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Levels of Persistent Organic Pollutants in Habanero Pepper Vegetable (*Capsicum Chinense*) from Abia and Imo States, Nigeria: The Health Implication

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Abstract

Levels of Dichlorodiphenyltrichloroethane (DDT), polychlorinated biphenyls (PCBs), perfluorooctanesulfonate (PFOS) and polycyclic aromatic hydrocarbons (PAHs) were established soil and vegetable samples obtained from four sites, Aba and Umuahia for Abia State then Mbaise and Amaigbo for Imo State of Nigeria. Multistep strategies for detection of persistent organic pollutants (POPs) were used which included sample preparation, highly selective and sensitive instrumental techniques and quality assurance and control. For the soil samples, ΣDDT ranged from <LOD to 0.011 mg/kg, suggesting contamination below detection limit in Aba and Umuahia and low contamination in Mbaise and Amaigbo soils; PCBs were in the range of <0.001 to 0.021 mg/kg indicating values within tolerable daily intake except in Amaigbo where the precautionary target value was slightly exceeded; ΣPFOS ranged from 0.001 to 0.005 mg/kg, exceeding trigger values in Umuahia, Mbaise and Amaigbo, while ΣPAH ranged from 0.068 to 0.276 mg/kg, suggesting slight pollution in Aba and no pollution in Umuahia, Mbaise and Amaigbo. For the pepper samples, values of ΣDDT ranged from not detected to 0.006 mg/kg which were far below World Health Organization (WHO) Maximum Residue Limit (MRL) value. PCBs recorded in the pepper vegetables were all safe according to WHO 2003 (<0.001 – 0.11 mg/kg). Values for ΣPFOS ranged from <0.001 to 0.010 mg/kg, which were below tolerable weekly intake according to EFSA, 2020, while ΣPAH_4 with range of values from 0.013 to 0.216 mg/kg do not present health risk to consumers based on the margin of exposure results. In conclusion, values of DDT were seen to be below WHO MRL values, the PCBs were safe going by WHO, values of PFOS were below tolerable weekly intake and PAH values in the pepper do not present health risk to consumers. However, observation of good agricultural practices, monitoring, controlling, risk assessments and reviewing of public policies and regulations are highly recommended to minimize POP contamination in food.

INTRODUCTION

Persistent organic pollutants (POPs), sometimes known as "forever chemicals" are organic compounds that are resistant to environmental degradation through chemical, biological, and photolytic processes (Ritter et al., 2007). They exert their negative effects on the environment through two processes, long range transport, which allows them to travel far from their source, and bioaccumulation, which re-concentrates these chemical compounds to potentially dangerous levels (Walker, 2001). These POPs enter the gas phase under certain environmental temperatures and volatize from soils, vegetation, and bodies of water into the atmosphere, resisting breakdown reactions in the air, to travel long distances before being re-deposited (Kelly et al., 2007). Persistent Organic Pollutants are not easily degraded in the environment due to their stability and low decomposition rates. Due to this capacity for long-range transport, POP environmental contamination is extensive, even in areas where POPs have never been used, and will remain in these environments years after restrictions implemented due to their resistance to degradation (Wania & Mackay, 1996).

Bioaccumulation of POPs is typically associated with the compounds high lipid solubility and ability to accumulate in the fatty tissues of living organisms for long periods of time (Wania and Mackay, 1996; Vallack *et al*., 1998). Persistent chemicals tend to have higher concentrations and are eliminated more slowly. Dietary accumulation or bioaccumulation is another hallmark characteristic of POPs. As POPs move up the food chain, they increase in concentration as they are processed and metabolized in certain tissues of organisms. The natural capacity for animals gastrointestinal tract concentrate ingested chemicals, along with poorly metabolized and hydrophobic nature of POPs makes such compounds highly susceptible to bioaccumulation *(*Yu et al., *2011).* Thus POPs not only persist in the environment, but also as they are taken in by animals they bio-accumulate, increasing their concentration and toxicity in the environment (Kelly et al., 2007; Lohmanna et al., 2007). Human exposure for some compounds and scenarios, even to low levels of POPs can lead among others, to increased cancer risk, reproductive disorders, alteration of the immune system, neurobehavioral impairment, endocrine disruption, genotoxicity and increased birth defects (WHO, 2010). Persistent Organic Pollutants as could be seen from above summary are indeed environmental threats that need to be addressed with all seriousness. Their mode of transmission is atmospheric, and they can find their ways in all aspects of the environment (air, water and soil), and what is making it more dangerous is their persistent nature and most of them being non-biodegradable (Donyinah, 2005).

Vegetables can be contaminated while growing in fields due to use of organic fertilizer or contaminated irrigation water or during harvesting, processing, distribution, sale and during consumption (Beuchat, 1995). As a result, they have been involved in food-borne outbreaks (WHO, 2002). Peppers have so many advantages associated with their consumption. They have a lot of good nutrition and are low in calories thereby making them a good dietary component. All the various varieties peppers come in act as an excellent source of vitamin A and vitamin C, folic acid, fiber, and potassium. In addition, they spice up the food and make it more tasty and satisfying. They come in a variety of shapes, sizes, colours as well as tastes. They are very versatile in preparation and can be added to quite a good number of dishes to obtain the nutritious benefits and also to add some taste to the meal (Miller, 2023).

MATERIALS AND METHODS

Sample Collection

A total of eight (8) samples comprising four soils and four habanero pepper vegetables were collected randomly from farms of the selected cities of the two States in the South-East Geopolitical Zone. The samples for each product collected were considered to be quite representative of the soils/vegetables in the city. For the vegetable analysis, only the edible portions were included. The collected samples were labelled and immediately transferred to Abia State Polytechnic Chemistry/Biochemistry Laboratory for analyses.

Determination of Persistent Organic Contaminant Residues

The detection of POPs in the pepper soils and vegetables were carried out using multistep strategies including sample preparation, highly selective and sensitive instrumental techniques, and quality assurance and quality control. Because an extremely low detection limit is required for POP analysis, sample preparation was needed to reduce the matrix effect when carrying out the analyses. Sample preparation used for the detection of POPs in the samples involved multiple steps which included filtration, pH adjustment, extraction, clean-up and enrichment procedures to ensure that the analytes are detected at a suitable concentration level (Loganathan and Masunaga, 2009, Dimpe and Nomngongo 2016, Capriotti et al,. 2013).

Data analysis

Statistical Package for Social Sciences (SPSS) for Windows (Version 20) at 5% level of significance was used, while data were presented using statistical tables.

RESULTS

Results of the levels of POPs on the soil samples

The results of the analysed soil samples collected from the different fields were summarised in table 1 below. Two of the sampled soils from Abia State (Aba and Umuahia) were free from detectable DDT residues (<LOD), while the other two from Imo State (Mbaise and Amaigbo) showed low contamination values (0.011 – 0.016 mg/kg), according to Polish Official Gazette, 2016.

Table 1: Results of the Levels of POPs in the Soil Samples

Locations/ Vegetables	Pesticides	Industrial Chemicals		By-Products of Industrial Processes
	ΣDDT (mg/kg)	PCBs (mg/kg)	ΣPFOS (mg/kg)	Σ_4PAHs (mg/kg)
Aba	<LOD	<0.001	0.001	0.276
Umuahia	<LOD	<0.001	0.011	0.078
Mbaise	0.016	0.012	0.012	0.126
Amaigbo	0.011	0.021	0.005	0.068

Key: ΣDDT=o'p-DDT+p'p-DDT+ p'p-DDE +p'p-DDD; Σ4PAH= BaA+Chr+ BbF+ BaP; ΣPFOS= PFOS+PFOS Substances+PFOS Polymers; ΣPCBs=(sum of indicator-PCBs and dioxin-like PCBs.

Values for total PCBs ranged from <0.001 to 0.021 mg/kg with lowest value recorded in Aba and Umuahia soil samples, while Amaigbo sample recorded the highest value of 0.21 mg/kg. ΣPFOS was observed to record values ranging from 0.001 to 0.012 mg/kg, with its lowest value observed in Aba soil sample and a highest value recorded in Mbaise soil. Values of total PAH (Σ_4PAHs) ranged from 0.0.068 mg/kg in Amaigbo soil to 0.276 mg/kg in Aba soil sample.

Results of the levels of POPs in the habanero pepper samples

The results of the levels of POPs in the habanero pepper analyzed at the various locations in the two States were summarized in table 2 below. Values of ΣDDT ranged from ND (not detected) in Aba, Umuahia and Amaigbo habanero pepper to 0.006 mg/kg in Mbaise pepper. Values ranging from <0.001 mg/kg in Umuahia to 0.011 mg/kg in Amaigbo pepper sample were observed for total PCBs. For ΣPFOS, values from <0.001 mg/kg in Aba pepper sample to 0.010 mg/kg in Mbaise sample. Values for Σ_4PAHs were in the range of 0.013 mg/kg in Umuahia sample to 0.215 mg/kg in Aba pepper sample.

Table 2: Results of the Levels of POPs in the Habanero Pepper Samples

Locations/ Vegetables	**Pesticides**	**Industrial Chemicals**		**By-Products of Industrial Processes**
	ΣDDT (mg/kg)	**PCBs (mg/kg)**	**ΣPFOS (mg/kg)**	**Σ_4PAHs (mg/kg)**
Aba	ND	0.001	<0.001	0.216
Umuahia	ND	<0.001	0.001	0.013
Mbaise	0.006	0.002	0.010	0.110
Amaigbo	ND	0.011	0.002	0.021

Key: ΣDDT=o'p-DDT+p'p-DDT+ p'p-DDE +p'p-DDD; Σ4PAH= BaA+Chr+ BbF+ BaP; ΣPFOS= PFOS+PFOS Substances+PFOS Polymers; ΣPCBs=(sum of indicator-PCBs and dioxin-like PCBs.

DISCUSSIONS

Considering that according to Polish law (Polish Official Gazette 2016) the level of DDT and its metabolites for agricultural soils should not exceed 0.120 mg/kg and that four contamination categories were foreseen, the analyzed samples were classified into the same categories, as follows: contamination below the detection limit (DDT was not detected), low contamination (detected concentration up to 0.025 mg/kg), medium contamination (0.025–0.120 mg/kg) and high contamination (>0.120 mg/kg). All samples were contaminated at low level (below detection limit). The detection of DDT in majority of the monitored soil samples strongly confirms the high persistence of this compound in soil (Lewis et al. 2016). In soil, DDT biodegrades to DDE under unflooded (generally aerobic) conditions and to DDD under flooded (generally anaerobic) conditions (ATSDR, 2002). For total PCBs, the maximum concentration according to the Bulgarian Regulation, 2008 is 200 µg/kg (0.2 mg/kg), the precautionary target value is 20 µg/kg (0.02 mg/kg) and the reference background value set by the ordinance is 5 µg/kg (0.005 mg/kg). Going by the Bulgarian Regulation standard, values of total PCBs for all soils surveyed were below the maximum concentration, while the value for Mbaise pepper soil was above the reference background value set by the ordinance. The total PCB levels in soil samples in this study were lower than those reported in Dutch action value, Australian and New Zealand ecological investigation level of 1000µg/kg (1mg/kg), as well as the Canadian soil guideline for residential areas of 1300 µg/kg (1.3mg/kg) (Canadian Environmental Quality Guidelines, 2003) . The Queensland End of Waste Code Biosolids (2019), published in January 2020, offers the most biosolids-specific guidance, with trigger values for PFOS in soils to be 0.001 mg/kg. With that, trigger values were observed in Aba pepper soil sample, while values higher than the trigger value were observed in other vegetable farm soils. Notification is required if any of the trigger values are exceeded following biosolids land application. It would therefore be prudent to know the PFAS contaminant concentrations in both the biosolids and soil prior to application. PAH-contaminated soil can be divided into four levels: no pollution (< 0.200 mg/kg), slight pollution (0.200–0.600 mg/kg), medium pollution (0.600–1.000 mg/kg) and serious pollution (> 1.000 mg/kg) (Zhengyu et al., 2013). It then means that no pollution was detected in Umuahia, Mbaise and Amaigbo soil samples while a slight pollution was observed in Aba soil sample.

The results of the habanero pepper vegetables surveyed showed that values of ΣDDT for pepper vegetables from Aba, Umuahia and Amaigbo were not detected for those that their soils were below limits of detection, while a value far below the recommended WHO

Maximum Residue Level (MRL) values of between 20 and 300 ng/g (0.02 and 0.3 mg/kg) in agricultural products (WHO, 2013) for most of the OCPs was observed in Mbaise pepper. This could be as a result of the fact that farmers in the surveyed locations did not apply pesticides during the period under study. The reasons that may account for the variations in pesticides found in the crop species are the quality of the soil, planting season, the quantity of pesticides used and the type of pesticide applied (Ahmad et al., 2006; NCEH, 2005). The levels therefore do not pose a health threat to the consumers of the vegetables. However, since DDT and its metabolites are bio-accumulative (Augustina *et al.*, 2008), long term health effects may arise due to DDT bio-accumulation, and over consumption or long-term exposure may lead to health effects especially in pregnant women and children (Nnamuyomba *et al*., 2014; Ssemugabo *et al*., 2022). Values of PCBs in the pepper vegetables ranged from <0.001 mg/kg in Umuahia pepper to 0.011 mg/kg in Amaigbo pepper.

According to WHO, 2003, the daily PCB intake that is safe is 20 ng/kg (0.02 mg/kg) of body weight and by that standard, the habanero peppers in all the locations sampled would not pose any health risk on human when consumed. A rare case was observed in Aba pepper vegetable where the value recorded in the vegetable was higher than that observed in its soil (table 1 and 2), and in Umuahia pepper where the value of the PCB is equal to the value recorded in the soil of the said vegetable. This could be as a result of the urban state and industries surrounding Aba and Umuahia cities since industrial areas are generally considered to be important emission sources of PCB (UNECE, 2011). They may also be unintentionally generated as by-products of activities involving combustion (Mansour, 2009). For ΣPFOS, recorded values ranged from <0.001 mg/kg in Aba pepper to 0.010 mg/kg in Mbaise pepper. The European Food Safety Authority (EFSA) recently (2020) released a scientific evaluation of a tolerable weekly intake (TWI) of 4.4 ng/kg (0.0044 mg/kg)$_{bw}$-week (0.63 ng/kg$_{bw}$-day) for the sum of PFOA, PFNA, PFHxS, and PFOS in food; this follows their previously established TWIs of 6 ng/kg$_{bw}$-week for PFOA (0.8 ng/kg$_{bw}$-day) and 13 ng/kg$_{bw}$-week for PFOS (1.8 ng/kg$_{bw}$-day) in 2018 (EFSA CONTAM 2018, EFSA CONTAM 2020). With the standard above, only Mbaise pepper exceeded the tolerable weekly intake (TWI) of 4.4 ng/kg (0.0044 mg/kg), while those from Aba, Umuahia and Amaigbo were below the limit. The total PAH (Σ_4PAHs) values were observed to range from 0.013 mg/kg in Umuahia pepper to 0.216 mg/kg in Aba pepper.

Considering the toxicity of PAHs, several countries have drafted legislation to establish tolerable limits for PAHs in foods, food products, and beverages as well as to enforce monitoring strategies for the most relevant compounds. Furthermore, health agencies such as the WHO and European Commission have launched efforts to decrease the concentration of PAHs in food, especially through strategies to control the processes that induce their formation [Abdel-Shafy and Mansour, 2016; Commission Regulation (EU), 2011; IARC, 2010; EFSA, 2012]. Until 2008, the EFSA used BaP as the only marker for PAHs occurrence in food. However, a re-evaluation study revealed that 30% of food samples with low levels of BaP had significant levels of other possible carcinogens (Mafra *et al*., 2010; Alexander *et al.*, 2008). Therefore, the European Union amended the previous legislation through Commission Regulation 835/2011, which introduced acceptable levels for the sum of PAH_4 (BaA, Chr, BbF, and BaP) while maintaining a separate maximum level for BaP to ensure comparability with previous and future data (Commission Regulation (EU), 2011). Recently, many studies reporting data on PAHs occurrence and health risk assessment have been published investigating whether a potential risk exists when consuming certain foods. The authors highlighted the necessity of avoiding the cultivation of vegetables, especially leafy ones, in heavily polluted areas.

CONCLUSION AND RECOMMENDATIONS

Considering the soil samples, this study showed that all samples were contaminated at low level (below detection limit). The detection of DDT in majority of the monitored soil samples strongly confirms the high persistence of this compound in soil (Lewis et al. 2016). Going by the Bulgarian Regulation standard, values of total PCBs for all soils surveyed were below the maximum concentration, while the value for Mbaise pepper soil was above the reference background value set by the ordinance. For PFOS, trigger values were observed in Aba pepper soil sample, while values higher than the trigger value were observed in other vegetable farm soils, going by trigger values were observed in Aba pepper soil sample, while values higher than the trigger value were observed in other vegetable farm soils. For total PAH_4, no pollution was detected in Umuahia, Mbaise and Amaigbo soil samples while a slight pollution was observed in Aba soil sample, which was also within acceptable limit.

For the habanero pepper samples, it was observed that total DDT were not detected in three out of the four locations sampled, representing 75%, while a value far below the WHO MRL was observed in one location, representing 25%. Values of PCBs were seen to be at safe levels even with rare cases where the values in the pepper were greater than/equal to the values obtained in the soil samples. Going by (EFSA CONTAM 2018, EFSA CONTAM 2020), only Mbaise pepper exceeded the tolerable weekly intake (TWI) of 4.4 ng/kg (0.0044 mg/kg), while those from Aba, Umuahia and Amaigbo were below the limit. For total PAH_4, there is yet to be an accepted tolerable limit in food generally, though there have been some regulations that have been accepted for specific foods.

This study therefore recommends that farmers should observe good agricultural and manufacturing practices to minimize POP contaminations in food.The presence of POPs in foods must be continually monitored and controlled, including risk assessments related to the consumption of contaminated foods and calculations to determine safe intake parameters. Public policies and regulations should be reviewed and updated to establish safe limits for POPs in foods. Such reviews comprising recent data on levels of POP contamination in foods and risk exposure are essential to aid regulatory agencies in updating or establishing these regulations.

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