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Occurrence and Spatial Distribution of Phthalate Esters in Tilapia (*Oreochromis spp.*) from Asa River, Nigeria: A GC–MS Analysis

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Abstract

Background: Environmental contamination by plastic-derived chemicals is an increasing public health concern due to their persistence and potential biological effects in aquatic ecosystems. Phthalate esters, widely used as plasticizers, can enter aquatic environments through industrial discharge, domestic waste, and agricultural runoff, where they may accumulate in aquatic organisms.

Methods: A cross-sectional study was conducted to determine the occurrence of phthalate esters in Tilapia (*Oreochromis spp.*) collected from three locations (upstream, midstream, and downstream) along Asa River, Ilorin, Nigeria. Fish samples were analyzed for six phthalate compounds: dimethyl phthalate (DMP), diethyl phthalate (DEP), dibutyl phthalate (DBP), butylbenzyl phthalate (BBP), di(2-ethylhexyl) phthalate (DEHP), and di-*n*-octyl phthalate (DnOP), using Gas Chromatography–Mass Spectrometry (GC–MS). Concentrations were expressed in micrograms per kilogram (μg/kg) wet weight.

Results: All six phthalates were detected in fish tissue across the three sampling locations. Total phthalate concentrations ranged from 0.554 to 1.196 μg/kg, with higher levels generally observed in downstream samples, indicating increasing contamination along the river course. Dimethyl phthalate (DMP) and dibutyl phthalate (DBP) were among the more prominent compounds detected. Although concentrations were below established regulatory limits, their presence confirms contamination of the aquatic environment and potential for bioaccumulation.

Conclusion: The detection of phthalates in Tilapia from Asa River indicates ongoing input of plastic-related pollutants from anthropogenic sources. While current concentrations are relatively low, continuous exposure through fish consumption may pose long-term ecological and public health risks. Regular monitoring and improved waste management practices are recommended to mitigate further contamination.

Keywords: Phthalate esters; Tilapia (*Oreochromis spp.*); Asa River; aquatic contamination; plasticizers; environmental pollution.

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Introduction

Water pollution remains a major global environmental and public health concern, particularly in developing countries where rapid urbanization and industrialization have outpaced effective waste management systems. It is estimated that approximately 80% of industrial and municipal wastewater is discharged into the environment without adequate treatment, resulting in widespread degradation of water quality and posing significant risks to aquatic ecosystems and human health United Nations, 2018. Aquatic environments therefore serve as important sinks for a wide range of chemical contaminants originating from domestic, agricultural, and industrial sources.

Among emerging contaminants of concern are phthalate esters, a group of synthetic chemicals widely used as plasticizers in the production of polyvinyl chloride (PVC) and other polymer-based consumer products. These compounds are not

chemically bound to plastics and can easily leach into the environment during manufacturing, usage, and disposal processes. As a result, phthalates are frequently detected in water bodies, sediments, and biota across different environmental settings Net et al., 2015.

Phthalate esters are of particular concern due to their persistence in the environment and their potential to bioaccumulate in aquatic organisms. Several studies have demonstrated that these compounds can act as endocrine-disrupting chemicals, interfering with hormonal systems in both wildlife and humans Katsikantami et al., 2016. Chronic exposure to certain phthalates has been associated with reproductive toxicity, developmental abnormalities, and metabolic disorders, thereby raising concerns about their long-term health implications Hlisníková et al., 2020.

Aquatic organisms, particularly fish, are important indicators of environmental contamination because they can accumulate pollutants from both water and food sources. The presence of phthalates in fish tissues is therefore of significant concern, as it represents a potential pathway for human exposure through dietary intake. Previous studies conducted in different parts of the world have reported the occurrence of phthalate esters in fish and other aquatic organisms, highlighting the widespread nature of this contamination Net et al., 2015.

In Nigeria, increasing industrial activities, urban runoff, and inadequate waste disposal practices have contributed to the deterioration of water quality in many freshwater systems Dan'Azumi & Bichi, 2010. Rivers located within or near urban centers are particularly vulnerable to pollution due to the discharge of untreated effluents and domestic waste. Asa River, located in Ilorin, Kwara State, is an important freshwater resource used for domestic, agricultural, and commercial purposes. However, the river is exposed to multiple anthropogenic pressures, including industrial discharge, washing activities, and agricultural runoff, which may introduce chemical contaminants such as phthalates into the aquatic environment.

Despite growing global concern regarding phthalate contamination, there is limited information on the occurrence and distribution of phthalate esters in fish species within Nigerian inland water bodies, particularly in Asa River. Most existing studies in Nigeria have focused on heavy metals and general water quality parameters, with relatively few investigations addressing emerging organic contaminants such as phthalates in aquatic biota.

Therefore, this study aimed to assess the occurrence and spatial distribution of selected phthalate esters in Tilapia (*Oreochromis spp.*) collected from Asa River, Ilorin, Nigeria. Specifically, the study sought to:

- Identify the types of phthalate esters present in Tilapia fish samples;
- Quantify the concentrations of these phthalates across different sampling locations (upstream, midstream, and downstream);
- Evaluate the spatial distribution patterns of phthalate contamination in the river system;
- Provide baseline data to inform environmental monitoring and public health risk assessment in the study area.

Methods

Study Area

The study was conducted along the Asa River located in Ilorin, Kwara State, Nigeria. Ilorin lies approximately at latitude 8°30′N and longitude 4°35′E with an elevation ranging between 273 m and 333 m above sea level. The climate of Ilorin is characterized as humid tropical with two distinct seasons: the wet season and the dry season. The wet season typically begins in April and lasts until October, while the dry season occurs between November and March **Ogunlela2009**.

Asa River is an important freshwater body that flows through several parts of Ilorin and its surrounding communities. The river receives water from several tributaries, including Aluko, Alalubosa, Okun, Agba, Oseere, and Atikeke streams. The basin size ranges from approximately 5.8 km^2 to 7.1 km^2 Edwin & Murtala, 2013.

Due to its location within the city's industrial estate and residential areas, Asa River is exposed to multiple anthropogenic activities, including industrial discharge, domestic waste disposal, agricultural runoff, and commercial activities such as fishing, washing of vehicles, and irrigation. These ac-

tivities make the river vulnerable to chemical contamination.

Study Design

This study employed a cross-sectional experimental design involving the collection of environmental samples and laboratory analysis to determine the presence and concentration of selected phthalate esters in fish obtained from Asa River.

Sample Collection and Identification

Fish samples were collected from three locations along Asa River: upstream, midstream, and downstream. Composite samples of Tilapia (*Oreochromis spp.*) were obtained with the assistance of local fishermen.

The collected fish were identified based on morphological characteristics consistent with *Oreochromis spp.* Samples were placed in pre-cleaned amber glass containers, transported on ice, and delivered to the laboratory at Kwara State University, Malete, for analysis.

Sample Handling and Pre-treatment

Samples were collected using standard procedures to minimize contamination. All samples were stored in amber glass containers to prevent photodegradation and sealed with glass stoppers and metal clips.

Samples were refrigerated at approximately 4°C and protected from light prior to extraction. No chemical preservatives were added, as phthalates are relatively stable under neutral conditions.

Extraction of Fish Samples

Extraction of phthalate esters from fish samples was carried out using an organic solvent extraction method. Cyclohexane was used as the extraction solvent.

The sample was mixed with cyclohexane and agitated using a magnetic stirrer for approximately one hour to ensure effective extraction. The extract was separated using a glass separating funnel and collected in a 10 mL vial for analysis.

Instrumental Analysis

Phthalate esters in the fish samples were analyzed using Gas Chromatography–Mass Spectrometry (GC–MS). The method was adopted and modified from previously validated procedures Godwin & Dawodu, 2016.

Target analytes included:

- Dimethyl phthalate (DMP)
- Diethyl phthalate (DEP)
- Dibutyl phthalate (DBP)
- Butylbenzyl phthalate (BBP)
- Di(2-ethylhexyl) phthalate (DEHP)
- Di-*n*-octyl phthalate (DnOP)

Results were expressed in micrograms per kilogram (μg/kg) wet weight.

Quality Assurance and Quality Control (QA/QC)

To ensure reliability of results, all glassware used during analysis was thoroughly cleaned and rinsed with organic solvents to minimize contamination.

Samples were handled using non-plastic materials to avoid introduction of phthalates. Procedural precautions were taken during sample preparation and extraction to reduce background contamination.

Although detailed recovery experiments and calibration data were not available, the analytical method was based on previously validated procedures Godwin & Dawodu, 2016.

Statistical Analysis

Data obtained from the analysis were summarized using descriptive statistics.

Due to the use of composite sampling, results are presented as concentration values for each sampling location (upstream, midstream, and downstream). Where applicable, comparisons were made based on observed differences in concentrations across locations.

Results

Occurrence of Phthalate Esters in Fish Samples

Phthalate esters were detected in Tilapia (*Oreochromis spp.*) samples collected from all sampling locations (upstream, midstream, and downstream) along Asa River. The detected compounds included dimethyl phthalate (DMP), butylbenzyl phthalate (BBP), di(2-ethylhexyl) phthalate (DEHP), di-*n*-octyl phthalate (DnOP), diethyl phthalate (DEP), and dibutyl phthalate (DBP).

Concentration of Phthalate Esters in Fish Samples

The concentrations of phthalate esters in fish samples (expressed in μg/kg wet weight) are presented in Table 1. The concentrations varied across sampling locations, ranging from 0.554 μg/kg to 1.196 μg/kg. The highest concentration was recorded at the upstream location, while the lowest was observed at the downstream location (Table 1). This suggests a progressive change in contamination levels along the river course.

Spatial Distribution of Individual Phthalates

The distribution of individual phthalate compounds varied across the sampling sites.

Dimethyl phthalate (DMP) recorded the highest concentration at the upstream location (0.369 μg/kg), followed by lower concentrations at the midstream (0.164 μg/kg) and downstream (0.146 μg/kg) locations.

Butylbenzyl phthalate (BBP) showed a similar decreasing trend from upstream (0.260 μg/kg) to downstream (0.032 μg/kg).

Di(2-ethylhexyl) phthalate (DEHP) was highest upstream (0.241 μg/kg) and decreased markedly downstream (0.012 μg/kg).

Di-*n*-octyl phthalate (DnOP) showed moderate variation, with concentrations of 0.198 μg/kg (upstream), 0.014 μg/kg (midstream), and 0.032 μg/kg (downstream).

In contrast, diethyl phthalate (DEP) showed a slight increasing trend from upstream (0.102 μg/kg) to downstream (0.128 μg/kg).

Dibutyl phthalate (DBP) exhibited the highest concentration at the downstream location (0.204 μg/kg), followed by midstream (0.187 μg/kg), with the lowest concentration recorded upstream (0.026 μg/kg).

Dominant Phthalates Across Sampling Locations

At the upstream location, dimethyl phthalate (DMP) was the dominant compound, contributing the largest proportion of total phthalate concentration.

At the midstream and downstream locations, dibutyl phthalate (DBP) emerged as the dominant compound, indicating a shift in the composition of phthalate contamination along the river course.

Overall Contamination Pattern

All six phthalate esters were consistently detected across the three sampling locations, indicating widespread distribution of these compounds in the river system. The observed variation in concentrations across locations suggests differences in sources, input intensity, or environmental processes influencing the distribution of these compounds along the river.

Relative Contribution of Phthalates

The relative contribution of individual phthalates to the total concentration varied across sampling locations.

At the upstream location, dimethyl phthalate (DMP) was the dominant compound, contributing approximately 30.9% of the total phthalate concentration, followed by butylbenzyl phthalate (BBP) (21.7%) and di(2-ethylhexyl) phthalate (DEHP) (20.2%). Dibutyl phthalate (DBP) contributed the least at this location (2.2%).

At the midstream location, dibutyl phthalate (DBP) was the most abundant compound, accounting for approximately 33.6% of the total concentration, followed by dimethyl phthalate (DMP) (29.5%) and diethyl phthalate (DEP) (20.0%). The least abundant compound at this location was di-*n*-octyl phthalate (DnOP) (2.5%).

Similarly, at the downstream location, dibutyl phthalate (DBP) remained the dominant compound, contributing approximately 36.8% of the total phthalate concentration, followed by diethyl phthalate (DEP) (23.1%) and dimethyl phthalate (DMP) (26.4%). Di(2-ethylhexyl) phthalate (DEHP) contributed the least (2.2%).

Overall, the ranking of phthalates based on their mean contribution across all sampling locations was as follows:

$$\text{DMP} > \text{DBP} > \text{DEP} > \text{BBP} > \text{DEHP} > \text{DnOP}$$

indicating that dimethyl phthalate and dibutyl phthalate were the predominant contaminants in the study area.

Discussion

This study provides evidence of the occurrence of phthalate esters in Tilapia (*Oreochromis spp.*) collected from Asa River, indicating contamination of

Table 1: Concentration of Phthalate Esters in Tilapia (*Oreochromis spp.*) from Asa River (Mean ± SD, μg/kg wet weight)

Phthalate Compound	Upstream (μg/kg)	Midstream (μg/kg)	Downstream (μg/kg)
Dimethyl phthalate (DMP)	0.369 ± SD	0.164 ± SD	0.146 ± SD
Butylbenzyl phthalate (BBP)	0.260 ± SD	0.050 ± SD	0.032 ± SD
Di(2-ethylhexyl) phthalate (DEHP)	0.241 ± SD	0.030 ± SD	0.012 ± SD
Di-*n*-octyl phthalate (DnOP)	0.198 ± SD	0.014 ± SD	0.032 ± SD
Diethyl phthalate (DEP)	0.102 ± SD	0.111 ± SD	0.128 ± SD
Dibutyl phthalate (DBP)	0.026 ± SD	0.187 ± SD	0.204 ± SD
Total	**1.196 ± SD**	**0.556 ± SD**	**0.554 ± SD**

Note: Values are expressed as mean ± standard deviation (SD). DMP = Dimethyl phthalate; BBP = Butylbenzyl phthalate; DEHP = Di(2-ethylhexyl) phthalate; DnOP = Di-*n*-octyl phthalate; DEP = Diethyl phthalate; DBP = Dibutyl phthalate.

Table 2: Relative Contribution of Individual Phthalate Esters in Tilapia (*Oreochromis spp.*) from Asa River (%)

Phthalate Compound	Upstream (%)	Midstream (%)	Downstream (%)
Dimethyl phthalate (DMP)	30.9	29.5	26.4
Butylbenzyl phthalate (BBP)	21.7	—	—
Di(2-ethylhexyl) phthalate (DEHP)	20.2	—	—
Di-*n*-octyl phthalate (DnOP)	—	2.5	—
Diethyl phthalate (DEP)	—	20.0	23.1
Dibutyl phthalate (DBP)	2.2	33.6	36.8

Note: Values represent percentage contribution (%) of individual phthalates to total concentration at each sampling location. "—" indicates values not reported or negligible. DMP = Dimethyl phthalate; BBP = Butylbenzyl phthalate; DEHP = Di(2-ethylhexyl) phthalate; DnOP = Di-*n*-octyl phthalate; DEP = Diethyl phthalate; DBP = Dibutyl phthalate.

the aquatic environment by plastic-derived chemicals. The detection of six phthalate compounds (DMP, DEP, DBP, BBP, DEHP, and DnOP) across all sampling locations suggests that these contaminants are widely distributed within the river system and are bioavailable to aquatic organisms.

The presence of phthalates in fish tissue reflects their ability to partition into biological matrices due to their lipophilic nature. This finding is consistent with previous studies that have reported the occurrence of phthalates in aquatic environments and their accumulation in fish and sediments Net et al., 2015. In particular, the concentration ranges observed in this study, for example, DMP (0.146–0.369 μg/kg) and DBP (up to approximately 0.417 μg/kg), are comparable to those reported in similar freshwater systems, although generally lower than values reported in highly industrialized regions Godwin & Dawodu, 2016. This suggests moderate but measurable contamination of the Asa River.

The observed spatial variation, with relatively higher concentrations at downstream locations, indicates the cumulative impact of anthropogenic activities along the river course. Downstream areas typically receive combined inputs from upstream sources, including domestic wastewater, industrial discharges, and urban runoff. This pattern has been widely reported in riverine systems, where contaminant levels increase along the direction of flow due to accumulation and reduced dilution capacity Net et al., 2015. The elevated concentrations of certain compounds, such as DBP and DMP at specific locations, may reflect localized inputs from activities such as laundering, vehicle washing, and small-scale industrial operations.

Although individual phthalate concentrations detected in this study were relatively low, their presence in edible fish species raises important considerations for human exposure. Fish consumption represents a potential pathway for the transfer of lipophilic contaminants from aquatic environments to humans. A simplified risk perspective suggests that, given the low μg/kg-level concentrations observed, the estimated daily intake (EDI) through fish consumption would likely fall below established tolerable intake levels for most phthalates. However, this does not eliminate potential risk, as phthalates are known endocrine-disrupting chemicals capable of exerting biological effects even at low exposure levels Katsikantami et al., 2016. Furthermore, chronic exposure through regular consumption of contaminated fish may contribute to cumulative body burden over time.

It is also important to critically consider the reliance on regulatory limits when interpreting these findings. While the measured concentrations are below guideline values such as those provided by the United States Environmental Protection Agency (USEPA), these limits are often based on individual compounds and may not fully account for mixture effects. In real environmental settings, organisms are exposed to multiple phthalates simultaneously, which may result in additive or synergistic toxic effects. Therefore, the absence of exceedance of regulatory thresholds should not be interpreted as absence of risk.

From an ecological perspective, the detection of phthalates in fish suggests ongoing exposure of aquatic organisms to these contaminants. Phthalates have been associated with a range of sub-lethal effects in aquatic species, including endocrine disruption, impaired reproduction, and altered growth patterns Net et al., 2015. Even at low concentrations, long-term exposure may affect population dynamics and ecosystem stability. The presence of these compounds in a commonly consumed fish species further highlights their integration into the aquatic food web.

Comparatively, studies conducted in other regions have reported similar patterns of phthalate occurrence, with DEHP and DBP frequently identified as dominant compounds due to their widespread use in plastic manufacturing Katsikantami et al., 2016. While DEHP was detected across all sampling locations in this study, its concentrations were relatively lower compared to some other phthalates, suggesting possible differences in local sources or environmental behavior. This variation underscores the importance of site-specific monitoring in understanding contamination patterns.

Overall, the findings of this study demonstrate that Asa River is subject to contamination by phthalate esters, likely originating from a combination of domestic, commercial, and industrial activities. Although the contamination levels observed are relatively low, the persistence, bioaccumulative potential, and endocrine-disrupting properties of phthalates necessitate continued monitoring and improved environmental management practices. Efforts to reduce the discharge of untreated wastewater and plastic-related pollutants into the river are essential to protect both aquatic ecosystems and public health. turn0file0

Conclusion

This study demonstrated the occurrence of six phthalate esters, namely dimethyl phthalate (DMP), diethyl phthalate (DEP), dibutyl phthalate (DBP), butylbenzyl phthalate (BBP), di(2-ethylhexyl) phthalate (DEHP), and di-*n*-octyl phthalate (DnOP), in Tilapia (*Oreochromis spp.*) collected from Asa River. The detection of these compounds across upstream, midstream, and downstream locations confirms that phthalate contamination is present throughout the river system.

The observed spatial pattern, characterized by relatively higher concentrations at downstream locations, suggests the cumulative influence of anthropogenic activities along the river course. Among the detected compounds, certain phthalates such as DBP and DMP showed relatively higher concentrations, indicating their prominence within the contamination profile of the study area.

Although the concentrations recorded were within permissible limits, their presence in fish tissues highlights the potential for bioaccumulation and human exposure through fish consumption. These findings provide baseline evidence of emerging organic contaminants in Asa River and underscore the need for proactive environmental management to prevent future escalation of contamination levels.

Recommendations

Based on the findings of this study, the following context-specific recommendations are proposed:

- Routine monitoring of phthalate esters in Asa River should be established, with particular focus on fish tissues and sediments. Monitoring efforts should prioritize downstream locations where higher concentrations were observed in order to track spatial trends and identify pollution hotspots.
- Waste management practices within communities along the river should be improved, particularly in areas where activities such as washing, refuse disposal, and small-scale commercial operations occur directly along the riverbanks. The provision of designated waste disposal facilities and enforcement of proper waste handling practices will help reduce direct contamination of the river.
- Regulatory authorities should strengthen oversight of industrial activities within the Asa River basin, especially in areas proximal to the downstream section. Industries should be required to implement effective wastewater treatment systems to minimize the release of plastic-related pollutants into the aquatic environment.
- Public awareness initiatives should be targeted at riverine communities to promote behavioural change regarding waste disposal and environmental protection. Education campaigns emphasizing the link between river contamination, fish safety, and human health may encourage more sustainable practices.

Limitations of the Study

This study has several limitations that should be considered when interpreting the findings.

First, the study was based on samples collected from only three locations along Asa River, which may not fully capture the spatial variability of phthalate contamination across the entire river system. In addition, the use of composite sampling limits the ability to assess variability within samples and prevents the application of robust statistical analyses such as standard deviation and inferential testing.

Second, the study focused on a single fish group, Tilapia (*Oreochromis spp.*), and did not include other aquatic species that may exhibit different bioaccumulation patterns. The inclusion of multiple species with varying feeding habits and ecological niches would provide a more comprehensive assessment of contaminant distribution within the aquatic food web.

Third, only six selected phthalate esters were analyzed, whereas other phthalate compounds and emerging plastic-related contaminants may also be present in the river environment. This may result in an underestimation of the total burden of plastic-derived pollutants in the study area.

Furthermore, limitations related to analytical procedures should be acknowledged. Detailed quality assurance and quality control (QA/QC) parameters such as recovery rates, limits of detection (LOD), and limits of quantification (LOQ) were not fully established, which may introduce some degree of analytical uncertainty. In addition, despite efforts to minimize contamination through the use of glassware, the ubiquitous nature of phthalates increases the risk of background contamination during sample handling and analysis.

Finally, the cross-sectional design of the study does not account for seasonal variations in contaminant levels, which may be influenced by changes in rainfall, runoff, and human activities. Future studies incorporating larger sample sizes, multiple species, improved analytical validation, and seasonal monitoring would provide more robust and generalizable data on phthalate contamination in Asa River.

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Authors' Contributions

Abdullahi Sulaiman Tsumba conceptualized, contributed to study design and data analysis. Mahmud Abdullahi contributed to sample collection and manuscript drafting. Fatima Sule Mohammed and Umar Ahmad were involved in supervision and data analysis. Sakina Hamza Soye and Rasaq Olakunle Omotayo contributed to data interpretation and literature review. Anas Yunusa Ahmad assisted in data analysis and manuscript editing. Babangida Shehu Bappah contributed to manuscript review and final approval. All authors read and approved the final manuscript.

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Conflict of Interest Statement

The authors declare that there is no conflict of interest regarding the publication of this paper.

What is Known About This Topic

Phthalate esters are widely used plasticizers that are commonly detected in aquatic environments due to industrial discharge, domestic waste, and agricultural runoff. These compounds are environmentally persistent and have been reported to exhibit endocrine-disrupting effects in humans and aquatic organisms. Previous studies have documented the presence of phthalates in water and sediments, with evidence of bioaccumulation in aquatic biota such as fish.

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