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Analysis of PAHs contents and Health risk quantification in *Telfairia occidentalis* (fluted pumpkin) cultivated in the Crude oil Ecozone of the Niger-Delta

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Abstract The results of this study showed the various concentrations of pyrenes, chrysene, benzo[a]pyrene, benzo[a]anthracene, and benzo[b]fluoranthene (0.85±0.11, 2.87±0.12, 2.86±0.10, 2.84±0.11, and 2.78±0.13 µg/kg), respectively in the fluted pumpkin. The non-carcinogenic risks from the possible ingestion of PAH congeners in the fluted pumpkin sourced from different locations in the Niger Delta revealed the progression in values for Beneze (a) pyrene, Benzo (a) anthracene, and Benzo (b) fluoranthene as; 14.55 > 1.46 > 1.42 respectively of the TEFs (Toxicity equivalent factors). However, it was noticed that the mean across the studied sites was considerably higher than the threshold set for this study. The values of the chronic daily intake (CDI) for the PAHs investigated, fluctuated in the sites studied. The incremental cancer risk (ILCR) for this study showed that the concentrations of PAHs were also higher than the set threshold. The study concluded that *T. occidentalis* is contaminated by PAHs above the EU 1255/2020 stipulated level of 2.0 µg/kg for PAHs in vegetables and thus not good for human ingestion It is recommended that the oil companies operating in the Niger Delta adopt world-best practices in their operations, mitigation mechanisms of impacts should be adopted, remediation should be commissioned, and monitoring agencies should be mandated to increase surveillance on the operations of the oil exploration and exploitation companies operating in the Niger Delta.

Keywords: Crude oil production, Polycyclic aromatic hydrocarbons, *T. occidentalis*, Bioaccumulation, Human health

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Introduction

Nigeria, a country in sub-Saharan Africa, is ranked as one of the major crude oil-producing countries producing about 85% of the GDP (Gross Domestic Products) and about 95% of its earnings in foreign exchange (Sanusi 2021; National Bureau of Statistics 2022; Oteriba, 2023; Ruwani 2023). Nigeria recorded 822 oil spill cases between 2020 and 2022 with 28,003 barrels spewed into the terrestrial and aquatic environment (National Oil Spill Detection and Response Agency, 2022; National Environmental Standards Regulations and Enforcement Agency, 2022; Friends of the Earth, 2022) and also flares 10 percent of the associated gas amounting to 7.4 billion cubic feet flared into the air (National Bureau of Statistics, 2022; Friends of the Earth, 2022; National Environmental Standards Regulations and Enforcement Agency, 2022; Oteriba, 2023).

The presence of crude oil in the sub or superficial regions of soil can cause the accumulation of PAHs in plants from the soil, while gas flaring results in particulate droplets of PAHs on the foliar parts of the crops (Osamede, 2018; Osugo, 2018; Enuneku *et al*., 2021; Ogwu *et al.,* 2022; Ogwu *et al*., 2024). Contamination of foods and vegetables by PAHs has been stated to elicit malignancy in humans (Yu *et al*., 2015a, b, c and d; Zafra, 2015; Zhang *et al*., 2015a, b and c; Zamani 2016; Yunker, 2015; Adetunji *et al*., 2021). It causes brain damage, nose, eye, and throat, irritation (Verma, 2015; Vignet, 2015).

Telfairia occidentalis is the major vegetable cultivated and consumed by the people of the crude oil region of the Niger Delta (Okonkwor, 2016; Okolie, 2017). Cultivation of *T. occidentalis* in soil contaminated by PAHs from crude oil and particulate PAH from flaring will result in bioaccumulation and biomagnification (Mao *et al*., 2012; Anani and Olomukoro, 2019; Ogwu *et al*., 2022).

PAHs are a group of naturally occurring chemicals ubiquitous in anthropogenic activities. They are chemically stable, comprising two or more atomic rings of hydrogen and carbon (Tesi *et al*., 2021).

PAHs occur on land, water, air, soil, sediments, and foods (Wu *et al*., 2014a b; Xu *et al*., 2016). Polycyclic hydrocarbons are formed through incomplete combustion and organic pyrolysis (Xu *et al*., 2014; Xu *et al*., 2016; Yang *et al*., 2016). They are also formed in volcanic eruptions, biosynthetic reactions, and forest fires (Yates, 2011; Yebro-Pimemtel 2014). PAHs are also formed in the combustion and incineration of tobacco and wood (Tesi *et al*., 2021). They are parts of the exhaust fumes and chimneys of gasoline-based industrial machines (Yu *et al*., 2014a, b). PAHs are several chemical compounds found in crude oil (petroleum) (Li *et al*., 2015; Liang *et al*., 2016; Liberti *et al*., 2016).

Aromatic compounds like PAHs contain at least 2 benzene rings fused but without substituents/heteroatom (Tesi *et al*., 2021). They are also heavy or lightweight (made up of four benzene rings or without), respectively, which gives them their noxious or stable nature (Wang *et al*., 2016a, b). Their presence in different compartments can elicit mutagenic and carcinogenic issues because of their dietary contact and persistent nature (Tesi *et al*., 2021). Seven congeners of PAHs like benzo[k] fluoranthene (BkF), indeno[1,2,3-cd] pyrene (IndP), dibenzo[a,h]anthracene (DahA), benzo[a]anthracene (BaA, benzo[b]fluoranthene (BbF), chrysene (chry), and benzo[a]pyrene (BaP), have been linked to causing these health effects (Tesi *et al*., 2021). This calls for special biomonitoring because of the hidden or unforeseen issues it portends.

Hence, the focus of this study was to assess the content of PAHs in *T. occidentalis* grown in crude oil communities of the Niger Delta and their potential health hazards when consumed.

Methodology

Area of study

The study area is comprised of nine states that are situated in the Delta of the River Niger, and these are Delta, Edo, Cross River, Rivers, Bayelsa, Imo, Akwa-Ibom, and Ondo Figure 1. The state's combined population accounts for 28 percent of Nigeria's population (National Population Census, 2006). The Niger Delta plays host to three refineries in Nigeria and has the only gas plant and liquefied natural gas trains in Okpai and Bonny Island, respectively.

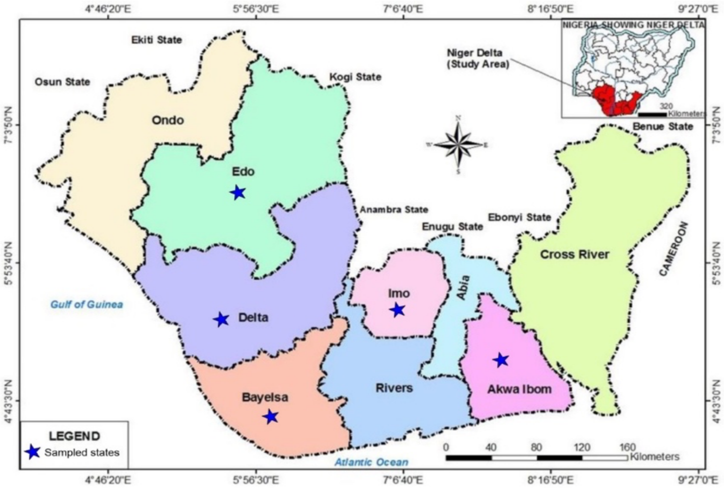

Figure 1. Map of Niger Delta showing the studied states

Ethical consideration

Samples were collected from vegetable farms after due permission was obtained from the owners of the farms and community leaders. Permits were not obtained from the government because there is no law in place prohibiting such sample collection.

Sampling

Five states in the 9 Niger Delta states: Akwa-Ibom, Bayelsa, Delta, Imo, and Rivers State. Were randomly selected to make the research sampling station, and samples of *T. occidentalis* were collected from (5 farms in 5 communities). This study spanned from May to September 2023 (6 months) and the sample collection was done by research assistants (the locals) from various villages and they were 25 in number.

The communities where samples were collected were Akwa-Ibom state (Edo, Ikot Iben, Uquo Ibeno, Eyet Ibeno, and Ukpenokang), Bayelsa state (Abuku, Adagbabiri, Ajoro, Agalabiri, and Agorogbene), Delta State (Uzede, Uweye, Afikero, Uhroko, and Iboro), Imo State (Orsu-Obodo, Igbukankwu, Nabachu, Abatu and Ishube), and Rivers state (Afradiku, Ajomotor, Agbadama, Agana and Agbaduchianna). The samples collected from the five communities from each state were bulked, swathed in an aluminum wrap, and kept in a temperature (4°C) control container (cooler) for posterior analysis.

Data analysis

The samples collected were screened thoroughly via washing and rinsing of debris with deionized fluid (water). The samples were later shredded using a blender and later homogenized using the technique of the European Commission Regulation 1255/2020, as described in (Quing *et al*., 2023). The homogenized vegetable samples were dehydrated in an Agilent door oven at 102 °C for 12 hours, and 2 g of each were weighed into flasks made with polypropylene tubes for centrifugation, and into 10 ml of 1:1 ethylene acetal cyclohexane was added, vortexed, and sonicated for 15 minutes. The supernatant was centrifuged at 4500 r/mm for 5 minutes. The top phase of the supernatant at the upper level was placed into a vial and then injected into a gas chromatograph gel permeation type that has a fitted purification column. 1:1 v/v ethylene acetate cyclohexene was adopted in the mobile phase. The eluent obtained was then used in the determination of the target PAHs using an HPLC-FLD (high-performance liquid chromatograph fitted with a flame detector) model 6900.

Valuing exposure

The PAH content in Nigerian fluted pumpkin was used to determine the health risks associated with PAH15. To calculate or estimate the TEFs of the PAH15 the following assumptions values of the selected PAHs: BaP, BaA, BaF, chrysene, and pyrene were: 1, 0.1, 0.1, 0.01, and 0.001, respectively, were used in this study with the following equation:

$$TEQBap = \sum_{i=1}^{n} Ci * TEFi$$

Where the concentration of PAHs in the fluted pumpkin is denoted by the values of TEFi and Ci. The entire set of PAHs (TEQBaP-Bapeq 15) was utilized to assess the risk of carcinogenesis. BaP is used as an equivalent standard to quantify TEEQ as reported by Dadar *et al.*, (2017) and US EPA (1992).

The chronic daily intake (CDI) of PAHs was calculated with the equation below:

$$CDI\ (ng * BaPeq\ per\ bw.d) = \sum Ci * lRi * ED * \frac{ET}{Bw * AT}$$

Where the concentration of PAH15 assessed for BaPeq in ng BaPeq/g for the fluted pumpkin is shown by the value of Ci. The fluted pumpkin was indicated in grams per day by the daily intake (IRi). The assumption of the following ED, EF, and AT (exposure days, exposure frequency, and average time) is 53, 365 days/years, and 70 years corresponding. While the IR and BW (ingestion rate and body weight) were 25,550 days and 65 days respectively (European Food Safety Agency, 2008). The carcinogenic risk was computed with the formula below:

ILCR = CDI*SF*CF

Where the incremental life cancer risk (ILCR) is dimensionless, the SF (slope factor) for the consumption of PAHs for BaP is estimated at 7.3 mg/kg/day. While the CF (conversion factor) is 0.000006 mg/mg.

Data computation

An analysis of variance (ANOVA) and the calculation of the mean and standard error (SE) were carried out using the Statistical Package for Social Scientists (SPSS) 29 and Microsoft Excel 2019 and the Windows 10 Pro application to determine whether there were any significant differences in the samples acquired.

Results

The results of the analysis of the PAHs content of *T. occidentalis* grown in crude oil communities of the Niger Delta were shown in Figures 2-6 and the mean comparison of the PAHs content from the crude oil communities (Figure 7). The mean results of the PAHs in *T. occidentalis* grown in the crude oil states of the Niger Delta were subjected to the test of significance with the statistical instrument of analysis of Variance (ANOVA) using a special package for social sciences (SPSS) model 29 (IBM) at the level of significance of 0.05 and p-value was 0.48 thus rejecting H_0.

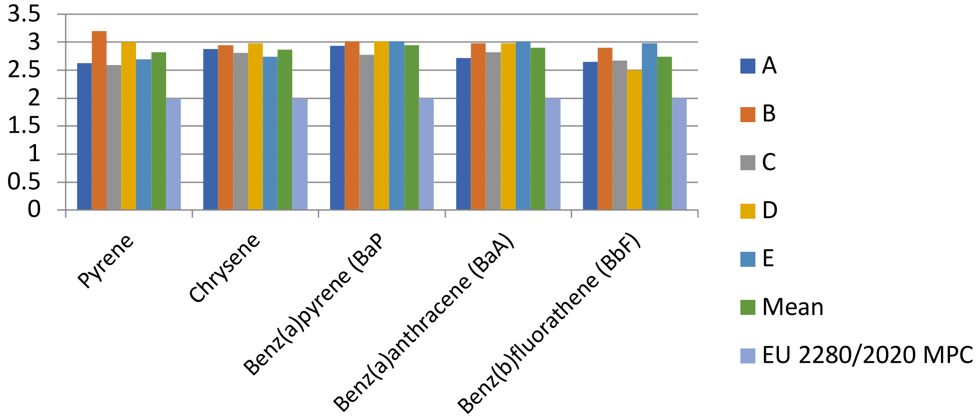

Figure 2. The analyses of PAHs content in *T. occidentalis* grown in Ibeno Akwa-Ibom state and EU 1255/2020 MPC for PAHs in vegetables in µg/kg

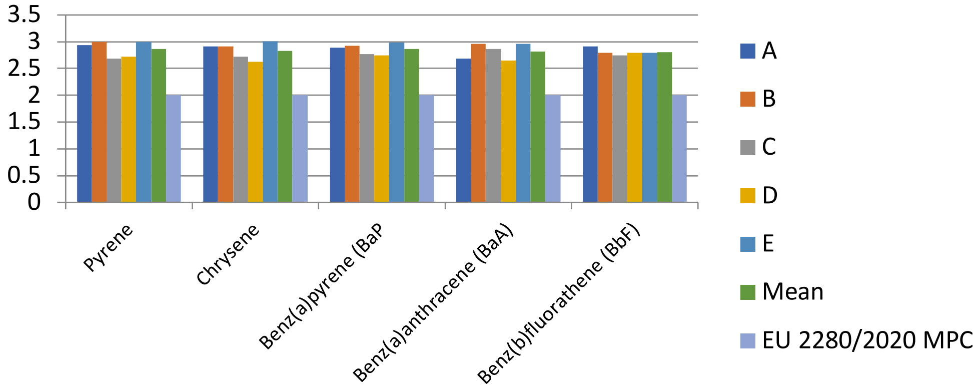

Figure 3. The analyses of PAHs in *T. occidentalis* cultivated in the Samagbama crude oil community of Bayelsa state and EU 1255/2020 MPC for PAHs in vegetables in µg/kg

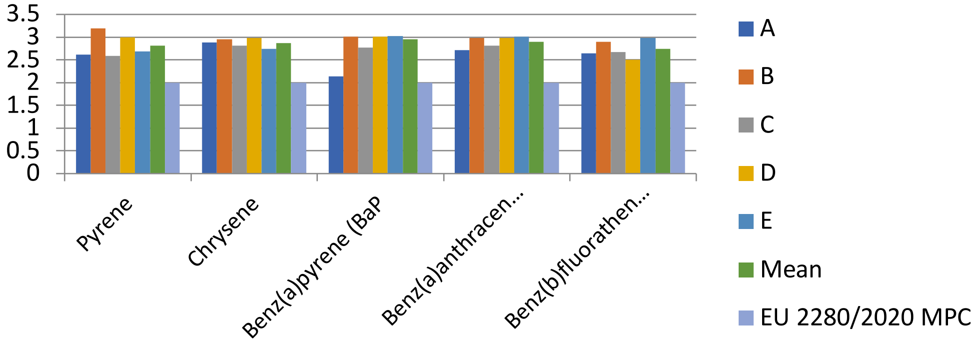

Figure 4. The PAHs in *T. occidentalis* grown in Uzere community, Delta state, and EU regulation 1255/2020 MPC for PAHs in vegetables in µg/kg

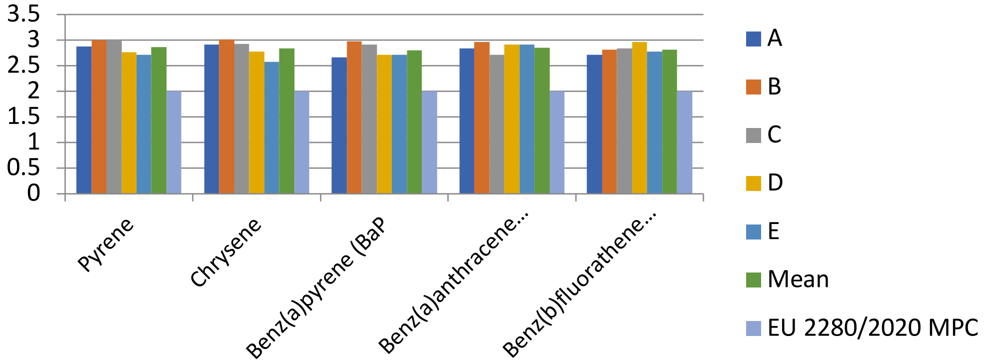

Figure 5. The PAHs content analysis of *T. occidentalis* in the Oguta crude oil community Imo state and EU 1255/2020 MPC for PAHs in vegetables in µg/kg

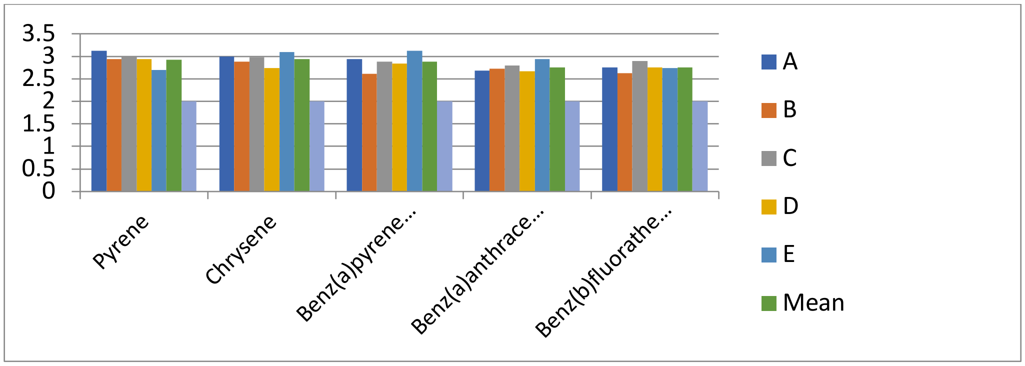

Figure 6. The analysis of the PAH concentrations in *T. occidentalis* harvested in the Ibeno crude oil community River state and EU regulation 1255/2020 MPC for PAHs in vegetables in µg/kg

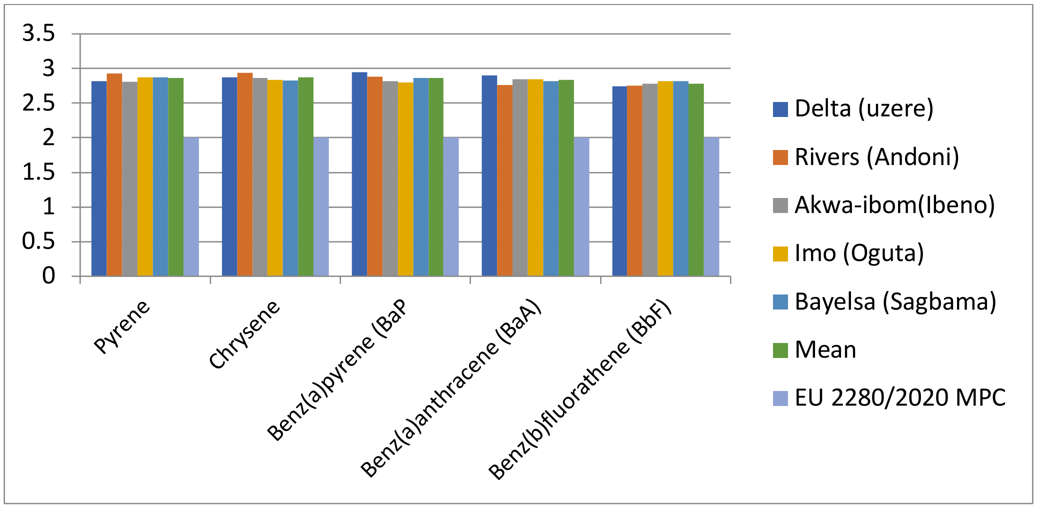

Figure 7. Comparative mean results of the PAHs harvested in crude oil states of the Niger Delta and EU regulation 1255/2020 MPC for PAHs in vegetables in µg/kg

Health risk calculation of PAHs in fluted pumpkin species

The results for the non-carcinogenic risks from the possible ingestion of PAH congeners in the fluted pumpkin sourced from different locations in the Niger Delta are shown in Tables 1 and 2. According to the data, the toxicity equivalent factors for benzo (a) pyrene were the highest at 14.55 µg/kg, followed by benzo (b) fluoranthene (1.46 and 1.42) and benzo (a) anthracene. In the meanwhile, the station-wide total was much more than the typical bounds (Table 1). The study's CDI values for each of the PAH congeners varied across the station and were notably high (Table 2).

Nevertheless, the ILCR findings indicated that the levels exceeded the predetermined threshold (Table 3).

Table 1. Factors that indicate the toxicity of certain PAH congeners in fluted pumpkin

		Beneze (a) pyrene (µg/kg)	Pyrene (µg/kg)	Chrysene (µg/kg)	Benzo (a) anthracene (µg/kg)	Benzo (b) fluoranthene (µg/kg)	
No fluted pumpkin sample	Designate	TEQBaP	TEQBaP	TEQBaP	TEQBaP	TEQBaP	Sum
5	Akwa-Ibom	2.95	0.00	0.03	0.29	0.27	**3.55**
5	Bayelsa	2.86	0.00	0.03	0.28	0.28	**3.45**
5	Delta	2.95	0.00	0.03	0.29	0.27	**3.55**
5	Imo	2.8	0.00	0.03	0.29	0.28	**3.40**
5	Rivers	2.99	0.00	0.03	0.31	0.31	**3.64**
	Σ	**14.55**	0.01	0.15	**1.46**	**1.42**	

*EU 1255/2020 MPC for PAHs in fluted pumpkin is 2.0 µg/kg

Table 2. Possible CDI in the fluted pumpkin of certain PAH congeners

No of fluted pumpkin sample	Designate	Benzo (a) pyrene (µg/kg)	CDI	Pyrene (µg/kg)	CDI	Chrysene (µg/kg)	CDI	Benzo (a) anthracene (µg/kg)	CDI	Benzo (b) fluoranthene (µg/kg)	CDI
5	Akwa-Ibom	2.95	461.46	2.82	441.13	2.87	448.95	2.9	453.64	2.98	466.16
5	Bayelsa	2.86	447.39	2.87	448.95	2.83	442.69	2.82	441.13	3.11	486.49
5	Delta	2.95	461.46	2.82	441.13	2.87	448.95	2.9	453.64	3.01	470.85
5	Imo	2.8	438.00	2.87	448.95	2.84	444.26	2.85	445.82	3	469.29
5	Rivers	2.99	467.72	3.2	500.57	3.11	486.49	3.1	484.93	3.1	484.93

*EU 1255/2020 MPC for PAHs in fluted pumpkin is 2.0 µg/kg

Table 3. Probable incremental life cancer risk of selected PAH (Benzo (a) pyrene) in fluted pumpkin

No Vegetable sample	Designate	Benzo (a) pyrene (µg/kg)	ILCR
5	LA-IB	2.95	0.02
5	AL-OT	2.86	0.02
5	OK-AB	2.95	0.02
5	OR-NT	2.8	0.02
5	LK-EP	2.99	0.02

US EPA (1992) standard for ILCR: $*1\times10^{-6}$ to 1×10^{-4}

Discussion

Quantification of PAH contents in the fluted pumpkin

Several studies have been carried out on the PAH contamination of foods and vegetables (Net 2014; Motorykin *et al*., 2015; Nam *et al*., 2015). However, research on PAH contamination of foods and vegetables in crude oil-producing areas remains scanty hence this study. The pyrene content of *T. occidentalis* grown in the Niger-Delta crude oil communities revealed a range from 2.81 µg/kg in the Ibeno crude oil community of Akwa Ibom state to 2.93µg/kg in the Andoni crude oil community Rivers state with a mean concentration of 2.86µg/kg. The elevated level of pyrene in the vegetables is the result of oil activities and bioaccumulation. A similar report of high content of pyrene in vegetables was in (Lin *et al*., 2014; Jung *et al*., 2015). Pyrene has been fingered in various cancers and mutations (Mao, 2016; Mahler, 2014; Ogwu *et al*., 2022).

The mean concentration of chrysene in the *T. occidentalis* analyzed was between 2.83 µg/kg in Sagbama Bayelsa state to 2.94 µg/kg in Andoni Rivers state with a group mean concentration of 2.87 µg/kg. The high content of chrysene in *T. occidentalis* in the Niger Delta is anthropogenic. This result corroboration reports by (An *et al*., 2012; Ranzi *et al*., 2013). Exposure of humans to higher doses of chrysene has been associated with lung cancer (Semedo, 2014).

Analysis of *T. occidentalis* grown in the Niger-Delta crude oil communities for BaP also revealed its concentration to be between 2.80 µg/kg in the Oguta crude oil community Imo state to 2.95µg/kg in Uzere crude oil community of Delta state with a mean concentration of 2.84 µg/kg. The concentration of BaP above EU regulation 12550/2020 is a result of man's interaction with the environment. This report is in agreement with (Moscoso *et al*., 2012; Monza *et al*., 2013). The health effects of BaP in humans are cancer of the bladder and osteoporosis (Na *et al*., 2011; Mulder *et al*., 2015; Alagić *et al*., 2015; Angion *et al*., 2015).

BaA content of analysis of the *T. occidentalis* cultivated in the Niger-Delta crude oil communities presented various concentrations ranging from 2.76 µg/kg in the Andoni oil-bearing community of River state to 2.90 µg/kg in Uzere in Delta state with a comparative mean concentration of 2.84 µg/kg. The high content of BaA is the result of its bioavailability and bioaccumulation. This result is in tandem with the reports presented by Araghi, (2014), Ratola, (2013), and Sazakli, (2015). BaA has been associated with lung cancer (Amezcua-Allieri, 2012; Ahmed, 2015). Skin and bladder cancer (Qin *et al*., 2014).

The analysis of *T. occidentalis* grown in the Niger-Delta crude oil communities for BbF content equally showed a range of concentration of 2.74 µg/kg in the Uzere oil-bearing community in Delta state to 2.82 µg/kg in Sagbama crude oil community of Bayelsa state with a comparative mean content of 2.78 µg/kg. This report of increased BbF in *T. occidentalis* aligns with reports of BbF in vegetables documented in (Ren *et al*., 2015). The consequences of prolonged exposure of man to BbF include skin irritations, and nasal and throat cancer (Mulder *et al*., 2015). Cancer of the bladder (Walker, 2015).

Likely dangerous to calculate the number of PAHs in fluted pumpkin

There was adequate concentration of the chosen PAHs in fluted pumpkins according to the results of the health risk assessment of the species. The fluted pumpkin of BaP, BbF, and BaA included significant amounts of PAHs, making the TEFs notable. There might not be a cancer risk if the species under study is consumed more than the daily allowance. The CDI data showed a positive synergy with the TEF outcomes. For fluted pumpkin species, the TEFs and CDI levels were both substantially greater than the EU regulatory requirements (2011 and 2020). Tesi *et al*., (2021), Igwe *et al*., (2022), and Liu *et al*., (2023) have also documented comparable situations.

The incremental life cancer risk result (1×10^{-2}) in this study showed a considerable increase when compared to the US EPA (1992) (1×10^{-6} to 1×10^{-4}) and Liu *et al*., (2023) guidelines for contact assessment for PAHs compounds. Therefore, eating fluted pumpkins increases the risk of cancer in humans.

The claims that PAHs in the soil, air, and water environments cause bioaccumulation and biomagnification in crops and animals in the environment have been further supported by this study. *T. occidentalis* cultivated in the Niger Delta crude oil region has been analyzed, and the results indicated that the vegetables are excessively polluted and unfit for human consumption or export. In light of these conclusions, the report suggested that oil firms doing business in the Niger Delta be forced to use international best practices in their operations to prevent environmental degradation, which is mostly caused by oil spills into the aquatic and soil environments. Models of mitigation must be developed, and the process of repairing soil harm must begin. The National Oil Spills Detection and Research Agency, a monitoring organization, ought to keep a closer eye on the Niger Delta's oil exploration and exploitation firms.

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