingredients for the making of cigarettes and cigars to enhance the tobacco smoke, many of which are not listed officially on the cigarette packaging in a bid to provide reassurance to health concerned smokers [15,16]. Many chronic diseases are associated with exposure to cigarette smoke, due to the presence of numerous pollutants which endanger the health of smokers [17]. Based on estimation, there are over 8000 chemical constituents in tobacco smoke, Out of which only 400 have been measured in mainstream and sidestream smoke [18]. Of the 400, a significant amount of data exists for about 100. Mainstream smoke is emitted at the end of a cigarette from which a smoker draws air through the burning cigarette to inhale, mainstream smoke consists of 5600 identified compounds [19]. The component of cigarette smoke pollutants includes Carcinogens, tars, Persistent Organic Pollutants (POPs), Volatile Organic Compounds (VOC), etc. Cigars are bigger and contain more tobacco, have fewer additives, and have unfiltered ends. Cigar smoke unknown to many contains many of the same pollutants as cigarette smoke, with a higher level of some carcinogenic substances due to the curing and fermentation process of the tobacco used in the cigar [13]. A smoker is exposed to several chemical compounds with each puff of a lit cigarette or cigar, many of which are toxic to human health [20]. Polychlorinated biphenyls (PCBs), a part of Persistent Organic Pollutants (POPs) are aromatic, synthetic chemicals that do not occur naturally in the environment, they are combustible at high temperature and the product of its combustion are more hazardous than the original material. Most studies carried out on PCBs around the world focused on environmental and human contaminants, ambient air and solid residues, soil samples, fish samples, etc. [21-26]. Over the years, studies on mainstream smoke from tobacco products have focused mostly on its emission, nicotine, tar, Polycyclic Aromatic Hydrocarbon (PAHs), Volatile Organic Compounds (VOC), particulate matter (PM) [27-31]. The determination of PCBs is of special interest for the tobacco industry as only a few studies have been carried out on PCBs in tobacco products. PCBs have been identified in unburned tobacco [32-33] Studies done in the past on PCB in mainstream cigarette smoke date far back as 1998, some studies focused on dioxin and dioxin-like compounds in cigarette mainstream smoke [34].

The increasing rate of tobacco product consumption amongst Nigerians has become a need for concern, as many smokers are unaware of the dangers and risks associated with inhaling pollutants produced during smoking. A number of studies have been done on the dangers of smoking [10,13,35]. Researches in Nigeria on tobacco products over the years have focused only on cigarettes and not cigars [6,9,10-13]. This study focuses on the level of Polychlorinated Biphenyls (PCBs) in the mainstream cigarette smoke of popular cigarette and cigar brands in Nigeria, and the associated risk. This will help with relevant data needed for National Smoking Cessation Strategies and Tobacco control in line with sustainable development goals currently hindered by lack of up to date data in Nigeria.

Materials and Method

Materials

13 top selling cigarettes and 2 cigar brands for this study were selected in Nigeria. Among these brands, 11 were manufactured by two multinational tobacco companies' residents in Nigeria, while the other 2 were imported into the country. The 2 cigars were foreign brands imported into the country. All the Cigarettes and Cigars were purchased from retail outlets in Ado-Ekiti, Ekiti State, Nigeria. Unique identification numbers were assigned to the cigarette and cigar pack, which was entered into a database. The samples were stored in the original packaging at room temperature until required for analysis. The use of Smoking machines to make reproducible samples of cigarette smoke extracts has become a known methodology; however, due to the cost of these machines, high maintenance, and its unavailability in Nigeria, easily assembled inexpensive materials were used in making an apparatus needed for the cigarette smoke extraction process, described in details by Gellner [36]. The set up consists of glass bottles, conical tube cap, two polypropylene tubing, epoxy glue, and glass syringe while Dichloromethane (DCM) was used as solvent. Two sticks were randomly drawn from each pack for the experiment. Upon lighting the cigarette, 50 ml puff is drawn with the aid of a syringe into the solvent, lasting 2-4 seconds, repeated every 30 seconds until cigarette butt is reached (this was done for an average of 35 times) The PCBs are extracted into the DCM, the extract is properly labeled and stored then sent immediately for analysis. The smoking process is repeated for all the cigarettes and cigars brands. The clean-up procedure on the extract was done with the aid of column chromatography consisting of packed alumina and silica. The concentration of the extracts was done using a rotary evaporator, under a gentle stream of Nitrogen. Gas chromatography (Agilent Model 7890 A) coupled with a mass selective detector (Varian 3800/4000 GC-MS) was used to analyze and quantify the PCBs concentration in the samples. HP-5 fused silica capillary column (5% phenyl 95% dimethyl polysiloxane) (30 m length \times 0.25 mm I.D \times 0.25 μ m film thickness) was the column used. The column condition was set at an initial temperature of 120°C with 1 min holding time, increased to 190°C at 20°C min-1, further increased to 230°C at 5°C min-1 and final temperature at 300°C at a rate of 25°C min-1 (10 mins holding time). Injector and transfer line temperatures were set at 280°C and 300°C respectively. The carrier gas used was high purity Helium with a constant flow rate of 0.8 ml/ min-1. 1 µL volume of sample injected into the GC-MS in split less mode was done. Electron impact ionization (EI) by automatic gain control used as a mode of operation storage window was programmed at full scan mode with range m/z 50-500, selected ion monitoring mode was used for data acquisition. The retention time of the authentic PCB standards together with the abundance of the quantification and confirmation ions were used to resolve the identities of the PCBs detected in the samples. PCBs quantification was done using internal standards.

Quality assurance/quality control

Blank was analyzed along the smoke samples to monitor any interference and contamination. Blank is obtained by analyzing uncontaminated DCM poured in a bottle for 1 min alongside other samples.

Health implications

Toxicity equivalence: The PCBs potential toxicity was calculated using the toxicity equivalence factor (TEF). The values were calculated by multiplying the toxicity equivalence factor (TEF) with the dioxin-like (DL) individual level of PCBs. (Eq. i) [37]. Toxic Equivalency (ng WHO-TEQ m⁽⁻³⁾)=C×TEF(i)

Where C is the individual level of PCBs in ng

Inhalation risk assessment: The associated risk with inhalation of PCBs was calculated using inhalation risk analysis (IRA) (Eq 2) [38,39]. Assessment was carried out for adults.

$$IRA = \frac{IRC_o f_r t_f}{BW} (ii)$$

IR is the inhalation rate, 20 m3/day was assumed for adults [38]. Co is the concentration of PCBs in terms of toxicity equivalence (ng TEQ/m3), Co=TEQ/Volume

Volume was calculated to be 0.00175 m3, fr is the alveolar fraction retained in the lungs, 0.75 was assumed, tf is the time of exposure, assumed to be 1. BW is the body weight, 70 kg was assumed for adults based on the average weight value in Nigeria.

Incremental lifetime cancer risk and hazard quotient: Incremental Lifetime Cancer Risk (ILCR) associated with inhalation of PCBs from tobacco product smoking was calculated using Eq 3, while the non-cancinogenic associated risk was assessed using Hazard Quotient (HQ) index (Eq. 4).

$$ILCR = \frac{C \times IR \times ED \times EF \times CF \times IUR}{AT \times BW} (iii)$$
$$HQ = \frac{C \times IR \times ED \times EF}{AT \times BW \times RfD} (iv)$$

C is the Σ individual level of Dioxin-like PCB (ng), ED is the exposure duration, a period of 10 years was used for this study, EF is the exposure frequency in days/years, it is calculated that an addict smokes daily, which translates to 365 days/year, AT is the average exposure time in days, 25550 days was used in the calculation, IUR is the Inhalation Unit Risk 5.7 × 10-3 (ngm3)-1. RfD is the reference dose of PCB (3.3 × 10-5).

Results and Discussion

In this study, a total of 14 PCBs were found in the analysis of the 15 cigarette samples, they are: 2-chlorobiphenyl (PCB 2), 4,4'-Dichlorobiphenyl (PCB 15), 2,2',3-Trichlorobiphenyl (PCB 16), 2,2,5'-Trichlorobiphenyl (PCB 18), 2,4,4'-Trichlorobiphenyl (PCB 28), 2,2',3,5'-Tetrachlorobiphenyl (PCB 44), 2,2',5,5'-Tetrachlorobiphenyl (PCB 52), 3,3',4,4'-Tetrachlorobiphenyl (PCB 77), 2,2',4,5,5'-Pentachlorobiphenyl (PCB 101), 2,3,3',4,4'-Pentachlorobiphenyl (PCB 105), 2,3,4,4',5-Pentachlorobiphenyl (PCB 114), 2,3',4,4'5-Pentachlorobiphenyl (PCB 118), 2,2'3,4',5,6-Hexachlorobiphenyl (PCB 149) and 2,2',3,4,4'5,6,6'-Octachlorobiphenyl (PCB 204). Table 1 shows the PCBs observed in the 15 different cigarette product brands. The cigarette brand with the most predominant PCB is CIG 15, with a 0.13-3.9 ng/m3 range, with a total PCB value of 31.14 ng/m3. CIG 2 has the lowest PCB level of 0-4.27 ng/m3 and \sum PCBs of 22.7 ng/m3. PCB classification is often based on "Dioxin-like" and "Non-dioxin-like". The dioxin-like PCBs cause the activation of the aryl hydrocarbon receptor which includes the multiplication of genes and causes the production of a dioxin-like effect. Four dioxin-like PCB compounds were observed in the analyzed samples, Figure 1 shows the level of Dioxin-like PCBs in all 15 tobacco product brand samples. They are PCB 77, PCB 105, PCB 114 and PCB 118 with a range of 1.2-3.37 ng, 1.22-3.61 ng, 0.13-4.21 ng, and 0.29-3.85 ng respectively. Figure 2 shows the level of $\Sigma PCBs$ in all tobacco product samples, the sample with the highest Σ PCBs is PCB 114 with a percentage of 13% and value 50.9 ng while PCB 149 has the least Σ PCBs percentage of 1% and value of 4.63 ng. The two Cigar samples CIG 3 and CIG 11 analyzed have a SPCB value of 27.28 and 29.92 ng respectively; there is no appreciable PCB level difference between the Cigar samples and that of Cigarette samples. Cig 9, a cigarette brand with a special filter has a significantly high value of 28.67 ng. PCB 149 and PCB 204 were found in 5 samples, which are CIG 6, CIG 9, CIG 11, CIG 14, and CIG 15, including a cigarette brand with a special filter (CIG 9), a menthol-flavored brand (CIG 6), and a cigar brand (CIG 11). Figure 3 shows the percentage distribution of Dioxin-like and Non-dioxin-like PCBs present in all tobacco product brand samples analyzed in the study. 40% of the total PCBs found in the sample are Dioxin-like while the Non-dioxin-like PCB has a percentage distribution of 60%; however, previous studies have shown that Non-dioxin-like compounds have the ability to modulate the overall toxic potency of Dioxin-like compounds. Figure 4 shows the PCB homolog pattern; mono to tetra-chlorinated congeners are the major contributors accounting for 62.5% of the total PCB levels found in the samples, while penta-, hexa and octa-chlorinated congeners account for the remaining 37.5%, no hepta-chlorinated congener was found during analysis. More PCB congeners were discovered in the tobacco product brand samples analyzed in this study compared to that found in Wilson.



Figure 1: Level of Dioxin-like PCBs in all 15 tobacco product brand samples.

Health implication

Toxicity equivalence quotient and inhalation exposure: Table 2 shows the Toxicity equivalency of Dioxin-like PCBs in all 15 tobacco product brand samples. The corresponding toxicity equivalency (WHO-TEQ) values of the tobacco product samples have a range of range of 2.35 \times $10-4 - 5.35 \times 10-4$ ng WHO-TEQ with a mean value of $3.24 \times 10-4$ ng WHO-TEQ. The determination of longterm risk from inhalation of PCBs from tobacco products is done by carrying out inhalation risk analysis. Table 3 shows the IRA for all 15 tobacco product brand samples. The calculated daily inhalation exposure values ranged between 0.0288-0.0655 ng TEQ kg⁻¹ day⁻¹, with CIG 6, a menthol flavored brand having the highest value, and CIG 12 having the lowest amount. The IRA values calculated in this study is several times higher than WHO proposed Tolerance Daily Intake (TDI) of 1000 fg TEQ kg⁻¹ day⁻¹. By implication, the continuous exposure to this pollutant via smoking of tobacco products could cause tremendous hazardous effect on the respiratory systems. Although, there is a dearth of literature in mainstream smoke from tobacco products, the calculated IRA values in this study are higher than calculated IRA values from studies that worked on ambient air and indoor environment.



Figure 2: Level of Σ PCBs in all Tobacco product brand samples.



Figure 3: Level of Dioxin-like (DL) and Non-Dioxin like (Non-DL) PCB present in all tobacco product brand samples.



10-9. The values calculated are lower than the permissible limit of $1.07 \times 10-6$ as stipulated by USEPA. The calculated ILCR is only for dioxin-like PCBs, there are other compounds present in the mainstream tobacco product smoke with carcinogenic properties such as Polyaromatic Hydrocarbons and Volatile Organic Compounds. There will be a significant increase in cancer risk of individuals exposed to the combination of all the carcinogenic pollutants present in the mainstream smoke from smoking of tobacco products. Hazard Quotient (HQ) is used to determine the non-carcinogenic risk associated with the exposure of an individual to a contaminant. The calculated HQ for the 15 different tobacco product brands analyzed in this study has a range of 68.66-114.37 and a mean value of 92.43. These values are significantly higher than the permissible limit of 1, hence a cause for serious concern. The health implication on a tobacco smoke addict is great due to the exposure to this pollutant. Such individual has a high risk of devel-

oping chronic non-cancer organ dysfunction [40-42].

Incremental lifetime cancer risk and hazard quotient: The

Incremental Lifetime Cancer Risk (ILCR) is used to deter-

mine the incremental probability of a person developing

cancer as a result of PCB exposure from mainstream to-

bacco product smoke. The calculated ILCR for this study is shown in Table 3. The values of ILCR were calculated

for adults of legal age (18 years) and above, ranging be-

tween 1.85×10^{-9} - 3.07×10^{-9} with a mean value of $2.48 \times$

Figure 4: Polychlorinated Biphenyls homolog pattern in tobacco product brands samples.

PCB (ng)	CIG 1	CIG 2	CIG 3	CIG 4	CIG 5	CIG 6	CIG 7	CIG 8	CIG 9	CIG 10	CIG 11	CIG 12	CIG 13	CIG 14	CIG 15
PCB 2	0.41	0.45	0.38	0.35	0.47	3.16	0.56	2.98	0.49	0.6	0.8	0.63	0.52	2.38	3.21
PCB 101	0.53	0.45	1.13	1.07	1.16	1.94	0.43	2.27	0.11	1.1	0.15	1.26	1.16	3.1	2.4
PCB 52	1.02	1.26	1.68	1.23	1.36	2.04	2.06	2.24	3.02	3.02	3.02	2.04	2.03	2.03	2.79
PCB 16	1.18	2.97	2.98	3.75	2.97	2.98	2.06	0.96	3.41	3.41	3.46	2	2.05	2.27	2.98
PCB 77	3.37	2.74	2.7	3	3.28	3.01	2.04	1.2	3.07	3.07	2.74	2.18	2.04	1.89	3.9
PCB 118	2.97	1.07	2.46	2	2.53	1.07	0.97	3.85	3.11	3.11	3	2.04	1.07	0.29	2
PCB 18	1.24	2.64	2.8	2.31	2.42	1.2	1.52	2.55	1.52	1.52	2.02	2.35	1.86	1.26	2.2
PCB 15	1.39	4.27	3.72	3.03	4.35	0.97	4.14	3.12	1.6	1.6	2.02	4.1	4.17	2.99	1.14
PCB 28	3.4	1.15	2.18	2.1	2.3	1.06	2.13	2.86	2.83	2.83	3	2.13	2.05	2.3	2.13
PCB 105	2.47	1.22	2.2	2.13	2.51	3.24	1.32	3.8	2.51	2.83	2.97	2.11	2.07	2.47	3.61
PCB 44	1.53	0.43	1	0.23	1	0.23	1.3	2	1.01	2.15	1.05	2.09	2.03	1.03	0.13
PCB 114	3.98	4.05	4.05	4.15	4.15	2.99	4.2	0.13	2.5	4.2	2.46	4.21	4.15	3.28	2.4
PCB 204	0	0	0	0	0	1.62	0	0	2.35	0	2.16	0	0	0.55	1.26
PCB 149	0	0	0	0	0	0.97	0	0	1.14	0	1.07	0	0	0.46	0.99

Table 1: Level of PCBs in the tobacco product brands.

Table 2: Toxicity equivalence for dioxin-like PCBs present in mainstream smoke of all cigarette samples.

TEF	РСВ	CIG01	CIG02	CIG03	CIG04	CIG05	CIG06	CIG07	CIG08	CIG09	CIG10	CIG11	CIG12	CIG13	CIG14	CIG15
0.0001	РСВ	4.10E-	4.50E-	3.80E-	3.50E-	4.70E-	3.16E-	5.60E-	2.98E-	4.90E-	6.00E-	8.00E-	6.30E-	5.20E-	2.38E-	3.21E-
	77	05	05	05	05	05	04	05	04	05	05	05	05	04	04	04
0.00003	PCB	7.41E-	3.66E-	6.60E-	6.39E-	7.53E-	9.72E-	3.96E-	1.14E-	7.53E-	8.49E-	8.90E-	6.33E-	6.21E-	7.41E-	1.08E-
	105	05	05	05	05	05	05	05	04	05	05	05	05	05	05	04
0.00003	PCB	1.19E-	1.22E-	1.22E-	1.25E-	1.25E-	8.97E-	1.26E-	3.90E-	7.50E-	1.26E-	7.38E-	1.26E-	1.25E-	9.84E-	7.20E-
	114	04	04	04	04	04	05	04	06	05	04	05	04	04	05	05
0.00003	PCB	8.91E-	3.21E-	7.38E-	6.00E-	7.59E-	3.21E-	2.91E-	1.16E-	9.33E-	9.33E-	9.00E-	6.12E-	3.21E-	8.70E-	6.00E-
	118	05	05	05	05	05	05	05	04	05	05	05	05	05	06	06
	SUM	3.24E- 04	2.35E- 04	2.99E- 04	2.83E- 04	3.23E- 04	5.35E- 04	2.51E- 04	5.31E- 04	2.93E- 04	3.64E- 04	3.33E- 04	3.14E- 04	2.35E- 04	2.35E- 04	2.99E- 04

Table 3: Incremental Lifetime Cancer Risk (USEPA), Hazard Quotient and Inhalation Risk Assessment values of PCBs in mainstream cigarette smoke.

	CIG 1	CIG 2	CIG 3	CIG 4	CIG 5	CIG 6	CIG 7	CIG 8	CIG 9	CIG 10	CIG 11	CIG 12	CIG 13	CIG 14	CIG 15
IL- CRx 109	2.98	2.11	2.65	2.62	2.9	2.4	1.98	2.09	2.6	3.07	2.6	2.45	2.17	1.84	2.77
HQ	110.74	78.61	98.79	97.66	107.97	89.26	73.85	77.75	96.88	114.37	96.71	91.26	80.78	68.66	103.12
IRA x 102	3.96	2.88	3.66	3.47	3.95	6.55	3.06	6.5	3.6	4.5	4.07	2.88	3.66	3.47	3.95

Conclusion

This study determined the level of Polychlorinated Biphenyls present in the mainstream smoke of popular tobacco product brands in Nigeria and its risk assessment. The results revealed that there are 14 different PCBs found in the analyzed samples. The Σ PCBs for all 15 samples used for analysis are: 17.39, 18.26, 30.84, 39.43, 40.23, 31.54, 29.41, 42.61, 34.45, 37.46, 17.21, 50.9, 7.94 and 4.63 ng. the mean Σ WHO toxicity equivalent quotient of the Dioxin-like PCBs in the tobacco products was 0.00032 ng WHO-TEQ. The ILCR values were lower than the permissible limit, while the HQ values were higher than the permissible limit in multiple folds.

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