DETERMINATION OF POLYCYCLIC AROMATIC HYDROCARBONS (PAHS) IN *OREOCHROMIS NILOTICUS* FROM OGUN RIVER AT AJEGUNLE, LAGOS STATE.

BY

Faith ILENBS-OKOISE (Miss)
AGR1800280

DEPARTMENT OF AQUACULTURE AND FISHERIES MANAGEMENT
FACULTY OF AGRICULTURE
UNIVERSITY OF BENIN
BENIN CITY, NIGERIA

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A REPORT SUBMITTED TO THE DEPARTMENT OF AQUACULTURE AND FISHERIES MANAGEMENT, FACULTY OF AGRICULTURE, UNIVERSITY OF BENIN, IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF THE BACHELOR OF AGRICULTURE DEGREE B.AGRIC (FISHERIES)

MAY, 2024.

CERTIFICATION

This is to certify that this research was carried out by ILENBS-OKOISE Faith in the Department of Fisheries, Faculty of Agriculture, University of Benin, Benin City, Edo State, Nigeria.

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Dr. O. M Wangboje **Date**

(Project Supervisor)

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Dr. O. M Wangboje **Date**

(Head of Department)

DEDICATION

I dedicate this project to God almighty for His sustenance and guidance throughout the period of study.

I also dedicate this project to my loving Parents; Mr and Mrs Ilenbs-Okoise, that gave the resources and values necessary to be who and where I am today and their support on every step and decision I make. Their prayers have kept me, for I am indeed grateful.

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I will like to thank God for His guidance, grace and unending love throughout this research journey.

I extend my heartfelt thanks and appreciation to my project supervisor Dr. O. M. Wangboje for his corrections and advise throughout this project. I would also love to appreciate my Head of Department Dr. O. M. Wangboje and all my esteemed lecturers for their dedication in imparting knowledge and skills that have shaped my academic journey

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Last but not the least I would love to acknowledge my friends, Greatness, Victory, Victor and Julian it wouldn't have been a success if not for this people.

Thank you all from the bottom of my heart, I LOVE YOU ALL

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ABSTRACT

Polycyclic aromatic hydrocarbons (PAHs) have received significant attention due to their potential health and environmental impacts. Some are toxic and pose risks to human health and ecosystems. This study was conducted to determine the levels of PAHs in *Oreochromis niloticus* from Ogun river in Ajegunle, South-West Nigeria. Gas Chromatography with Flame Ionization Detector (GC/FID) was used to measure the concentrations of PAHs in the fish tissues, following United State Environmental Protection Agency (USEPA) methods. The objectives of the research evaluated the toxic equivalency (TEQ) of PAHs, toxic/hazard quotient for PAHs, estimated daily intake with annual intake and cancer risk factor for PAHs in *O.niloticus.*

Naphthalene had a low value in the fish sample analyzed and could be related to the fish species inability to absorb it irrespective of its presence in the water and sediments. Total PAH concentrations found in the fish sample exceeded the acceptable limit set by the European Commission (2 µg/kg Fluroanthene and

Benzo(a)pyrene) for fish considered safe for human consumption. The quota for PAHs ranged from 0.037% for Naphthalene to 73.97% for Fluroanthene. The continual consumption of fish from the river by the inhabitants could lead to bioaccumulation with adverse health implications since PAHs are carcinogenic, mutagenic and teratogenic. Furthermore, the high concentrations and the persistent nature of PAHs make the fish that inhabit the river unfit for human consumption.

CHAPTER ONE

1.0 INTRODUCTION

Water pollution and the lack of access to clean water are general global problems that result from the expansion of industrial and agricultural activities (Wang *et al*.,2019). In recent decades, organic compounds such as polycyclic aromatic hydrocarbons (PAHs) have commonly been observed in aquatic environments. Moreover, the number of new organic compounds arriving the worldwide market is increasing remarkably every year, and most of these compounds, including pharmaceuticals, pesticides, personal care products, and PAHs surfacants are used worldwide in high amounts in industrial activities, after which they are discharged in to various water bodies, where they can persist, causing severe health and environmental problems (Zambianchi *et al*.,2017).

Polycyclic Aromatic Hydrocarbons (PAHs) are described as an assemblage of chemicals formed as a result of incomplete combustion of organic matter. They are especially recognized in the scientific and medical community for their carcinogenicity, mutagenicity and teratogenicity. PAHs are documented to be priority contaminants according to the United States Environmental Protection Agency (Barakat *et al*., 2011; Li *et al* 2015; Cui *et al*., 2016; Adekunle *et al*., 2018; Kosek and Ruman, 2021; Shariatifar *et al*., 2021; Ambade *et al*., 2023; He *et al*., 2023). Nwaichi and Ntorgbo, (2016), also stated in their report that there is an alarmingly high levels of PAH-based pollutants in the aquatic ecosystem due to significant increases in anthropogenic activities along with unavoidable process of biotransformation and biomagnification.

PAHs have been classified into pyrolytic and petrogenic groups based on their mode of formation and orgin. The former is produced via the combustion of organic matter, while the latter is directly linked with the petroleum industry. Sources of PAHs in the environment include volcanic eruptions, forest fires and biogenic formations (El-Maradny *et al*., 2023).

These pollutants are capable of presenting significant health risk to human by oral intake through food, inhalation, and even dermal interaction. Exposures through any of the listed routes could bring about health challenges of short and long-term effects, including some major respiratory and cardiovascular diseases (Perez- Padilla *et al*., 2010; WHO 2014). Their reactive metabolites, such as epoxides and dihydrodiols, are considered more concerning, given their ability to bind to cellular proteins and DNA and adduct formation (Balbo *et al*., 2014; Blaszczyk *et al*., 2017).

In addition, on the basis of their molecular structure, PAHs can be classified into two categories, namely the low-molecular-weight (LWM) PAHs which contain four or fewer aromatic rings and the high-molecular-weight (HWM) PAHs which contain five or more aromatic rings (Wang *et al*., 2022).

Considering the dangers PAHs represent to the environment, wildlife, and the health of man, scientific research on them has attracted considerable attention in many developed nations. However, there are significant data gaps regarding PAHs sources and levels in developing nations (Ogunfowokan *et al*., 2003; Nieuwoudt *et al*., 2011; Okedeyi *et al*., 2013; Mirza *et al*., 2014; Zheng *et al*., 2020; Sharma *et al*., 2021; Usese and Egbuta, 2021; Ambade *et al*., 2023). Fish are important bioindicator species and play an important role in the monitoring of water pollution because they respond with great sensitivity to changes in the aquatic environment (Naigaga *et al*., 2011). A previous report by Xia *et al*. (2010) however noted that dietary intake constitutes a major pathway of PAHs exposure in humans. In addition, increased risks of cancer in humans have been attributed to dietary exposure to elevated concentrations of PAHs (Yoon *et al*., 2007; Stacewicz Sapurtzakis *et al*., 2008). Hence, the risk derived from exposure to chemical pollutants via frequent consumption of fish has been an issue of concern in contrast to the potential health benefits of dietary fish intake (Nwaichi and Ntorgbo, 2016).

1.1 Justification of Study

In Nigeria, a number of studies have shown a steady increase in the levels of pollutants of priority concerns including PAHs and their bioaccumulation in fishery resources from several ecosystems (Nkpaa *et al*., 2013; Nwaichi and Ntorgbo, 2016; Tongo *et al*., 2017; Usese *et al*., 2017; Igbo *et al*., 2018; Ekere *et al*., 2019; Olayinka *et al*., 2019).

Lagos lagoon ranks first among most polluted African ecosystem (Alani *et al*., 2017) and is primarily impacted by effluents from oil and textile industries, urban sewage carried by Ogun and Osun Rivers, farming activities along the river course that uses agrochemicals to boost production and contribute to National food security. Activities along Ogun River include abbatoirs, market, residential communities and brewery. Ogun river is one of the most major source of fish, other aquatic products and livelihood for fishermen in South Western part of Nigeria.

Also, studies have shown the risk associated with dietary exposures of PAHs in muscle tissues of *Oreochromis niloticus* from creeks in South-West Nigeria where they are abundant, which has resulted in consequent health implications on the consumers (Okenyi *et al*., 2016). Therefore, this research has extended the data on PAHs in order to protect consumers health and to direct existing and prospective consumers of such fish in that geographical zone.

1.2 Aim and Objectives of the Study

The aim of the study was to determine the levels of polycyclic aromatic hydrocarbons (PAHs) in *Oreochromis niloticus* (Nile Tilapia) in Ogun River at Ajegunle, Lagos state, Nigeria.

1. The specific objectives of the study were to determine the;

2. polycyclic aromatic hydrocarbons (PAHs) (Benzo(a)pyrene, Anthracene, Fluroanthene and Napthalene) levels in *O. niloticus* from Ogun river at Ajegunle, Lagos State, Nigeria.

3. toxic/hazard quotient for PAHs in *O. niloticus* ;

4. toxic equivalency (TEQ) for PAHs;

5. estimated daily intake and annual intake

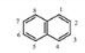
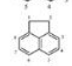
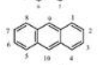
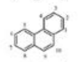
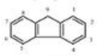
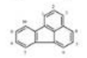
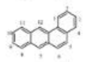
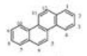
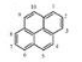
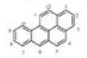
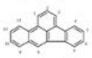
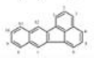
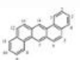
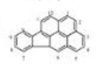
6. cancer risk factor for PAHs

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 POLYCYCLIC AROMATIC HYDROCARBONS

Polycyclic aromatic hydrocarbons are organic pollutants and composed of two or more fused aromatic rings of carbon and hydrogen atoms, which are primarily colorless, white, or pale yellow solid compounds (Abdel-Shafy and Mansour, 2016; Suman *et al*., 2016). The molecular arrangements of aromatic rings in space can be linear, angular, or in clusters (Abdel-Shafy and Mansour, 2016). With the number of rings present in the compounds, PAHs are classified into light-molecular weight PAHs (LMW PAHs; having two or three aromatic rings) and high-molecular weight PAHs (HMW PAHs; having four or more aromatic rings). Depending upon their molecular weight, they are emitted either as gaseous phase (LMW PAHs) or in the particulate form (HMW PAHs). Further, based on the structure of rings, PAHs are also classified as: alternant PAHs, which contain only fusion of six carbon benzene rings, whereas the non-alternant PAHs like fluorene contain fusion of six carbon benzene rings along with an additional ring of less than six carbon (Gupte *et al*., 2016). The existence of dense π electrons on aromatic rings is responsible for the biochemical persistence of PAHs that make PAHs more resistant to nucleophilic attack. The United States Environmental Protection Agency (USEPA) has declared 16 PAHs as priority pollutants in 1983 based on their existence of highest concentrations, greater exposure, recalcitrant nature, and toxicity (Zheng *et al*., 2018; Mojiri *et al*., 2019). PAHs are characterized through their low water solubility, low vapor pressure, and high melting and boiling points, depending on their structures. PAHs with increased molecular weight are tending to decrease water solubility and increase lipophilicity, making them more recalcitrant compounds. The table below shows the physiochemical properties of 16 aromatic hydrocarbons.

Name	Formula	Structure	Molecular weight (g/mole)	Solubility in water (mg/L)	Phase distribution	Melting point (°C)	Boiling point (°C)	Vapor pressure (mmHg)	Log Kow	Log Koc	Toxicity as per IARC
Naphthalene	$C_{10}H_8$		128.17	31	Gas	80.26	218	0.087	3.29	2.97	2B
Acenaphthene	$C_{12}H_{10}$		154.21	3.8	Gas	95	96	4.47×10^{-3}	3.98	3.66	3
Acenaphthylene	$C_{12}H_8$		152.20	16.1	Gas	92–93	265–275	0.029	4.07	1.40	3
Anthracene	$C_{14}H_{10}$		178.23	0.045	Particle gas	218	340–342	1.75×10^{-6}	4.45	4.15	3
Phenanthrene	$C_{14}H_{10}$		178.23	1.1	Particle gas	100	340	6.8×10^{-4}	4.45	4.15	3
Fluorene	$C_{13}H_{10}$		166.22	1.9	Gas	116–117	295	3.2×10^{-4}	4.18	3.86	3
Fluoranthene	$C_{16}H_{10}$		202.26	0.26	Particle gas	110.8	375	5.0×10^{-6}	4.90	4.58	3
Benzo(a)anthracene	$C_{20}H_{12}$		228.29	0.011	Particle	158	438	2.5×10^{-6}	5.61	5.30	2B
Chrysene	$C_{18}H_{12}$		228.29	0.0015	Particle	254	448	6.4×10^{-9}	5.9	No data	2B
Pyrene	$C_{16}H_{10}$		202.26	0.132	Particle gas	156	393–404	2.5×10^{-6}	4.88	4.58	3
Benzo(a)pyrene	$C_{20}H_{12}$		252.32	0.0038	Particle	179–179.3	495	5.6×10^{-9}	6.06	6.74	1
Benzo(b)fluoranthene	$C_{20}H_{12}$		252.32	0.0015	Particle	168.3	No data	5.0×10^{-7}	6.04	5.74	2B
Benzo(k)fluoranthene	$C_{20}H_{12}$		252.32	0.0008	Particle	215.7	480	9.59×10^{-11}	6.06	5.74	2B
Dibenz(a,h)anthracene	$C_{22}H_{14}$		278.35	0.0005	Particle	262	No data	1×10^{-10}	6.84	6.52	2A
Benzo(g,h,i)perylene	$C_{22}H_{12}$		276.34	0.00026	Particle	273	550	1.03×10^{-10}	6.50	6.20	3
Indeno[1,2,3-cd] pyrene	$C_{22}H_{12}$		276.34	0.062	Particle	163.6	530	10^{-10}–10^{-16}	6.58	6.20	2B

Kow is n-Octanol/Water Partition Coefficient. Log Kow is useful for predicting the distribution of compound in the environment, high Log Kow indicates low water affinity and high hydrophobicity. Compounds with Log Kow > 4.5 have higher bioaccumulation rates.

Koc is Soil Adsorption Coefficient. Log Koc is useful for predicting the mobility of compound in soil, i.e., distribution and exposure level of compound, high Log Koc indicates strong adsorption onto soil and organic matter. Compounds with Log Koc > 4.5 have potential adverse effects on terrestrial organisms.

Toxicity as per International Agency for Research on Cancer (IARC): group 1-human carcinogen, group 2A-probable human carcinogens, group 2B-possible human carcinogens, and group 3-not classifiable as

The sources of PAH pollution are categorized mainly into two, such as anthropogenic emission sources and natural emission sources (Mojiri *et al*., 2019). Natural emission sources such as volcanic eruptions, natural forest fire, and moorland fire caused by lightning flashes are negligible or less important(Abdel-Shafy and Mansour, 2016). Anthropogenic emission sources are the main determinants of PAH pollution, which can be divided into four types, i.e., industrial, mobile, domestic, and agricultural emission sources (Ravindra *et al*., 2008). Incomplete combustion is the prime source of PAH emissions by various industrial activities such as waste incineration, iron and steel production, aluminum production, cement manufacturing, coal-tar pitch production, dye manufacturing, asphalt industries, rubber tire manufacturing, fungicide and insecticide production, exhaust from refineries, and power production (Abdel-Shafy and Mansour, 2016; Gupte *et al*., 2016; Mojiri *et al*., 2019). Other industrial emission sources are coal gasification, electric arc furnace, oxygen furnace, diesel engine, and gasoline-powered engines of large machineries. Mobile emission sources include exhaust from many vehicles like aircrafts, ships, trains, and off-road heavyweight and lightweight vehicles (Srogi *et al*., 2007). The figure below shows different types of PAHs emissions.

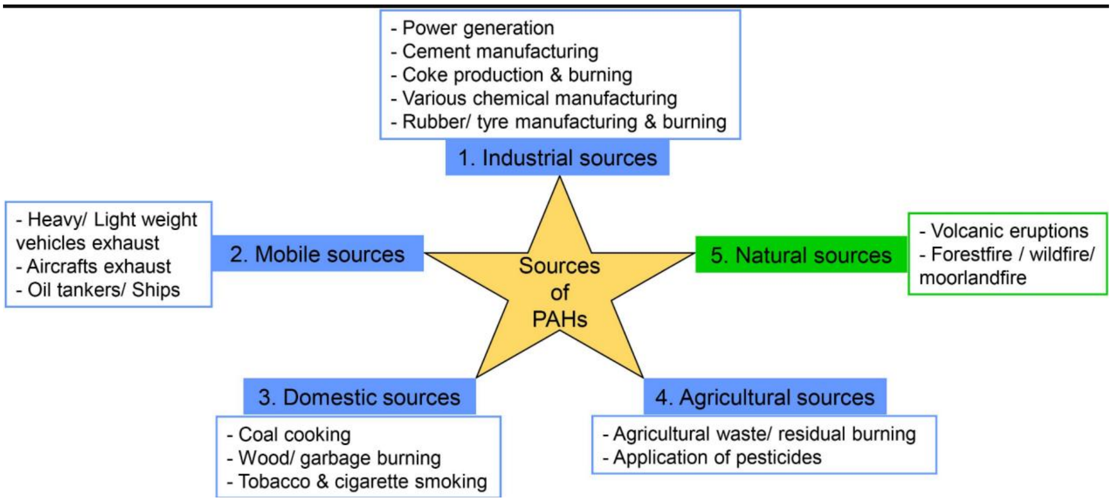


Figure 1: The different types of PAHs emissions

Atmospheric PAHs (gaseous phase as aerosols) are deposited in water, soil, and plants in the particulate phase through dry/wet deposition processes (Abdel-Shafy and Mansour, 2016). PAHs with three or more aromatic rings are very strong adsorbent to the soil particles due to low vapor pressure and high hydrophobicity (Abdel-Shafy and Mansour, 2016). Accumulation of PAHs in soil/sediment is responsible for further transport of pollution to the groundwater, plants, and food, e.g., plant roots absorb PAHs from polluted soil and translocate to the farther plant parts. Exposure to PAHs is unavoidable in the current situation. PAH exposure occurs mainly via three routes, i.e., inhalation, ingestion, and dermal contact. It is also possible that exposure can occur via more than one route, simultaneously, e.g., dermal and inhalation exposures from contaminated soil (Rengarajan *et al*., 2015; Abdel-Shafy and Mansour, 2016).

2.2 Biotoxicity of Noxious Polycyclic Aromatic Hydrocarbon Pollutants.

Individual PAH compounds do not have the exact same health effects (Rengarajan *et al*., 2015; Abdel-Shafy and Mansour, 2016). Many PAHs are mutagenic, carcinogenic, teratogenic, and immunotoxic to living organisms, including microorganisms, animals, and humans (Burchiel and Gao, 2014; Rengarajan *et al*., 2015; Bolden *et al*., 2017). PAHs have ecotoxic effects on aquatic life and birds (Abdel-Shafy and Mansour, 2016). The mode of exposure, exposure duration, and exposure dose are important parameters for the severity of PAHs' toxic effects (Rajpara *et al*., 2017; Tong *et al*., 2018). Zheng *et al*. (2018) found that PAHs from different sources showed different risk levels, and they calculated incremental lifetime cancer risk (ILCR) in humans for soil-bound PAHs upon the three different exposure routes; the highest cancer risk was found for ingestion, i.e., 98.1–99.3%, followed by dermal contact, i.e., 0.66–1.83%, and inhalation, i.e., 0.03–0.04%. Toxic effects of PAHs may vary according to factors such as pre health status and age. Acute health effects include eye irritation, vomiting, diarrhea, confusion, skin irritation, and inflammation (Abdel-Shafy and

Mansour, 2016). Naphthalene, anthracene, and benzo(a)pyrene are direct skin irritants and skin sensitizers for animals and humans (Rengarajan *et al*., 2015). Chronic health effects include eye cataracts, kidney and liver damages, breathing problems, decreased immune function, lung malfunctions, and asthma-like symptoms (Abdel-Shafy and Mansour, 2016). Naphthalene can cause the breakdown of red blood cells if inhaled or ingested in high amounts (Rengarajan *et al*., 2015).

2.3 Phototoxicity of Polycyclic Aromatic Hydrocarbons

Sunlight has three components, i.e., 91.0% visible light (400–700 nm), 8.7% UVA light (320–400 nm), and 0.3% UVB light (280–320 nm) (Yan *et al*., 2004). PAHs can absorb UVA and visible light. Due to the absorption of UVA light, formation of reactive species upon electron or energy transfer from the excited PAHs and formation of reactive intermediates upon the reaction of excited PAHs with oxygen or other molecules occur in the cells. These reactive species or intermediates are responsible for damages to cellular components such as cell membrane, nucleic acid, or proteins. PAH-contaminated human skin exposed to sunlight irradiation can cause DNA single-strand cleavage, oxidation of DNA bases, and formation of DNA covalent adducts, indicating PAH toxicity can exceed over 100 times in the presence of light as compared to dark (Yu, 2002; Yan *et al*., 2004).

2.4 Immunotoxicity of Polycyclic Aromatic Hydrocarbons

Polycyclic aromatic hydrocarbons have some immune system-related adverse effects like inhibition of pre B, pre T, and myeloid cell development, B and T cell suppression, apoptosis of lymphoid tissues, disruption of myelopoiesis, and altered cytokine production by macrophages and monocytes (Burchiel and Luster, 2001; Burchiel and Gao, 2014; Rengarajan *et al*., 2015). Under specific circumstances, tumor development, hypersensitivity (allergy), and autoimmunity may develop (Abdel-Shafy and Mansour, 2016). PAHs bind with specific aryl hydrocarbon receptors (AhRs) in lymphocytes and accessory cells of the immune system, upregulate AhR-controlled metabolic enzymes cytochrome P450, and produce immune toxic oxidative and electrophilic metabolites (Burchiel and Gao, 2014; Marris *et al*., 2020).

2.5 *Orechromis niloticus* (Nile Tilapia)

Nile tilapia (*Oreochromis niloticus*) is a significant species in both aquaculture and the wild, with a wide distribution and potential ecological impact. In aquaculture ponds, it has been found to have a complex relationship with the ecosystem, with its stocking density affecting fish growth and trophic levels . However, its introduction as an invasive species in South China has led to ecological problems, including a decrease in native fish species and water quality (Gu, 2017). In the Pearl River, its invasion has disrupted the functional patterns of the fish community, particularly in terms of body size (Fangmin, 2019). In the African Great Lakes, including Lakes Victoria and Kyoga, the introduction of Nile tilapia has led to significant changes in the food web, with potential implications for nutrient turnover.

2.5.1 Physiological responses of Nile Tilapia to PAHs

The physiological responses of Nile tilapia to polycyclic aromatic hydrocarbons (PAHs) have been extensively studied. Pathiratne (2010) found that PAHs with four aromatic rings, such as pyrene and chrysene, induced hepatic EROD activity, while those with two to three rings, like naphthalene and phenanthrene, had no effect or inhibited EROD activity. Cheevaporn (2010) further demonstrated that exposure to PAHs increased EROD activity and bile fluorescence intensity in Nile tilapia. Liang (2007) identified the presence of PAHs in the sediments and fish of Mai Po Marshes Nature Reserve, Hong Kong, with higher levels in larger fish and in viscera compared to muscle. Sogbanmu (2018) investigated the toxicity of sublethal concentrations of naphthalene, phenanthrene, and pyrene in Clarias gariepinus, finding sex-specific responses and histological alterations in the gills, liver, and ovary.

2.5.2 PAH Exposure As It Affects Physiological functions In Nile Tilapia

Exposure to polycyclic aromatic hydrocarbons (PAHs) can significantly impact the physiological functions of Nile tilapia. Lemos (2018) found that exposure to high stocking density and acidic pH levels can lead to increased plasma cortisol and glucose levels, as well as changes in haematological variables. Similarly, Pathiratne (2010) demonstrated that certain PAHs can induce hepatic damage and modulate enzyme activities in the fish. Mohamed (2021) further highlighted the effects of salinity stress on the physiological and ion changes in Nile tilapia, including alterations in blood parameters and stress hormone levels. Lastly, Abdel-Tawwab (2012) showed that acute exposure to commercial petroleum fuels, which often contain PAHs, can reduce growth performance and alter physiological variables in the fish. Studies showing that PAH exposure can have a range of negative effects on the physiological functions of Nile tilapia.

2.5.3 Biochemical And Metabolic Changes.

The biochemical and metabolic changes of PAHs in Nile tilapia are complex and multifaceted. Pathiratne (2010) found that certain PAHs, such as pyrene and chrysene, can induce hepatic EROD activity, while others, like fluoranthene, can inhibit it. These changes can be used as biomarkers for PAH exposure. Salinity stress, as studied by Mohamed (2021), can significantly affect the blood parameters, ion levels, and gene expression in Nile tilapia, leading to physiological changes. Osman (2012) further demonstrated that exposure to mixed pollutants, including PAHs, can lead to alterations in enzyme activities and histological lesions in the tissues of Nile tilapia. Also, Hegazi (2010) showed that chronic sublethal ammonia exposure can lead to changes in the activities of various enzymes related to ammonia detoxification in the brain of Nile tilapia.

2.6 Behavioral And Reproductive Impacts Of PAHs In Nile Tilapia.

Exposure to palm oil mill effluent (POME) has been found to impact the reproductive performance and gonad histopathology of female Nile tilapia, leading to a decline in gonadosomatic index and oocyte diameter (Zulfahmi, 2018). Similarly, exposure to di(2-ethylhexyl)phthalate (DEHP) has been shown to disturb the expression of genes associated with the reproductive system in Nile tilapia (Zhang, 2018).

2.6.1 Ecological Impact of PAHs On Nile Tilapia.

On the other hand, exposure to the steroidal pre-ovulatory pheromone 17,20βP has been found to improve the seminal characteristics of male Nile tilapia, enhancing sperm quality and motility. Lastly, sublethal exposure to chlorpyrifos, an insecticide, has been found to induce visible behavioral changes in Nile tilapia, including irregular body movements and mucus secretion (Ihsan, 2018). Di-(2-ethylhexyl) phthalate (DEHP) is currently the most frequently detected phthalic acid esters (PAEs) compounds and can induce diverse toxicities on aquatic organisms.

Therefore, it can be concluded that exposure time and variation of concentration have an effect on behavioral changes of Nile tilapia such as gestures, operculum, anal excretion and breathing at the surface of water, so that the longer the exposure time, the more visible behavioral changes in the fish, as well as the variation of concentration, it means that the higher the concentration, the more visible changes in the physiological behavior of the fish.

Despite ubiquity of polycyclic aromatic hydrocarbons (PAHs) in the tropical environments, little information is available concerning responses of tropical fish to PAHs and associated toxicity. Exposure to polycyclic aromatic hydrocarbons (PAHs) can have significant ecological implications for Nile tilapia populations and their habitats. PAHs can induce hepatic damage and modulate enzyme activities in Nile tilapia, affecting their ability to metabolize these compounds.

High levels of PAHs have been found in the River Nile, indicating potential risks to both the ecosystem and human health (Omar 2017). The introduction of Nile tilapia into aquatic systems can lead to increased water turbidity, nutrient levels, and phytoplankton growth, as well as decreased benthic algal growth (Zhang 2016). Biomarker studies have shown that high concentrations of contaminants can affect fish metabolism, with potential implications for the health of Nile tilapia populations .

2.6.2 Contaminants Pathways And Accumulation.

PAHs enter aquatic ecosystems through wet deposition and suspended particulates, with higher levels detected in larger fish due to aqueous route dominance. In Nile tilapia, these pollutants can accumulate in tissues, leading to alterations in enzyme activities and histological lesions (Osman 2012). The effects of BaP, a common PAH, on biotransformation pathways in Nile tilapia depend on the route of exposure, with barrier tissues like gills and intestine playing a significant role (Costa 2011). Light PAHs, such as naphthalene, can also accumulate in the brain tissues of fish, potentially impairing normal brain function (Domingos 2011).

The use of biomarkers has become an important tool for modern environmental assessment as they can help to predict pollutants involved in the monitoring program. Currently, the use of biomarkers for monitoring environmental quality has gained considerable interest in the assessment of river condition in many places around the world. There are many different biomarkers that occur at many different levels of organization from sub-cellular to whole-organisms. The biomarker may be the chemical itself (bioaccumulation). It may also be molecular biomarkers, biochemical biomarkers and tissues biomarkers.

2.7 Toxicological Assessments

The role of dose-response in human risk assessment is crucial, with new technologies providing unprecedented capabilities to explore this relationship.

Toxicological data provide the basis for evaluating the potential health risks of chemicals to humans.

The use of toxicogenomic approaches, such as gene expression technology and proteomics, is providing new tools to predict potential toxicity of chemicals and drugs. These studies highlight the importance of understanding dose-response relationships in toxicology and the potential health impacts of chemical exposure.

Research in Nigeria has consistently found high levels of Polycyclic Aromatic Hydrocarbons (PAHs) in aquatic environments, particularly in the Niger Delta region (Ofori, 2021). These levels often exceed permissible limits, posing significant health risks to both humans and aquatic organisms. The distribution of PAHs in sediments varies across different aquatic ecosystems, with factors such as sediment characteristics influencing their presence (Inam, 2018). Despite these findings, there is a lack of specific regulations addressing PAH levels in Nigerian aquatic environments.

The presence of Polycyclic Aromatic Hydrocarbons (PAHs) in the Ogun River, Nigeria, has been a cause for concern due to its potential health risks. Studies in other Nigerian water bodies, such as the Ovia River, Lagos Lagoon, and Oburun Lake, have found varying levels of PAHs, with some exceeding recommended guidelines (Tongo 2017, Davies 2018).

The Ogun River in Nigeria is contaminated with Polycyclic Aromatic Hydrocarbons (PAHs), with levels exceeding the guideline value for drinking water (Tongo, 2017). The dominant PAHs in the river are naphthalene, acenaphthylene, and fluoranthene, with a mixed origin of pyrogenic and petrogenic sources (Tongo, 2017). Despite the low risk of exposure to PAHs through dietary and non-dietary sources, there is a potential non-carcinogenic health risk from direct ingestion of sediment (Tongo, 2017). The presence of both lower and higher molecular PAHs in the river indicates organic pollution from various sources (Davies, 2018). Continuous monitoring of PAH levels in the Ogun River is crucial to prevent future human health effects (Tongo, 2017).

PAHs, a common environmental pollutant, pose a significant threat to Nile tilapia and their habitats. Polycyclic aromatic hydrocarbons (PAHs) are derived from both natural and anthropogenic sources and are released from a wide range of industries and everyday activities. Unlike many other organic chemical contaminants that are manufactured and regulated, PAHs continue to be released on a global scale because of the world's dependence on fossil fuels.

Omar (2017) underscores the importance of regular monitoring and mitigation efforts in heavily impacted areas.Low egg production per spawning and lack of spawning synchrony are the major problems of mass seed production in mouthbrooding tilapias. Collection of eggs or fry from the mouths of incubating females reared in large hapas suspended in fertilized ponds and incubating them artificially has been found to be commercially viable. However, fouling of the hapa is a major problem causing inconsistent performance of broodfish. In addition, other factors influencing with the tilapia seed output in hapa within pond systems are age and the size of the broodfish, feeding and feed management, environmental factors and management techniques. Behera (2018) suggests the use of biosensors and bioremediation strategies for efficient detection and management of PAHs in inland aquatic ecosystems. These strategies could be explored further for their potential application in mitigating the impact of PAHs on Nile tilapia and their habitats.

The Consumption of fish contaminated with polycyclic aromatic hydrocarbons (PAHs) can pose significant health risks to humans. Jafarabadi (2020) found high levels of PAHs in coral-reef fish from the Persian Gulf, leading to a considerable risk of PAH intake through fish consumption.

Dietary intake has been reported as an important route for human exposure to PAHs, except for smokers and occupationally exposed populations [6,7]. Pollution by persistent chemicals is potentially harmful to the organisms at higher tropic levels in the food chain. The marine organisms like fish are able to accumulate severalfold higher concentration of PAHs than the surrounding water. Fish is a major source of proteins and healthy lipids for people. In particular, the long-chain omega-3 fatty acids have been shown to have numerous beneficial roles in the human health. Despite the human benefits of a fish diet, an issue of concern related to frequent fish consumption is the potential risk arising from exposure to toxic chemicals. In a recent year, a number of epidemiologic studies have reported that a large portion of human cancers, such as lung and prostate cancers, are attributable to dietary sources. Certain groups of population may have higher risks from dietary exposure of PAHs than the general populations.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Statement of Research

The study was carried out between September-October 2023 for wet month and January-February 2024 for dry month. Sampling was done within the first week of the aforementioned months. Three sampling stations were established within the creeks of Ogun river at Ajegunle, Lagos State of exact positions which are: Station 1(Latitude 6.30°N and Longitude 3.23°E), Station 2(Latitude 6.27°N and Longitude 3.19°E) and Station 3(Latitude 6.45°N and Longitude 3.33°E) in order to determine their Polycyclic Aromatic Hydrocarbon content. The exact positions of the stations were determined using a Smart GPS Location Model.

3.2 Description of Study Area

The study was conducted from Ogun River, at Ajegunle, Lagos State, which lies between Latitude 6.60°N and Longitude 3.42°E. The city is located within the mangroove ecological zone of the South-West Nigeria. The mean annual rainfall in Lagos State is approximately 1216.83mm, with a long wet season from February/March to November and a short dry season from December to January/February (Adejuwon, 2012).

Figure 2 : Map Of Study Area
Source : Google map 2024

Plate 1: Pictures of some parts of the Ogun river at Ajegunle.

Plate 2: *Oreochromis niloticus* (Nile Tilapia)

3.3 Experimental design

The study is a factorial experiment within a Completely Randomized Design (CRD), with three (3) stations × four (4) months × one (1) specific fish species (*Oreochromis niloticus*) × four (4) PAHs replicated thrice.

3.4 Sample Collection

Samples of *O.niloticus* were randomly collected from three station which was earlier mentioned in Ajegunle, Lagos State Nigeria. From personal survey, these stations were visibly loaded with *Eichornia crassipes* (water hyacinth) due to nutrient enrichment from increased anthropogenic activities which causes algal blooms and the proliferation of water hyacinth and other aquatic weeds as seen above in plate 1. These samples were caught with the help of skilled artisanal fishermen in the study area and were conveyed in an ice packed cooler to prevent PAH degradation (Wang *et al*., 2023) and taken to the laboratory within 24 hours for further analysis and study.

3.5 Conditioning of Instruments

Hewlett Packard (HP) 5980 series ‖ Gas chromatograph was equipped with an Agilent 7683B injector (Agilent Technologies, Santa Clara, CA, USA) 0.30m, 0.25mm ID. HP-5MS capillary column (Hewlett Packard Palo Alto, CA, USA) coated with phenyl-methylsiloxane (film thickness 0.25µm) and an Agilent 5975 flame ionization detector (FID) was used to separate and quantify the PAHs compounds.

3.6 Treatment and Extraction of Sample for Analysis of PAHs

The fish samples were milled using an electric blender. Extraction of PAHs was then carried out based on the method described by Pena *et al*., (2006). Ten (10g) of homogeneously grinded fish sample was weighed using an electric scale balance, and the solid sample was placed in an extraction thimble which was thoroughly mixed with anhydrous Na2So4 to dehydrate the sample. 20ml of the extraction solvent (di-chloromethane) was added to the sample. Samples was covered with aluminium foil to prevent evaporation and sonicated to separate supernatants of extracts. Extracts was concentrated using an evaporator and was cleaned up using a chromatographic column, moderately packed at the bottom with 1cm glass wool. 2g of silica gel and 1cm of anhydrous Na2So4 was added to the column while the column was pre-eluted with 20ml dichloromethane. After subsequent drying over anhydrous sodium sulphate, and concentrated to 1.0ml using a rotary evaporator, an internal standard mixture (naphthalene-d8, acenaphtene-d10, phenathrene-d10, crysene-d12, and perylene-d12) solution was then added to the extract analyzed, using Hewlett Packard HP 5890 series Ⅱ Gas chromatograph with flame ionization detector (GC-FID). Extracts was concentrated and collected in 2ml vials.

3.7 Chromatographic Analysis

The cleaned up extracts was analyzed and corresponding results was obtained using Gas chromatography (GC, Hewlett-Packard HP-5890 Series Ⅱ with flame ionization detection (GC-FID). The GC will be programmed as follows: initial temperature of 60°C for 2mins and will be ramped at 25°C/min to 300°C for 5mins and allowed to stay for 15mins giving a total of run time of 22mins. A 2L volume split-less injection mode was used and the injection port temperature will be set at 2500C, while 3000C was maintained for the injection port of the FID detector. A standard mixture of 4 priority PAHs (Benzo(a)pyrene, Benzo(a)anthracene, Benzo(b)fluroanthene and Napthalene) were obtained and used for the analysis. Compounds identified were then compared by the retention time of standards with that obtained from the extracts and individual analysis of PAHs was identified for quantification. All PAHs value were expressed in µg/kg.

3.8 Calculation of Toxic Quotient (TQ) for PAHs

The Toxic Quotient (TQ) expresses the possibility of a contaminant being an ecological risk or a contaminant of potential ecological concern.

$$TQ = \frac{\text{Measured concentration of PAH}}{\text{Toxicity reference value or selected screening benchmark}}$$

• TQ < 1: Indicates no potential for adverse effects.

• 1 ≤ TQ ≤ 5: Suggests potential for low-level effects, warranting further investigation.

• TQ > 5: Raises concern for potential adverse effects, requiring more immediate action.

3.9 Calculation of Toxic Equivalency (TEQ) for PAHs

Toxic equivqlency factors are toxicity potency factors used as a consistent method to evaluate the toxicity of variable mixtures of organic compounds. These factors are based on health effects assessed in laboratory animals and wildlife by an intake route such as ingestion through diet. According to the United States Environmental Protection Agency (USEPA), the toxic equivalency estimates the individual PAH potencies relative to that of benzo(a)pyrene, in order to obtain a benzo(a)pyrene equivalent (USEPA,1992).

$$TEQ = \sum Ti \times TEF$$

Where: TEQ = Toxic Equivalency

Ti = PAH concentration in fish

TEF = Toxic Equivalency Factor

Compounds having TEF value of zero are not important in calculating TEQ. Benzo(a)pyrene has a TEF value of 1 and it serves as an index PAH for the other PAH compound.

Benzo(a)anthracene and Benzo(b)flouranthene have TEF value of 0.1.

3.10 Estimated Annual Intake (EAI) and Estimated Daily Intake (EDI) Of PAHs

$$EAI\ (mg/person/year) = \frac{\text{Concentration of PAH in fish} \times \text{per capital figure}}{\text{Adult body weight (Assumed to be 70kg)}}$$

Where Per capita figure is 13.3kg/person/year for Nigeria (World Fish Center, 2021).

$$EDI\ (mg/person/year) = \frac{EAI}{365\ \text{days}}$$

3.11 Calculation of cancer risk factor

The general equation for estimating human exposure to cancer through the consumption of fish is stated below (USEPA, 1989).

$ECR = EI \times ED \times CSF/BW \times AT$

Where, ECR = Excess Cancer Risk; EI = Estimated Intake; ED = Exposure Duration (30 years for adults); CSF = Oral Cancer Slope Factor; BW = Body Weight (assuming 60kg weight); AT = Average Time for carcinogens (70 years for adults). Cancer Risk Guideline Value = 1.0×10^{-6}

3.12 Statistical Analysis

A GENSTAT® computer software (Version 12.1 for Windows) was used for statistical analysis. Data generated from the study was subjected to one way Analysis of Variance (ANOVA) to determine significant differences between mean values of PAHs at 5% level of significance. Significant means was separated using Duncan Multiple Range Test while Microsoft Excel (for Windows 2010), was used for all graphical representation.

CHAPTER FOUR

4.0 RESULTS

As shown in Table 1, the mean concentration of PAHs congeners in *O. niloticus* ranged from 0.0059 µg/kg in Naphthalene to 11.789 µg/kg in Fluoranthene.

Table 1: Summary Statistics for PAHs congeners (µg/kg) in *Oreochromis niloticus* from Ogun River.

PAHs	Mean	Minimum	Maximum	Threshold (µg/kg)
Benzo(a)pyrene	3.737	0.001	11.46	
Anthracene	0.405	0.001	0.698	
Fluoroanthene	11.789	0.001	38.5	2.0
Naphthalene	0.0059	0.001	0.01	
Total	15.9369	0.004	50.668	

* Commission Regulation (2008)

4.1 PAHs congeners (µg/kg) in *Oreochromis niloticus* from Ogun River.

As shown in Table 1, the mean concentration of PAHs congeners in *O. niloticus* ranged from 0.0059 µg/kg in Naphthalene to 11.789 µg/kg in Fluoranthene. There were significant differences ($P < 0.05$) in the mean concentrations of all PAHs in the fish.

Table 2: Summary statistics for PAHs congeners (µg/kg) in *Oreochromis niloticus* by months.

PAHs	September	October	January	February
Benzo(a)pyrene	0.04±0.05a	0.04±0.05a	0.04±0.05a	0.04±0.05a
Anthracene	0.43±0.32a	0.43±0.32a	0.38±0.29a	0.38±0.29a
Fluoroanthene	0.13±0.18a	0.13±0.18a	0.11±0.17a	0.11±0.16a
Naphthalene	0.01±0.00a	0.01±0.00a	0.00±0.00b	0.00±0.00b
Total PAHs conc. µg/kg	0.61±0.37a	0.61±0.38a	0.54±0.33a	0.54±0.32a

Values are expressed as mean ± Standard deviation. Means with different superscripts are statistically significant at $p < 0.05$

4.2 PAHs congeners (µg/kg) in *Oreochromis niloticus* by months

As shown in Table 2, the mean concentration of PAHs congeners in *O. niloticus* ranged from 0.01 ± 0.00 (SD) µg/ul in Naphthalene to 0.43 ± 0.32 (SD) µg/kg in Anthracene by the month of September, 2023. The mean concentration of PAHs congeners also ranged from 0.01 ± 0.00 (SD) µg/kg in Naphthalene to 0.43 ± 0.32 (SD) µg/kg in Anthracene by the month of October, 2023. The mean concentration of PAHs congeners also ranged from 0.00 ± 0.00 (SD) µg/kg in Naphthalene to 0.38 ± 0.29 (SD) µg/kg in Anthracene by the month of January, 2024. The mean concentration of PAHs congeners also ranged from 0.00 ± 0.00 (SD) µg/kg in Naphthalene to 0.38 ± 0.29 (SD) µg/kg in Anthracene by the month of February, 2024. There were no significant differences between (P>0.05) in the mean concentrations of PAHs congeners in the fish by months.

Table 3: Summary statistics for PAHs congeners (µg/kg) in *Oreochromis niloticus* by Stations

PAHs	Upstream	Midstream	Downstream
Benzo(a)pyrene	0.04±0.05a	0.04±0.05a	0.04±0.05a
Anthracene	0.40±0.30a	0.41±0.30a	0.41±0.30a
Fluoroanthene	0.12±0.17a	0.12±0.17a	0.12±0.17a
Naphthalene	0.01±0.00a	0.01±0.00a	0.01±0.00a
Total PAHs conc. µg/kg	0.57±0.34a	0.58±0.35a	0.58±0.35a

Values are expressed as mean ± Standard deviation. Means with different superscripts are statistically significant at $p < 0.05$

4.3 PAHs congeners (µg/kg) in *Oreochromis niloticus* by Stations

As shown in Table 3, the mean concentration of PAHs congeners in *O. niloticus* ranged from 0.01 ± 0.00 (SD) µg/kg in Naphthalene to 0.40 ± 0.30 (SD) µg/kg in Anthracene in the Upstream Station. The mean concentration of PAHs congeners also ranged from 0.01 ± 0.00 (SD) µg/kg in Naphthalene to 0.41 ± 0.30 (SD) µg/kg in Anthracene in the Midstream Station. The mean concentration of PAHs congeners also ranged from 0.01 ± 0.00 (SD) µg/kg in Naphthalene to 0.41 ± 0.30 (SD) µg/kg in Anthracene in the Downstream Station. There were no significant differences between ($P>0.05$) in the mean concentrations of PAHs congeners in the fish by station.

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Table 4: Calculated cancer risk factors for polycyclic aromatic hydrocarbons

PAHs	EI	ED	CSF (USEPA values)	BW	AT	ECR
Benzo(a)pyrene	0.00011231	30	NA	60	70	NA
Anthracene	0.000001215	30	NA	60	70	NA
Fluoroanthene	0.000035467	30	NA	60	70	NA
Naphthalene	0.0000000177	30	NA	60	70	NA

USEPA cancer risk guidelines value = 1.0 x 10-4   NA = Not Available

4.4 Calculation of Toxic Quotient (TQ) for PAHs

The TQ values of *O. niloticus* ranged from 0.000059 for Naphthalene to 3.737 for Benzo(a)pyrene as shown in Fig. 2.

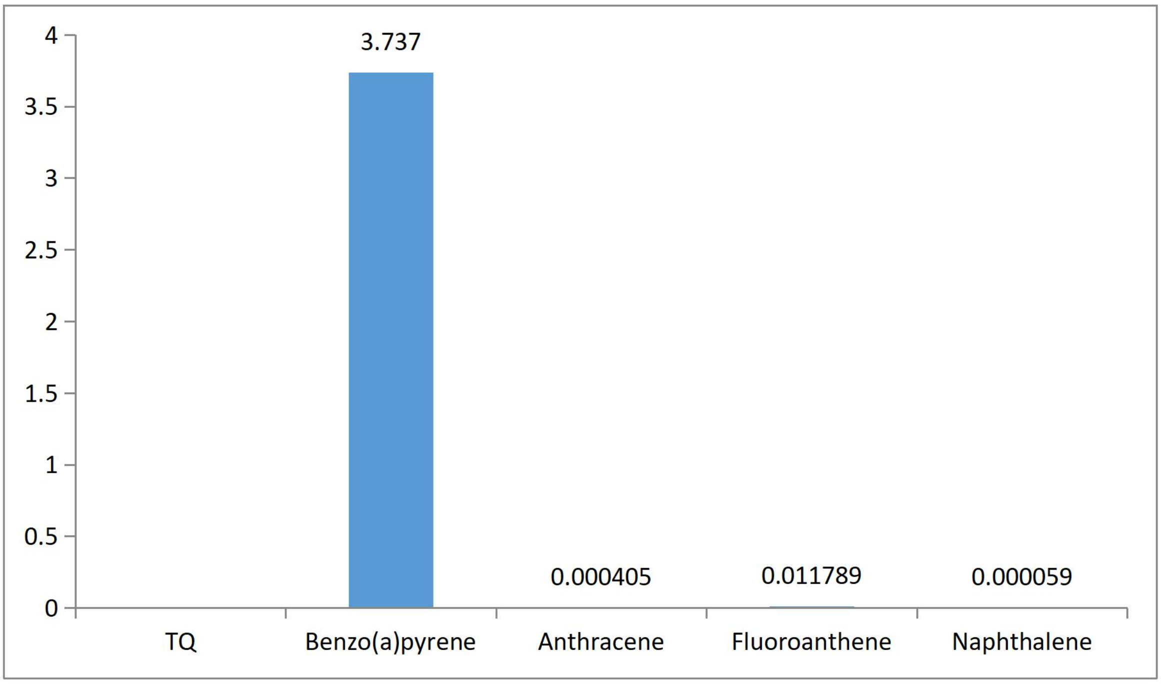

Figure 3: Toxic quotient (TQ) values for PAHs in fish species (*O. niloticus*)

4.5 Calculation of Toxic Equivalency (TEQ) for PAHs

The TQ values of *O. niloticus* ranged from 0.0059 for Naphthalene to 11.789 for Fluoranthene as shown in Fig. 3.

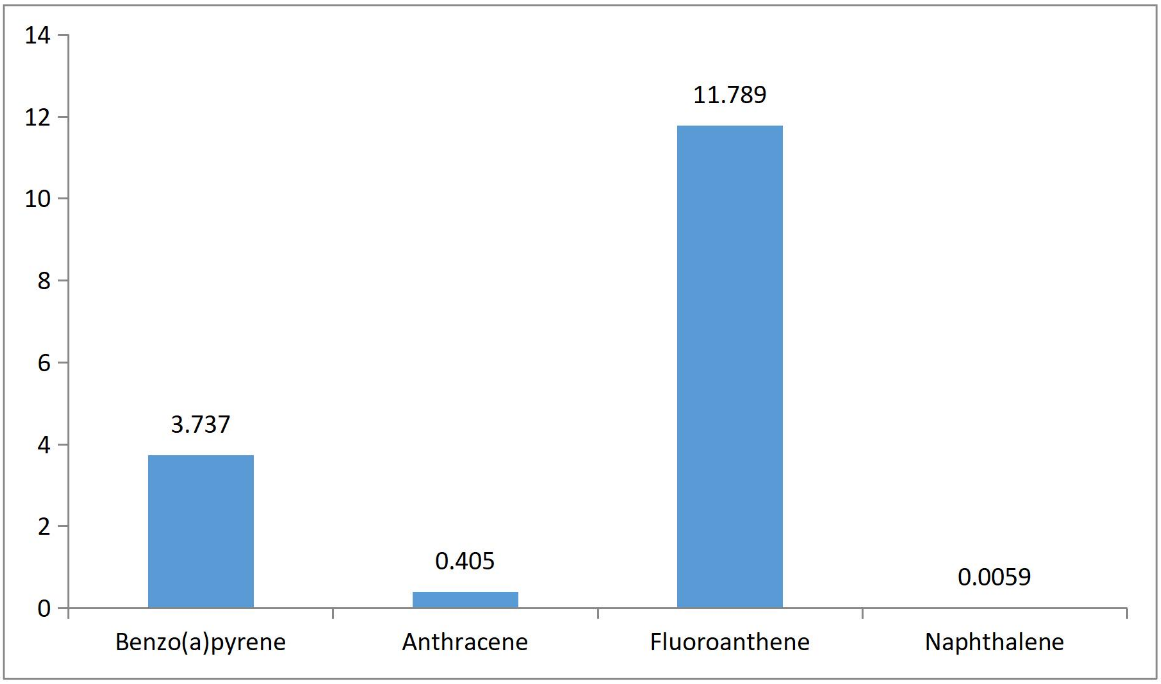

Figure 4: Toxic Equivalency (TEQ) values for PAHs in fish species (*O. niloticus*)

4.6 Estimated Daily Intake (EDI) values for PAHs

The EDI values of *O. niloticus* ranged from 0.00000307 for Naphthalene to 0.00613 for Fluoranthene as shown in Fig. 4.

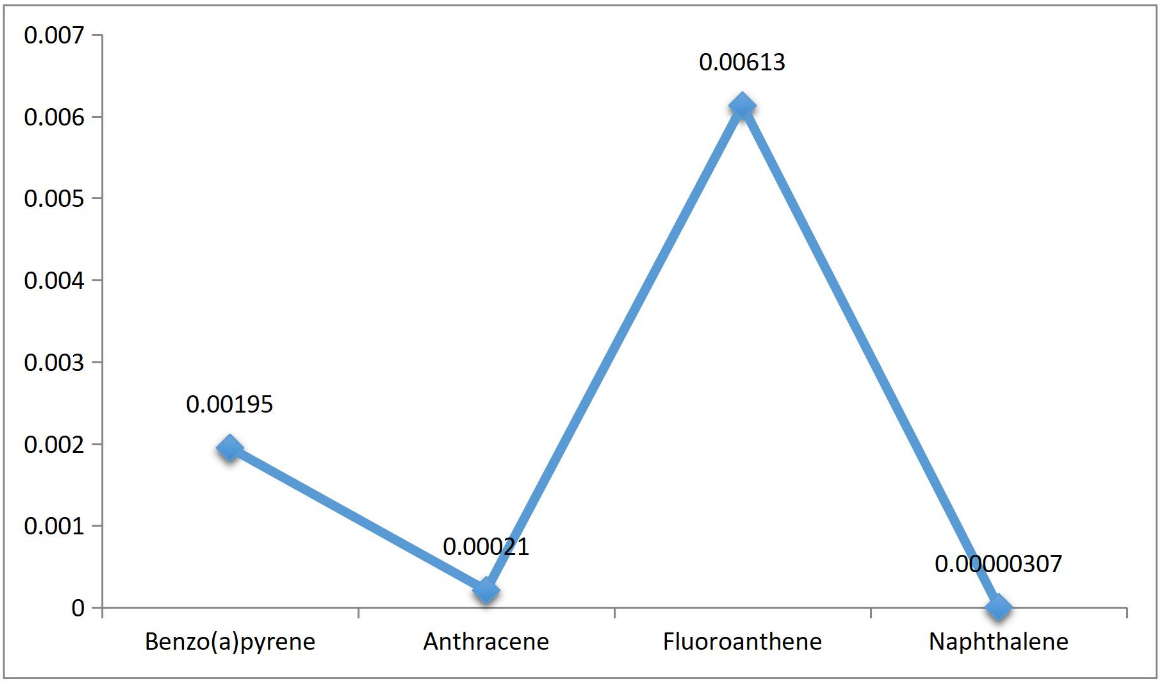

Figure 5: EDI Values (mg/person/day) for PAHs in *O. niloticus* from Ogun River

4.7 Estimated Annual Intake (EAI) values for PAHs

The EDI values of *O. niloticus* ranged from 0.001121 for Naphthalene to 2.28991 for Fluoranthene as shown in Fig. 5.

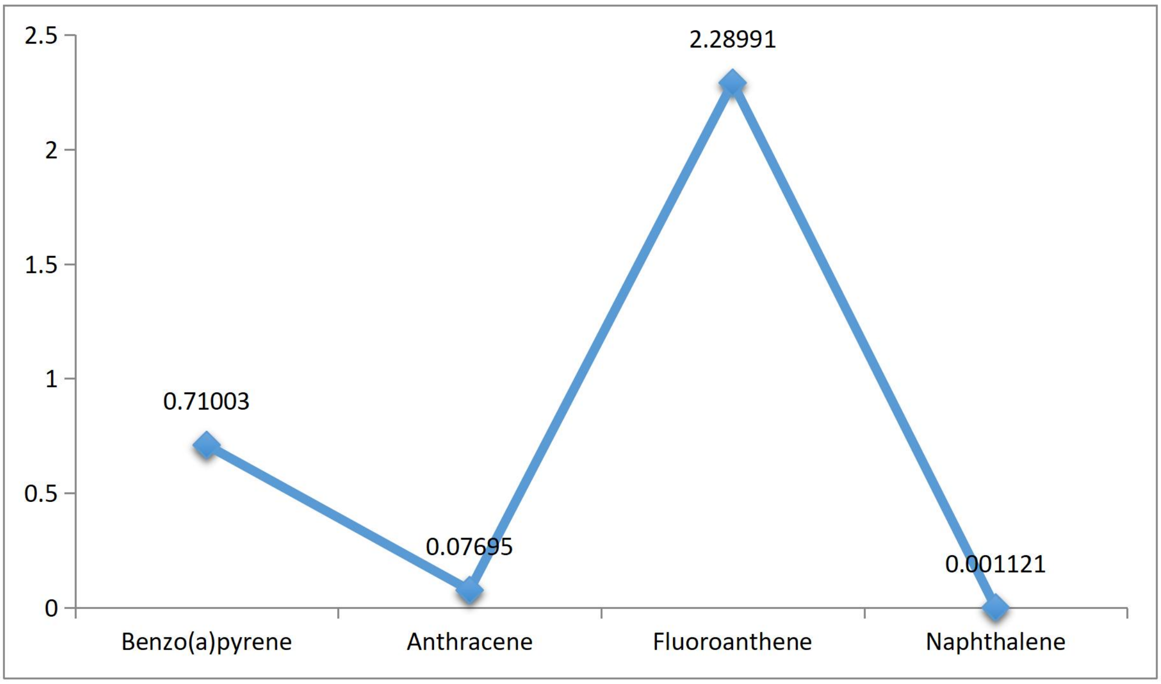

Figure 5: EDI Values (mg/person/day) for PAHs in *O. niloticus from* Ogun River

4.8 Quota (%) for PAHs in *O. niloticus* from Ogun River

The quota for PAHs in *O. niloticus* ranged from 0.037% for Naphthalene to 73.97% for Fluoranthene as shown in Fig. 7 below.

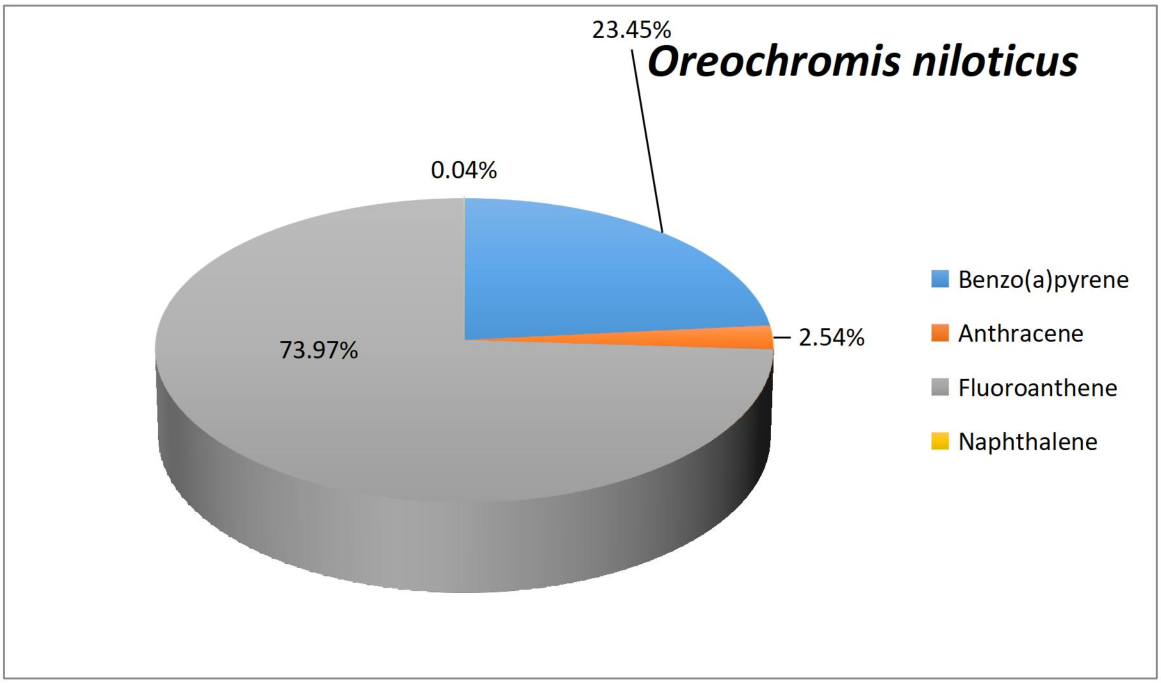

Figure 6: Quota of PAHs in *O. niloticus* from Ogun River

4.3.7 Total PAHs content in *O. niloticus* by river stations

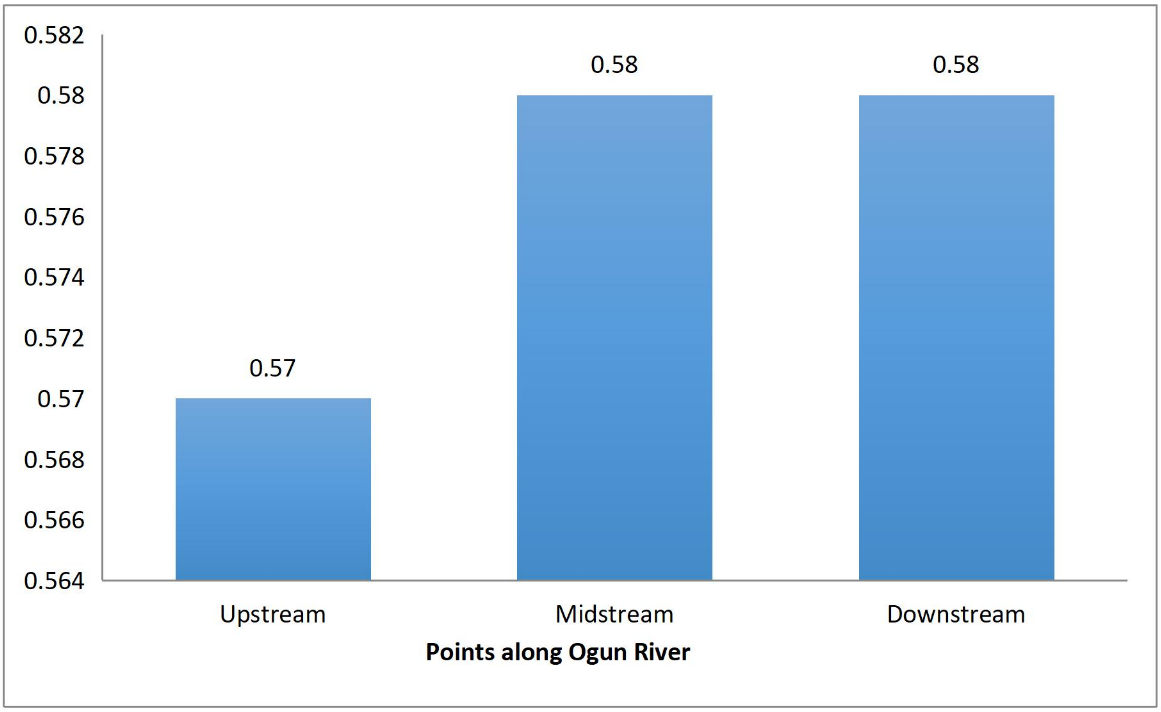

Figure 7: Total PAHs content in *O. niloticus* by river stations

CHAPTER FIVE

5.0 DISCUSSION

The laboratory findings of this study demonstrate that the Ogun river Nile Tilapia (*O. niloticus*) possesses four congeners with an apparent PAHs rank profile sequence of Naphthalene > Fluoranthene > Anthracene > Benzo(a)pyrene.

Polycyclic Aromatic Hydrocarbons are typically components of complex mixtures containing carbon and hydrogen atoms arranged in a minimum of two fused aromatic ring structures (CCME, 2010). Humans commonly encounter PAHs through direct or indirect pathways. PAHs exhibit a high affinity for lipids, volatility, and persistence, rendering them readily bioavailable (Palm *et al*., 2011). However, exposure to PAHs can stem from diverse sources, including consuming river fish (Ayandiran *et al*., 2022). PAH concentrations in river fish vary based on water and sediment PAH levels and the fish's feeding habits. PAHs present in river water and sediments are bioaccumulated by fish and other aquatic organisms, potentially leading to biomagnification of these compounds. PAHs have demonstrated carcinogenic, mutagenic, and teratogenic effects on aquatic animals and humans relying on river water and fish for sustenance, particularly those with occupational exposure (Ekere *et al*., 2019).

Numerous factors contribute to elevated PAH levels in river fish, including fossil fuel and biomass combustion, soil contamination, industrial and municipal waste, atmospheric deposition, agricultural practices, and natural sources. Incomplete combustion of fossil fuels and biomass like petroleum products, coal, and wood releases PAHs into the atmosphere, which may subsequently deposit into rivers and other water bodies (Chizhova *et al*., 2020). Soil pollution, often caused by the combustion of petroleum products, can also contribute to riverine PAH levels as soil-borne PAHs may be washed into rivers during rainfall or flooding. Industrial and municipal waste, such as coal-tar pitch, creosote, and asphalt, may contain substantial PAH concentrations and can contaminate river water and sediments upon release. Atmospheric deposition is another source of PAHs in rivers, as these compounds can be transported through the air and settled into water bodies. Agricultural practices, including the application of specific pesticides and herbicides, can also contribute to PAH levels in rivers. Natural processes such as volcanic eruptions and forest fires can produce PAHs naturally. PAHs persist in the environment and can accumulate in sediments and aquatic organisms, posing potential risks to aquatic ecosystems and human health. This is consistent with the findings of Fatima *et al*., (2022), who examined the distribution of polycyclic aromatic hydrocarbons in water and sediment, highlighting the critical role of PAH exchange between water and sediment in determining their environmental fate and potential impacts on aquatic biota. This study indicates higher total PAH concentrations in the river's midstream and downstream sections compared to other locations, which may be attributed to increased exposure to pollutants upstream. Fluoranthene and Benzo(a)pyrene were found to be the most prevalent PAH congeners.

Fluoranthene, a PAH congener, poses various health concerns, particularly its potential to induce cancer and, in conjunction with other PAHs, hemolytic anemia, liver damage, and neurological impairment in infants. Inhalation or ingestion of fluoranthene can result in acute effects such as headaches, nausea, vomiting, diarrhea, malaise, anemia, jaundice, seizures, and coma. In this study, fluoranthene concentrations were highest across various months and sampling locations along the river and exhibited the highest toxic equivalency and quota in *O. niloticus* (73.97%). Therefore, the consumption of fish contaminated with fluoranthene may pose a health risk.

Benzo(a)pyrene (BaP) is a PAH known for its carcinogenic and toxic effects. It is generated during the incomplete combustion of organic materials, including fossil fuels, tobacco smoke, and charred foods (Ba *et al*., 2015). BaP exposure can occur through inhalation, ingestion, or dermal contact and is transported to various organs by blood and lymph following absorption. The cytochrome P450-dependent monooxygenase system metabolizes BaP, converting it into reactive, toxic metabolites that covalently bind to cellular components like DNA. This metabolic activation can produce reactive oxygen species, causing damage to cellular macromolecules (Ba *et al*., 2015). In humans, BaP exposure has been linked to reproductive and immune system dysfunctions, including decreased sperm count, ovarian weight, and follicle numbers, as well as decreased immunoglobulin and B cell numbers and thymus weight. Research also suggests that BaP exposure can disrupt cardiovascular development, elevate systolic blood pressure, and potentially cause adverse birth outcomes, neurobehavioral effects, and reduced fertility (Chenghao *et al*. 2022). Furthermore, BaP has been found to impair the migration pattern of human gonadotropin-releasing-hormone-secreting neuroblasts and induce genome-wide mutations in mouse offspring (Traini *et al*., 2023). This study revealed that Benzo(a)pyrene had the highest toxic quotient value (3.737) and the highest cancer risk factor (0.00011231), indicating a potential health concern regarding its consumption in fish.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATION

6.1 Conclusion

Polycyclic aromatic hydrocarbons (PAHs), an array of chemical substances, are prevalent in environmental mixtures. These compounds pose significant health risks as carcinogens and mutagens, particularly when concentrated in marine life. Limited information regarding PAH levels in *Oreochromis niloticus* from Ogun river prompted this investigation. The evaluation of PAH content in these fish species highlights potential health hazards for consumers and underscores the imperative to limit prolonged exposure to carcinogenic effects.

The study recognized Fluoranthene and Benzo(a)pyrene as the primary PAH congeners in *O. niloticus* obtained from the Ogun river in Ajegungle, Lagos State, Nigeria, emphasizing the need for comprehensive data on PAH levels for public awareness. Excessive consumption of these pollutants elevates cancer risks.

To address these concerns, it is essential to educate local communities about minimizing pollutant discharge into the river, fostering a healthier environment for both fish and consumers. Collaborative efforts between health authorities and organizations like the World Health Organization (WHO) are crucial for implementing awareness campaigns and mitigating the risks associated with long-term consumption of PAH-contaminated *O. niloticus* sourced from rivers.

6.2 Recommendations

Based on the outcomes of this study, the following recommendations are made;

1. It is suggested that PAHs not contained within the scope of this study be integrated into future research to expand the PAH baseline for the area.

2. Investigations should be carried out to ascertain the correlation amidst PAHs contamination, its dissemination throughout freshwater ecosystems, and its noxiousness towards bottom-dwelling fauna

3. Public enlightenment with respect to health implications of exposure to PAHs should be encouraged.

REFERENCES

Abdel-Shafy, H. I., and Mansour, M. S. (2016). A review on polycyclic aromatic hydrocarbons: Source, environmental impact, effect on human health and remediation. *Egyptian Journal of Petroleum*, 25(1), 107-123.

Ayandiran, T. A., Fawole, O. O., & Ogundiran, M. A. (2022). Polycyclic aromatic hydrocarbon concentrations in Clarias gariepinus from Oluwa River, Ondo State, Nigeria. *Research Journal of Environmental Toxicology, 16*, 1-11.

Ba, Q., Li, J., Huang, C., Qiu, H., Li, J., Chu, R., Zhang, W., Xie, D., Wu, Y., and Wang, H. (2014). Effects of benzo[a]pyrene exposure on human hepatocellular carcinoma cell angiogenesis, metastasis, and NF-κB signaling. Environmental Health Perspectives, 123(3), 246-254.

Balbo, S., Hecht, S. S., Upadhyaya, P., and Villalta, P. W. (2014). Application of a high-resolution mass-spectrometry-based DNA adductomics approach for identification of DNA adducts in complex mixtures. Analytical Chemistry, 86(3), 1744-1752.

Barakat, A. O., Mostafa, A., Wade, T. L., Sweet, S. T., and El Sayed, N. B. (2011). Distribution and characteristics of PAHs in sediments from the Mediterranean coastal environment of Egypt. Marine Pollution Bulletin, 62(9), 1969-1978.

Błaszczyk, E., Rogula-Kozłowska, W., Klejnowski, K., Fulara, I., and Mielżyńska-Švach, D. (2017). Polycyclic aromatic hydrocarbons bound to outdoor and indoor airborne particles (PM2.5) and their mutagenicity and carcinogenicity in Silesian kindergartens, Poland. Air Quality, Atmosphere & Health, 10, 389-400.

Burchiel, S. W., and Gao, J. (2014). Polycyclic aromatic hydrocarbons and the immune system. Encyclopedia Immunotoxicology.

Cheevaporn, V., Pindang, M., and Helander, H. F. (2010). Polycyclic aromatic hydrocarbon contamination in Nile Tilapia (*Oreochromis niloticus*): Analysis in liver and bile. Environment Asia, 3(2), 8-14.

Chen, F., Tan, M., Ma, J., Zhang, S., Li, G., and Qu, J. (2016). Efficient remediation of PAH-metal co-contaminated soil using microbial-plant combination: A greenhouse study. *Journal of Hazardous Materials, 302*, 250-261.

Chizhova, T., Koudryashova, Y., Prokuda, N., Tishchenko, P., and Hayakawa, K. (2020). Polycyclic aromatic hydrocarbons in the estuaries of two rivers of the Sea of Japan. *International Journal of Environmental Research and Public Health, 17*(17), 6019.

Commision Regulation (2008). Setting maximum levels for certain contaminants in foodstuffs. Official Journal of the European Union. 20pp

Costa, P. M., Neuparth, T. S., Caeiro, S., Lobo, J., Martins, M., Ferreira, A. M., ... and Costa, M. H. (2011). Assessment of the genotoxic potential of contaminated estuarine sediments in fish peripheral blood: Laboratory versus in situ studies. Environmental Research, 111(1), 25-36.

Ekere, N. R., Yakubu, N. M., Oparanozie, T., and Ihedioha, J. N. (2019). Levels and risk assessment of polycyclic aromatic hydrocarbons in water and fish of Rivers Niger and Benue confluence Lokoja, Nigeria. *Journal of Environmental Health Science and Engineering, 17(*1), 383-392.

El-Maradny, A., Orif, M., AlKobati, A., *et al*. (2023). Polycyclic aromatic hydrocarbons in the sediments of highly polluted coastal area in the Red Sea: levels, spatial distribution, and risk assessment. Environmental Monitoring and Assessment, 195, 1547. https://doi.org/10.1007/s10661-023-12157-x

Fátima, J., Pereira, J. L., Campos, I., Martha, S., Ana, R., Jacob, K., António, N., Fernando, J. M. Gonçalves, Nelson, A., and Dalila, S. (2022). A review on polycyclic aromatic hydrocarbons distribution in freshwater ecosystems and their toxicity to benthic fauna. Science of The Total Environment, 820.

Fu, C., Li, Y., Xi, H., Niu, Z., Chen, N., Wang, R., Yan, Y., Gan, X., Wang, M., Zhang, W., and Zhang, Y. (2022). Benzo(a)pyrene and cardiovascular diseases: An overview of pre-clinical studies focused on the underlying molecular mechanism. Frontiers in Nutrition, 9.

Gao, F. Y., Zhang, D., Lu, M. X., Cao, J. M., Liu, Z. G., Ke, X. L., ... and Yi, M. M. (2018). MHC class IIA polymorphisms and their association with resistance–susceptibility to Streptococcus agalactiae in Nile tilapia, *Oreochromis niloticus*. *Journal of Fish Biology, 93*(6), 1207-1215.

Gupte, A., Tripathi, A., Patel, H., Rudakiya, D., and Gupte, S. (2016). Bioremediation of polycyclic aromatic hydrocarbon (PAHs): A perspective. The Open Biotechnology Journal, 10(1).

Ihsan, T., Edwin, T., and Anggraeni, W. (2018). Behavioral responses of Nile tilapia (*Oreochromis niloticus*) by sublethal exposure to chlorpyrifos: A case study in Twin Lakes of West Sumatra. *Journal of Environmental Health Science & Engineering, 5*(4), 205-210.

Inam, E., Etuk, I., Offiong, N. A., Kim, K. W., Kang, S. Y., and Essien, J. (2018). Distribution and ecological risks of polycyclic aromatic hydrocarbons (PAHs) in sediments of different tropical water ecosystems in Niger Delta, Nigeria. Environmental Earth Sciences, 77, 1-14.

Jafarabadi, A. R., Mashjoor, S., Bakhtiari, A. R., and Jadot, C. (2020). Dietary intake of polycyclic aromatic hydrocarbons (PAHs) from coral reef fish in the Persian Gulf—Human health risk assessment

Ke, C. L., Gu, Y. G., and Liu, Q. (2017). Polycyclic aromatic hydrocarbons (PAHs) in exposed-lawn soils from 28 urban parks in the megacity Guangzhou: Occurrence, sources, and human health implications. Archives of Environmental Contamination and Toxicology, 72, 496-504.

Liang, Y., Tse, M. F., Young, L., and Wong, M. H. (2007). Distribution patterns of polycyclic aromatic hydrocarbons (PAHs) in the sediments and fish at Mai Po Marshes Nature Reserve, Hong Kong. Water Research, 41(6), 1303-1311.

Liu, B., Xue, Z., Zhu, X., and Jia, C. (2017). Long-term trends (1990–2014), health risks, and sources of atmospheric polycyclic aromatic hydrocarbons (PAHs) in the US. Environmental Pollution, 220, 1171-1179.

Mojiri, A., Zhou, J. L., Ohashi, A., Ozaki, N., and Kindaichi, T. (2019). Comprehensive review of polycyclic aromatic hydrocarbons in water sources, their effects and treatments. Science of the Total Environment, 696, 133971.

Naigaga, I., Kaiser, H., Muller, W. J., Ojok, L., Mbabazi, D., Magezi, G., and Muhumuza, E. (2011). Fish as bioindicators in aquatic environmental pollution assessment: A case study in Lake Victoria wetlands, Uganda. Physics and Chemistry of the Earth, parts A/B/C, 36(14-15), 918-928.

Nkpaa, K. W., Wegwu, M. O., and Essien, E. B. (2013). Assessment of polycyclic aromatic hydrocarbons (PAHs) levels in two commercially important fish species from crude oil polluted waters of Ogoniland and their carcinogenic health risks. Journal of Environmental Earth Science, 3(8).

Nwaichi, E. O., and Ntorgbo, S. A. (2016). Assessment of PAHs levels in some fish and seafood from different coastal waters in the Niger Delta. Toxicology Reports, 3, 167-172.

Ofori, S. A., Cobbina, S. J., Imoro, A. Z., Doke, D. A., and Gaiser, T. (2021). Polycyclic aromatic hydrocarbon (PAH) pollution and its associated human health risks in the Niger Delta Region of Nigeria: A systematic review. Environmental Processes, 8, 455-482.

Ogunbisi, M. A., Olujimi, O. O., Sojinu, O. S., Xian, Q., and Arowolo, T. A. (2023). Occurrence, source, and risk assessment of polycyclic aromatic hydrocarbons in Ogun River and Lagos Lagoon, Southwest, Nigeria. *International Journal of Environmental Science and Technology*, 20(4), 4391-4404.

Okenyi, A. D., Ubani, C. S., Oje, O. A., and Onwurah, I. N. E. (2016). Levels of polycyclic aromatic hydrocarbon (PAH) in fresh water fish dried with different drying regimes. Journal of Food Measurement and Characterization, 10, 405-410.

Omar, W. A. M., and Mahmoud, H. M. (2017). Risk assessment of polycyclic aromatic hydrocarbons (PAHs) in River Nile up-and downstream of a densely populated area. *Journal of Environmental Science and Health, Part A, 52*(2), 166-173.

Osman, A. G., Abuel-Fadl, K. Y., and Kloas, W. (2012). In situ evaluation of the genotoxic potential of the river Nile: II. Detection of DNA strand-breakage and apoptosis in *Oreochromis niloticus* niloticus

Pathiratne, A., and Hemachandra, C. K. (2010). Modulation of ethoxyresorufin O-deethylase and glutathione S-transferase activities in Nile tilapia (*Oreochromis niloticus*) by polycyclic aromatic hydrocarbons containing two to four rings: Implications in biomonitoring aquatic pollution. Ecotoxicology, 19, 1012-1018.

Pena, T., Pensado, L., Casais, C., Mejuto, C., Phan-Tan-Luu, R., and Cela, R. (2006). Optimization of a microwave-assisted extraction method for the analysis of polycyclic aromatic hydrocarbons from fish samples. *Journal of Chromatography A, 1121*(2), 163-169.

Pérez-Padilla, R., Schilmann, A., and Riojas-Rodriguez, H. (2010). Respiratory health effects of indoor air pollution. *The International Journal of Tuberculosis and Lung Disease*, 14(9), 1079-1086.

Ravindra, K., Sokhi, R., and Van Grieken, R. (2008). Atmospheric polycyclic aromatic hydrocarbons: Source attribution, emission factors and regulation. Atmospheric Environment, 42(13), 2895-2921.

Sogbanmu, T. O., Osibona, A. O., Oguntunde, O. A., and Otitoloju, A. A. (2018). Biomarkers of toxicity in Clarias gariepinus exposed to sublethal concentrations of polycyclic aromatic hydrocarbons. *African Journal of Aquatic Science, 43*(3), 281-292.

Srogi, K. (2007). Monitoring of environmental exposure to polycyclic aromatic hydrocarbons: A review. Environmental Chemistry Letters, 5, 169-195.

Tongo, I., Ogbeide, O., and Ezemonye, L. (2017). Human health risk assessment of polycyclic aromatic hydrocarbons (PAHs) in smoked fish species from markets in Southern Nigeria. Toxicology Reports, 4, 55-61.

Traini, G., Tamburrino, L., Ragosta, M. E., Guarnieri, G., Morelli, A., Vignozzi, L., Baldi, E., and Marchiani, S. (2023). Effects of benzo[a]pyrene on human sperm functions: An in vitro study. *International Journal of Molecular Sciences, 24*(19), 14411.

United States. Environmental Protection Agency. Environmental Criteria and Assessment Office (Cincinnati, 1993). Provisional guidance for quantitative risk assessment of polycyclic aromatic hydrocarbons (Vol. 600). Environmental Criteria and Assessment Office, Office of Health and Environmental Assessment, US Environmental Protection Agency.

Vijayanand, M., Ramakrishnan, A., Ramakrishnan, Dr, Issac, P., Nasr, M., Khoo, K. S., Rajagopal, R., Babett, G., Wan Azelee, N. I., Jeon, B.-H., Chang, S.-W., and Balasubramani, R. (2023). Polyaromatic hydrocarbons (PAHs) in the water environment: A review on toxicity, microbial biodegradation, systematic biological advancements, and environmental fate. Environmental Research, 227, 115716.

Wang, S., Li, C., Zhang, L., Chen, Q., and Wang, S. (2024). Assessing the ecological impacts of polycyclic aromatic hydrocarbons petroleum pollutants using a network toxicity model. Environmental Research, 245, 117901.

Wang, T., Xiang, K., Zeng, Y., Gu, H., Guan, Y., and Chen, S. (2023). Polycyclic aromatic hydrocarbons (PAHs) in air, foliage, and litter in a subtropical forest: Spatioseasonal variations, partitioning, and litter-PAH degradation. Environmental Pollution, 328, 121587.

Xia, Z., Duan, X., Qiu, W., Liu, D., Wang, B., Tao, S., ... and Hu, X. (2010). Health risk assessment on dietary exposure to polycyclic aromatic hydrocarbons (PAHs) in Taiyuan, China. Science of the Total Environment, 408(22), 5331-5337.

Yoon, E., Park, K., Lee, H., Yang, J. H., & Lee, C. (2007). Estimation of excess cancer risk on time-weighted lifetime average daily intake of PAHs from food ingestion. Human and Ecological Risk Assessment, 13(3), 669-680.

Zambianchi, M., Durso, M., Liscio, A., Treossi, E., Bettini, C., Capobianco, M. L., ... and Melucci, M. (2017). Graphene oxide doped polysulfone membrane adsorbers for the removal of organic contaminants from water. *Journal of Membrane Science, 326*, 130-140. [ISSN 1385-8947]

Zheng, H., Kang, S., Chen, P., Li, Q., Tripathee, L., Maharjan, L., ... and Santos, E. (2020). Sources and spatio-temporal distribution of aerosol polycyclic aromatic hydrocarbons throughout the Tibetan Plateau. Environmental Pollution, 261, 114-144.

Zulfahmi, I., Muliari, M., Akmal, Y., and Batubara, A. S. (2018). Reproductive performance and gonad histopathology of female Nile tilapia (*Oreochromis niloticus* Linnaeus 1758) exposed to palm oil mill effluent. *The Egyptian Journal of Aquatic Research, 44*(4), 327-332.

APPENDIX

Benzo(a)pyreneAnthraceneFluroantheneNapthaleneTotalPAHsconc.µg/kg*Stations

Stations		Benzo(a)pyrene	Anthracene	Fluroanthene	Napthalene	TotalPAHsconc.µg/kg
1	Mean	.04203333	.40016667	.12008333	.00683333	.56911667
	Std.Deviation	.051581433	.297746028	.169317055	.003040136	.343693936
	Minimum	.002000	.002000	.002000	.001000	.117600
	Maximum	.113500	.688000	.385000	.010000	.994000
2	Mean	.04158333	.40808333	.12183333	.00516667	.57666667
	Std.Deviation	.052464028	.300653367	.173438401	.003214550	.349342625
	Minimum	.001000	.004000	.001000	.001000	.118900
	Maximum	.114600	.680000	.383000	.010000	.987000
3	Mean	.04122500	.40750000	.12266667	.00575000	.57714167
	Std.Deviation	.052259425	.301182669	.173475874	.003441062	.353177587
	Minimum	.003000	.001000	.002000	.001000	.119200
	Maximum	.112800	.698000	.377000	.010000	1.005000
Total	Mean	.04161389	.40525000	.12152778	.00591667	.57430833
	Std.Deviation	.050593552	.291193836	.167102618	.003219361	.338669190
	Minimum	.001000	.001000	.001000	.001000	.117600
	Maximum	.114600	.698000	.385000	.010000	1.005000

Benzo(a)pyreneAnthraceneFluroantheneNapthaleneTotalPAHsconc.uMonth

Month		Benzo(a)pyrene	Anthracene	Fluroanthene	Napthalene	TotalPAHsconc. µg/kg
Sept	Mean	.04360000	.42677778	.12977778	.00888889	.60904444
	Std.Deviation	.052411902	.316139122	.183011460	.001269296	.372102117
	Minimum	.007000	.007000	.006000	.007000	.136300
	Maximum	.114600	.698000	.385000	.010000	.994000
Oct	Mean	.04416667	.42644444	.13166667	.00755556	.60983333
	Std.Deviation	.051674800	.316599949	.184771480	.001810463	.376884412
	Minimum	.005000	.006000	.005000	.004000	.137000
	Maximum	.113700	.688000	.383000	.010000	1.005000
Jan	Mean	.03954444	.38477778	.11388889	.00322222	.54143333
	Std.Deviation	.053584352	.290374232	.167669649	.002818589	.332154678
	Minimum	.001000	.002000	.001000	.001000	.117600
	Maximum	.111400	.656000	.347000	.010000	.895000
Feb	Mean	.03914444	.38300000	.11077778	.00400000	.53692222
	Std.Deviation	.053727998	.290553351	.161337207	.002692582	.324660614
	Minimum	.002000	.001000	.002000	.001000	.119200
	Maximum	.111200	.644000	.351000	.010000	.884000
Total	Mean	.04161389	.40525000	.12152778	.00591667	.57430833
	Std.Deviation	.050593552	.291193836	.167102618	.003219361	.338669190
	Minimum	.001000	.001000	.001000	.001000	.117600
	Maximum	.114600	.698000	.385000	.010000	1.005000

One way

ANOVA

		SumofSquares	df	MeanSquare	F	Sig.
Benzo(a)pyrene	BetweenGroups	.000	3	.000	.022	.995
	WithinGroups	.089	32	.003		
	Total	.090	35			
Anthracene	BetweenGroups	.016	3	.005	.059	.981
	WithinGroups	2.951	32	.092		
	Total	2.968	35			
Fluroanthene	BetweenGroups	.003	3	.001	.034	.991
	WithinGroups	.974	32	.030		
	Total	.977	35			
Napthalene	BetweenGroups	.000	3	.000	13.416	.000
	WithinGroups	.000	32	.000		
	Total	.000	35			
TotalPAHsconc.	BetweenGroups	.045	3	.015	.120	.948

	WithinGroups	3.970	32	.124		
	Total	4.014	35			

PostHocTests

HomogeneousSubsets

Benzo(a)pyrene

Duncan[a]

Month	N	Subsetforalpha=0.05 1
Feb	9	.03914444
Jan	9	.03954444
Sept	9	.04360000
Oct	9	.04416667
Sig.		.856

Meansforgroupsinhomogeneoussubsetsaredisplayed.

a.UsesHarmonicMeanSampleSize=9.000.

Anthracene

Duncan[a]

Month	N	Subsetforalpha=0.05 1
Feb	9	.38300000
Jan	9	.38477778
Oct	9	.42644444
Sept	9	.42677778
Sig.		.784

Meansforgroupsinhomogeneoussubsetsaredisplayed.
a.UsesHarmonicMeanSampleSize=9.000.

Fluroanthene

Duncan[a]

Month	N	Subsetforalpha=0.05 1
Feb	9	.11077778
Jan	9	.11388889

Sept	9	.12977778
Oct	9	.13166667
Sig.		.820

Meansforgroupsinhomogeneoussu bsetsaredisplayed.

a.UsesHarmonicMeanSampleSize

=9.000.

Napthalene

Duncan[a]

		Subsetforalpha=0.05	
Month	N	1	2
Jan	9	.00322222	
Feb	9	.00400000	
Oct	9		.00755556
Sept	9		.00888889
Sig.		.467	.216

Meansforgroupsinhomogeneoussubsetsaredis played.

a.UsesHarmonicMeanSampleSize=9.000.

TotalPAHsconc. µg/kg

Duncan[a]

		Subsetforalpha=0.05
Month	N	1
Feb	9	.53692222

Jan	9	.54143333
Sept	9	.60904444
Oct	9	.60983333
Sig.		.694

Meansforgroupsinhomogeneoussu

bsetsaredisplayed.

a.UsesHarmonicMeanSampleSize

=9.000.

ONEWAYBenapAntracfluronnapthTotaPAHBYStations

/MISSINGANALYSIS

/POSTHOC=DUNCANALPHA(0.05).

Oneway

ANOVA

		SumofSquares	df	MeanSquare	F	Sig.
Benzo(a)pyrene	BetweenGroups	.000	2	.000	.001	.999
	WithinGroups	.090	33	.003		
	Total	.090	35			
Anthracene	BetweenGroups	.000	2	.000	.003	.997
	WithinGroups	2.967	33	.090		
	Total	2.968	35			
Fluroanthene	BetweenGroups	.000	2	.000	.001	.999
	WithinGroups	.977	33	.030		
	Total	.977	35			
Napthalene	BetweenGroups	.000	2	.000	.820	.449
	WithinGroups	.000	33	.000		
	Total	.000	35			
TotalPAHsconc.ug/uL	BetweenGroups	.000	2	.000	.002	.998
	WithinGroups	4.014	33	.122		
	Total	4.014	35			

PostHocTests

HomogeneousSubsets

Benzo(a)pyrene

Duncan[a]

Stations	N	Subsetforalpha=0.05
		1
3	12	.04122500
2	12	.04158333
1	12	.04203333
Sig.		.972

Meansforgroupsinhomogeneoussubsetsaredisplayed.

a.UsesHarmonicMeanSampleSize=12.000.

Anthracene

Duncan[a]

Stations	N	Subsetforalpha=0.05
		1
1	12	.40016667
3	12	.40750000
2	12	.40808333
Sig.		.952

Meansforgroupsinhomogeneoussubsetsaredisplayed.

a.UsesHarmonicMeanSampleSize=1

2.000.

Fluroanthene

Duncan[a]

Stations	N	Subsetforalpha=0.05
		1
1	12	.12008333
2	12	.12183333
3	12	.12266667
Sig.		.973

Meansforgroupsinhomogeneoussubs

etsaredisplayed.

a.UsesHarmonicMeanSampleSize=1

2.000.

Napthalene

Duncan[a]

Stations	N	Subsetforalpha=0.05
		1
2	12	.00516667
3	12	.00575000
1	12	.00683333
Sig.		.243

Meansforgroupsinhomogeneoussubs

etsaredisplayed.

a.UsesHarmonicMeanSampleSize=

12.000.

TotalPAHsconc. µg/kg

Duncan[a]

Stations	N	Subsetforalpha=0.05 1
1	12	.56911667
2	12	.57666667
3	12	.57714167
Sig.		.958

Meansforgroupsinhomogeneoussubs

etsaredisplayed.

a.UsesHarmonicMeanSampleSize=

12.000.