African Scientist Vol. 25, No. 4 December 31, 2025
Printed in Nigeria
1595-6881/2023 $80.00 + 0.00
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https://africansciientistjournal.org

afs2025052/26409

Comparative Acute Toxicity Assessment of Industrial Effluents from Benin City, Nigeria Using the African Catfish (*Clarias gariepinus*)

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(Received December 23, 2025; Accepted in revised form December 27, 2025)

ABSTRACT: This study investigated effluents from three industrial facilities (alcoholic, non-alcoholic, and abattoir) located in Benin City, Nigeria, to ascertain the suitability of the discharges for aquatic life using the African catfish (*Clarias gariepinus*). The physicochemical properties and heavy metals assessment of the effluents were measured using standard methods, while the acute toxicity assays were done using a static bioassay with continuous aeration for 96 h under laboratory conditions. The results indicate non-conformation with the standards set by national and international regulatory bodies for most of the physicochemical parameters (78.79%), while the levels of the heavy metals were majorly (76.67%) below the permissible limits. The alcoholic industrial effluent recorded the highest mortality with a 96-h LC_{50} of 8.71 mg/L despite having better effluent quality. The observed mortality showed alcoholic (60%) > non-alcoholic (42.50%) > abattoir (41.25%) effluents, whereas the effluent quality assessment result showed alcoholic > non-alcoholic > abattoir effluents. The dissolved oxygen levels and the overall effect of the complex mix of pollutants in the effluents may have exerted a considerable contribution to their toxicity to the fish. Hence, toxicity assays should be carried out complementarily to physicochemical and microbiological evaluations in effluents before discharge into water bodies.

Keywords: Industrial effluent; Acute toxicity; Toxicity assessment; African catfish; Benin City

Introduction

Civilisation and modernisation of the world and its inherent processes have bequeathed adverse environmental challenges to the global community. Industrialisation is a key component of the World's civilisation and modernisation. The vast arrays of industries expends huge volumes of water in carrying out their operations, and equally churn out wastewater of varying degrees of contamination, depending on the industry. Over the years, the quality of effluent discharged into water bodies has attracted growing interest due to environmental concerns and the contamination of freshwater sources by untreated or improperly treated effluents (Akharame *et al.*, 2017). The perceived challenge is heightened by the contamination of the Earth's surface and groundwater sources, which are gradually being depleted in quantity and quality (Oputu and Akharame, 2022). Hence, it becomes pertinent to carry out routine monitoring and quality assessment of effluent from industries deemed as major contributors to ascertain the quality and possible environmental implications of their discharges.

In Nigeria, the National Environmental Standards and Regulations Enforcement Agency (NESREA), established in the year 2007, is saddled with the protection of the environment. The body is charged with the regulation of wastewater and effluent discharges, and enforcing the compliance status of the various industries

(NESREA,2007). The government agency has formulated set guidelines and permissible limits for physical, chemical, and microbial contaminants for the monitoring of industrial effluents (NESREA, 2011). And there is an ongoing concerted effort by it to carry out its statutory monitoring and enforcement functions to ensure that Nigerians get a cleaner and healthier environment. The daunting task of enforcing compliance and ensuring that industries meet the quality requirements before discharging their effluents has been a challenge. Reports from literary works suggest that some facilities still cut corners and discharge untreated or improperly treated effluents into water bodies or the environment (Akharame *et al*., 2017; Gbarakoro *et al*., Imoobe & Koye, 2011). The discharge of untreated or improperly treated effluents into the aquatic environment may cause toxic effects on the organisms present in the receiving water bodies. The adverse environmental effects of the effluents may affect the abundance, location, and/or sizes of the aquatic organisms, and can also lead to their death (Olorunfemi *et al*., 2019). More so, some aquatic organisms may accumulate hazardous compounds in their tissues, which when consumed by humans, may have deleterious effects.

Fishes are important aquatic organisms that constitute a source of protein in the meals of average Nigerians (Nwafili & Gao, 2007); and they tend to be extremely sensitive to evaluating and monitoring toxicity in aquatic systems (Olorunfemi *et al*., 2019). The African catfish (*Clarias gariepinus*) has proven to be a model test species for the assessment of effluents or surface water quality (Ibrahem *et al*., 2011). It serves as an effective biomarker for contaminants and can be used to monitor the health of an aquatic ecosystem (Olaniran *et al*., 2019). The African catfish possesses strong adaptability characteristics; they can survive in extreme environmental conditions and can survive in oxygen-depleted waters due to their uniquely developed air-breathing organ (Graaf & Janssen, 1996). The fish is largely consumed in Nigeria and is found in aquatic environments such as swamps, rivers, and lakes in Africa and the Middle East (Olaniran *et al*., 2019). Its adaptive characteristics and abundance in different water systems make it a distinct and valuable candidate for numerous aquatic toxicity monitoring programs covering acute toxicity, and biochemical and histological assays (Olaniran *et al*., 2019; Olajumoke *et al*., 2022; Ariyomo *et al*., 2021; Dahunsi *et al*., 2012).

In this study, we sought to evaluate the potential environmental impact of the effluents from selected industries located in Benin City, Nigeria. The effluents were obtained from a non-alcoholic (carbonated drink), an alcoholic (brewery), and a meat processing (abattoir) facility. Several literature works have reported investigations on the effluent quality from these industries, but the reports are mostly limited to physical, chemical, and biological evaluations (Akharame *et al*., 2017; Imoobe & Koye, 2011; Akpoka *et al*., Adegbite *et al*., 2018). Consequently, our research is focused on using the African catfish (*C. gariepinus*) to assess the toxicological and possible environmental impacts of the effluent discharges from these facilities. Acute toxicity tests were carried out to measure the responses and effects of the effluents on the African catfish. This report will provide scarce data on the ecological effects of the effluents from the selected industries on the various stakeholders (owners, regulatory agencies, policymakers, and others) for necessary actions to protect the aquatic ecosystems of the receiving water bodies.

Materials and methods

Sampling sites and collection of effluents: The effluent samples were collected from the discharge points of a non-alcoholic (carbonated drink), an alcoholic (brewery), and a meat processing (abattoir) facility located in Benin City, Nigeria. The GPS (global positioning system) coordinates of the sampling locations are: the alcoholic (6°20′ N and 5°39′ E), non-alcoholic (6°27′8′′ N and 5°36′33′′ E), and abattoir (6°13′ N and 5°46′ E). The effluent samples were collected with 10-litre plastic containers, which were thoroughly washed and rinsed with distilled water and rinsed three times with the effluents before filling up at the discharge points. The samples were preserved with ice during transportation to the laboratory and eventually stored in the refrigerator at 4 °C before the physicochemical and heavy metals analyses, and further utilisation for the acute toxicity assessments.

Water quality parameters measurements: The physicochemical parameters measurements carried out were for pH, electrical conductivity (EC), dissolved oxygen (DO), total hardness, total suspended solids (TSS), ammonia, nitrates, phosphates, sulphates, and chlorides. The heavy metals evaluated were aluminium (Al), arsenic (As), cadmium (Cd), chromium (Cr), cobalt (Co), copper (Cu), lead (Pb), manganese (Mn), nickel (Ni), and zinc (Zn). The pH measurements were carried out *in situ* using a handheld HANNA pH/Temperature Tester (HANNA Instruments, USA). A HACH DR 3900 spectrophotometer (HACH, USA) was utilised to assess the levels of ammonia, nitrates, phosphates, sulphates, and chlorides in the effluents (APHA, 1998). The EC values were obtained using an Adwa multi-parameter meter with model number AD8000 (Adwa Instruments, Hungary), the total hardness of the samples was evaluated using the EDTA titrimetric method, and the measurements for the TSS levels were done following the method established in APHA, 1998) . The DO levels in the effluents were evaluated using the Azide modified Winkler's method (APHA, 1998). The heavy metals assessment was carried out using the Unican 929 AA spectrometer (PCB Equipment, The Netherlands) with flame atomisation. Prior digestion of the samples was done as described by Radojevic *et al*. (1999) before the analyses to eliminate the interference from particulates and organic matter.

Toxicity assessment: The acute toxicity assay was carried out following the procedure recommended by UNEP (1989). The juveniles of the African catfish (*C. gariepinus*) were sourced from a commercial fish farm located in Benin City, Nigeria (6°15′ N, 5°25′ E). A total of 260 juveniles measuring 17±1.0 cm, with an average weight of 12.4±0.5 g, were obtained for the toxicity assessment. The acclimatisation of the fish to laboratory conditions was done for two (2) weeks in glass tanks (20×15×30) with de-chlorinated water (pH 7.0), at room temperature (26.0 - 28.0 °C) and relative humidity of 78.0 ± 2%. Feeding of the fish was done two times daily (morning and evening) with a commercial fish meal (Topfeed®) at 3% body weight, with the feeding stopped 24 h before and during the assay timeline. Regular changing of the water was carried out every 24 h during the acclimatisation period to prevent waste metabolites and food debris accumulation.

The toxicity evaluation was done by placing eight (8) fish in each glass tank containing different dilution percentiles (effluent in water: 5%, 25%, 50%, 100%) of the effluents, and de-chlorinated water used for the control. Examination of the fish was done at 12, 24, 48, 72, and 96 h, respectively, to observe their behavioural patterns and record the mortalities. The absence of mobility, opercula, and tail movement was used to confirm the mortality, and dead fish were evacuated immediately, once observed.

Data analysis: The Duncan Multiple Range Test (DMRT) was used to analyse the physicochemical and heavy metals data, while Probit analysis data interpretation using the arithmetic method of percentage mortality was used to assess the 96 h LC_{50} toxicity (Randhawa, 2009). The confidence limits (lower and upper) of the LC_{50} toxicity were determined following the UNEP (1989) procedure. The MedCalc® Statistical Software version 20.113 (MedCalc Software Ltd, Ostend, Belgium) was utilised to carry out the regression analysis.

Results and Discussion

Physicochemical assessment of the effluent quality: The physicochemical assessment data obtained for the study show varying values and concentrations for the parameters of the different industrial effluents (Table 1). The pH values of 5.62, 7.17, and 5.64 were recorded for the non-alcoholic, alcoholic, and abattoir effluents, respectively. The pH values obtained for the non-alcoholic and abattoir effluent were both below the permissible limit of 6.5-8.5 set by the Nigerian environmental regulatory body- National Environmental Standards and Regulations Enforcement Agency, NESREA (NESREA, 2011) and 6.5-9.0 recommended by the United States Environmental Protection Agency (USEPA, 2023). Accordingly, the discharged effluents from the two facilities were slightly acidic and required pH adjustment before their discharge to the receiving water body. The acidic condition of the effluents may adversely impact the fishery population in the receiving water body, especially around the discharge points. Generally, the metabolic activities of aquatic fauna are pH dependent, making them easily responsive to changes in the environmental pH conditions (Ogbu *et al*., 2016). The DO levels of the effluents ranged from 2.50-6.19 mg/L, with only the non-alcoholic industry meeting the minimum limit for effluent discharge (4.0 mg/L) and fisheries and other aquatic organisms (6.0 mg/L) set by NESREA (NESREA, 2011). Turbidity set limit for effluent discharge is not stipulated in the NESREA (NESREA, 2011) guideline, however, the range of 11.35-15.13 NTU is well above (double/triple) the 5 NTU recommended for drinking water by WHO (WHO, 1996). Similarly, the TSS values (0.40-1.40 mg/L) followed the observed trend for turbidity (i.e. abattoir > non-alcoholic > alcoholic effluent), and this corroborates the strong linear relationship that exists between turbidity and TSS (Hannouche, 2011). Although the levels recorded for ammonia (0.014-0.043 mg/L) were below the maximum permissible limit of 2.0 mg/L stipulated for effluent discharges, as well as the 0.05 mg/L recommended for the sustainability of fisheries and aquatic fauna (NESREA, 2011), and as such may not pose a threat to the aquatic ecosystem.

Table 1: Physicochemical parameter values in the effluent samples from the industrial facilities

	Effluent samples			NESREA Standards (2011)		USEPA Standards (2023^{α} & 1986^{β})
Parameter	Non-alcoholic	Alcoholic	Abattoir	Effluent discharge, irrigation & reuse	Fisheries & recreational quality	Freshwater
pH	5.62±0.02^{a}	7.17±0.01^{c}	5.64±0.04^{a}	6.5 - 8.5	6.5 - 8.5	6.5 – 9.0$^{\alpha\beta}$
DO	6.19±0.08^{c}	2.50±0.10^{a}	2.77±0.03^{b}	4.0	4.0 - 6.0	4.0 - 6.0$^{\beta}$
Turbidity	12.06±0.49^{b}	11.35±0.19^{a}	15.13±0.06^{c}	-	-	-

Table 1: Physicochemical parameter values in the effluent samples from the industrial facilities (contd.)

	Effluent Samples			NESREA Standards (2011)		USEPA Standards (2023^{α} & 1986^{β}) Freshwater
Parameter	Non-alcoholic	Alcoholic	Abattoir	Effluent Discharge, Irrigation & Reuse	Fisheries & Recreational Quality	
TSS	$0.89±0.04^{b}$	$0.40±0.10^{a}$	$1.40±0.10^{c}$	0.75	0.75	0.08^{β}
Ammonia	$0.043±0.01^{a}$	$0.017±0.13^{a}$	$0.014±0.08^{a}$	2.0	0.05	-
Conductivity	$1031.00±1.00^{b}$	$864.32±0.58^{a}$	$1187.33±2.51^{c}$	-	-	-
Chloride	$751.00±1.00^{a}$	$633.67±1.53^{b}$	$766.33±1.53^{c}$	350	300	230^{α}
Hardness	$343.67±3.51^{b}$	$319.67±1.53^{a}$	$373.33±2.08^{c}$	-	-	$0 - 75^{\beta}$
Nitrates	$45.33±3.05^{b}$	$36.33±0.57^{a}$	$52.00±1.00^{c}$	40	9.1	-
Phosphates	$311.50±1.32^{a}$	$633.67±1.53^{b}$	$766.33±1.53^{c}$	3.5	3.5	-
Sulphates	$585.33±30.75^{a}$	$703.67±5.51^{b}$	$723.67±5.51^{b}$	500	100	-

Values are means ± standard deviation of three replicates. Different superscripts in the same row for the effluents indicate significant differences at $p < 0.05$ according to Duncan Multiple Range Test (DMRT). All values are expressed in mg/L except turbidity (NTU), conductivity (µs/cm), and pH (no unit). DO: Dissolved Oxygen, TSS: Total Suspended Solids. NESREA: National Environmental Standards and Regulations Enforcement Agency (2011); USEPA: United States Environmental Protection Agency (1986 & 2023), maximum permissible limits for freshwater (aquatic life)

The EC concentrations, which represent the effluents' dissolved solids (salts), ranged from 864-1187 µs/cm. Chlorides and total hardness are contributors to high EC levels in wastewaters (Ogbu *et al*., 2016), and the high EC values recorded for the different effluent corroborate the trend in the chlorides (634-766 mg/L) and total hardness (320-373 mg/L) concentrations. Both parameters exceeded the limits of 350 mg/L (NESREA, 2011) and 75 mg/L (USEPA, 1986) stipulated for effluent discharges. Essentially, water with hardness levels of more than 75 mg/L is characterised as moderately hard and very hard water, and the effects of the hardness on fisheries (freshwater) and other aquatic organisms are related to the ions responsible for the hardness (USEPA, 1986). The levels of the nitrates (36-55 mg/L), phosphates (312-766 mg/L), and sulphates (585-724 mg/L) were mostly above the stipulated maximum permissible limits of 40.0 mg/L, 3.5 mg/L, and 500 mg/L for effluent discharges, respectively. The high phosphates (312-766 mg/L) correlate with the high chloride levels (634-766 mg/L) and may have been influenced by the phosphate-based and chlorinated detergents utilised for cleaning and washing purposes in the different facilities (Akharame *et al*., 2017; Adeogun *et al*., 2013). The observed results trend was similar to the values in the literature for effluent discharges from carbonated drinks (Imoobe & Koye, 2011; Nwokedi *et al*., 1992), breweries (Adegbite *et al*., 2018; Ologbosere *et al*., 2016), and abattoirs (Olaniran *et al*., 2019; Egesi *et al*., 2019) facilities. More so, several parameters, such as pH, DO, chlorides, nitrates, and phosphates, were mostly non-compliant in these reports.

The overall assessment of the industrial effluents as analysed using the Duncan Multiple Range Test indicated that the quality follows the trend Alcoholic (a:7; b:3; c:1) > non-alcoholic (a:5; b:5; c:1) > abattoir (a:2; b:2; c:7). The effluent from the alcoholic facility had seven (7) parameters in the low range, five (5) in the middle range, while the one (1) parameter recorded in the high range was the pH value which is actually within the NESREA stipulated limit. The other effluent samples, albeit recording a low range in the DMRT, had values below the stipulated limit.

Heavy metals assessment of water quality: Heavy metals evaluation in effluents is a critical quality assessment criterion (Akharame *et al*., 2017). Trace levels of some heavy metals portend grave, damaging and toxic consequences to the organisms present in the ecosystem (Ramirez *et al*., 2020). More so, they tend to be persistent and bioaccumulate in the environmental matrices (Masindi *et al*., 2018). The heavy metal levels for the different effluents are presented in Table 2. The measured concentrations of some of the assayed heavy metals were lower than the maximum permissible limits for effluent discharges. These include As, Al, Cd, Cr, Ni, and Pb, whose NESREA's set limits for effluent discharge are 0.05 mg/L, 0.20 mg/L, 0.01 mg/L, 0.5 mg/L, 0.1 mg/L, and 0.1 mg/L, respectively (NESREA). The levels of Cu (0.017-0.113 mg/L) and Zn (2.161-7.049 mg/L) were above the set limits of 0.01 mg/L and 0.2 mg/L, respectively. The recorded levels for Mn ranged from 0.030-0.064 mg/L. Manganese tends to bioaccumulate in molluscs at exceedingly high levels, which can portend health hazards for humans; hence, a set point of 0.1 mg/L is recommended for the sustainability of aquatic life in water (USEPA, 1986). Also, the criterion of 0.05 mg/L is stipulated for drinking water.

Generally, the effluent for the brewery facility may be adjudged better using the levels of the heavy metals as it relatively recorded the least concentrations. Although the levels of the heavy metals were mostly (76.67%) compliant for the effluent discharges, some of the values (50%) tend to be much higher in comparison to the permissible limits for fisheries and other aquatic organisms (NESREA, 2011). This poses a possible threat to the aquatic fauna in the receiving water bodies in the vicinity of these facilities.

Table 2: Heavy metal levels in the effluent samples from the industrial facilities

Parameter	Effluent Samples: Non-alcoholic	Alcoholic	Abattoir	NESREA Standards (2011): Effluent Discharge, Irrigation & Reuse	Fisheries & Recreational Quality	USEPA Standards (2023[α] & 1986[β]) Freshwater
Aluminium	0.130±0.001[a]	ND	0.107±0.013[a]	0.200	0.200	-
Arsenic	0.003±0.001[a]	0.003±0.001[a]	0.007±0.001[b]	0.050	0.050	0.150[α]
Cadmium	0.003±0.001[b]	ND	0.014±0.001[c]	0.010	0.005	
Chromium	0.071±0.002[b]	0.040±.0.010[a]	0.085±0.005[c]	0.500	0.001	0.011[α]
Cobalt	0.0127±0.001[b]	ND	0.024±0.001[c]	-	-	-
Copper	0.113±0.001[c]	0.017±0.001[a]	0.046±0.001[b]	0.010	0.001	-
Lead	0.03±0.005[b]	0.01±0.002[a]	0.027±0.006[b]	0.100	0.010	0.003[α]
Manganese	0.043±0.002[a]	0.030±.010[b]	0.064±0.001[c]	-	-	0.100[β]
Nickel	0.026±0.001[c]	ND	0.009±0.001[b]	0.100	0.010	0.052[α]
Zinc	5.073±0.008[b]	2.161±0.007[a]	7.049±0.041[c]	0.200	0.020	0.120[α]

Values are means ± standard deviation of three replicates. Different superscripts in the same row for the effluents indicate significant differences at $p < 0.05$ according to Duncan Multiple Range Test (DMRT). All values are expressed in mg/L. ND: Not detected; NESREA: National Environmental Standards and Regulations Enforcement Agency (2011); USEPA: United States Environmental Protection Agency (1986 & 2023), maximum permissible limits for freshwater (aquatic life)

Acute toxicity responses of the African catfish to the industrial effluents: Acute toxicity testing regimes are designed for short-term evaluation of the adverse effects of toxic agents on aquatic organisms (*Ebrahimpour et al*., 2010). The observed behavioural patterns of the African catfish in the varying dilution percentile (effluent in water ratio: 0%, 5%, 25%, 50%, and 100%**)** of the different effluents include restlessness and erratic movements, and gasping for breath, which was more noticeable with increases in the dilution percentile (concentration) of the effluents. This led to sluggishness in movement and the eventual death of some fish. The observed responses followed the three-phase behavioural pattern, viz, active, fatigue, and collapse, posited by Ibrahim and Imam (Ibrahim *et al*., 2019). More so, previous literary works have reported these behavioural responses in abattoirs (Adeogun *et al*., 2013), breweries (Ariyomo *et al*., 2021), paint (Otong *et a*l., 2018), synthetic resins (Dahunsi & Oranusi, 2012), and tannery (Ibrahim & Imam, 2019) industrial effluents.

There was a noticeable and significant ($p<0.05$) trend indicating severe adverse effects (mortality) on the fish with increasing concentration of the industrial effluents; also, the extended exposure duration (12, 24, 48, 72, and 96 h) resulted in more mortality of the fish (Table 3). The observed effluent-induced concentration-dependent mortality result corroborates the investigations of Adeogun *et al*. (2013). Comparatively, the effluent obtained from the alcoholic industrial facility recorded the highest mortality with 60% of the fish, whereas the non-alcoholic industry and abattoir effluents resulted in a 42.50% and 41.25% mortality rate, respectively. The Probit regression data indicate that the 96-h LC_{50} (the dose required to achieve 50% mortality from toxicity) were 8.71 mg/L, 25.70 mg/L, and 28.84 mg/L for the alcoholic, non-alcoholic, and abattoir effluents, respectively (Figures 1 - 3). The 96-h LC_{50} result shows that the effluent from the alcoholic industry poses more risk to the African catfish, with a lethal concentration dose of 8.71 mg/L of the effluent capable of 50% mortality. The LC_{50} values for the non-alcoholic and abattoir effluent were quite close and correlated with the recorded mortalities. Overall, the toxicity of the effluents based on the 96-h LC_{50} values was alcoholic > non-alcoholic > abattoir effluents, accordingly.

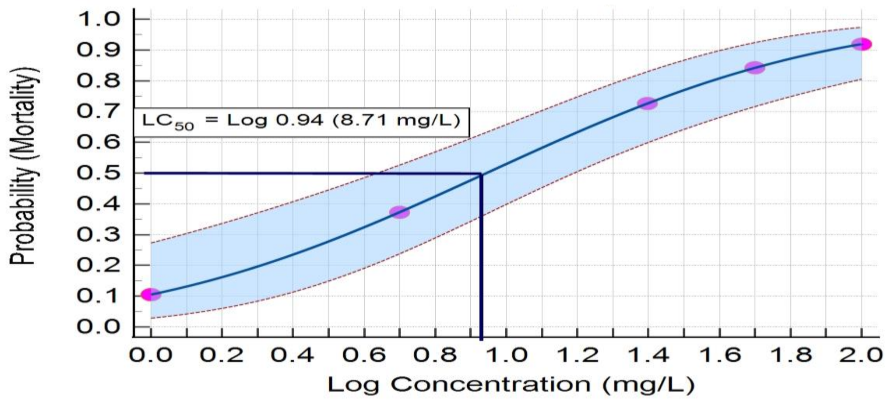

Fig. 1: Probit mortality and exposure concentration of *C. gariepinus* juveniles exposed to alcoholic industrial effluent for 96 h

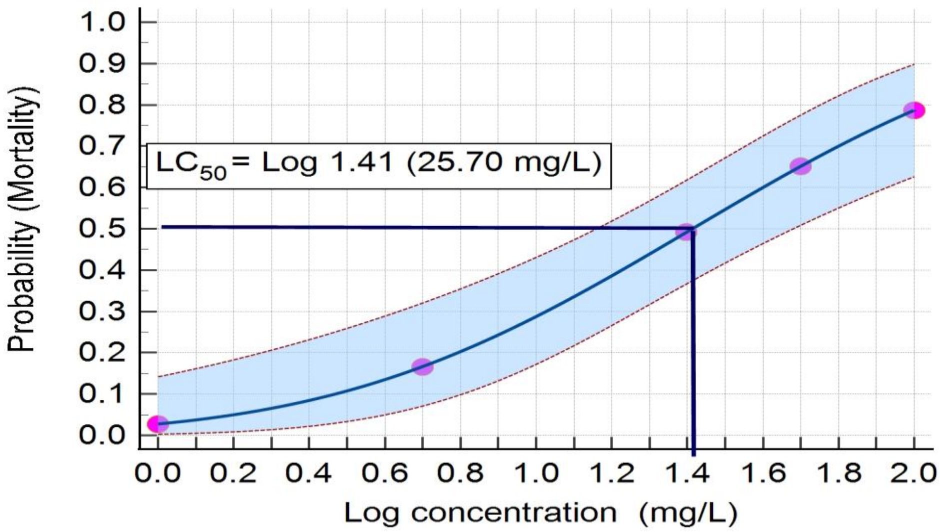

Fig. 2: Probit mortality and exposure concentration of *C. gariepinus* juveniles exposed to non-alcoholic industrial effluent for 96 h

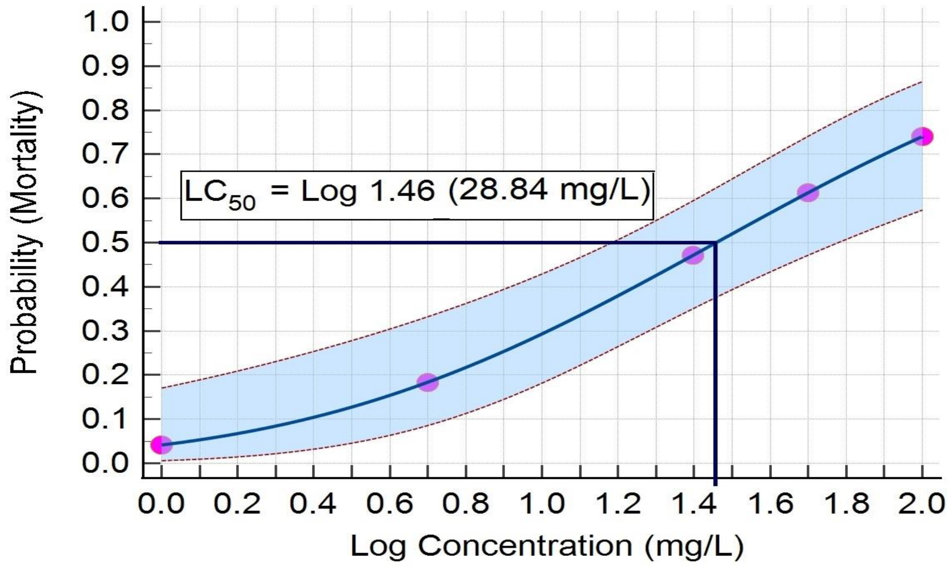

Fig 3: Probit mortality and exposure concentration of *C. gariepinus* juveniles exposed to abattoir effluent for 96 h

Table 3: Mortality rate of African catfish (*C. gariepinus)* juveniles exposed to varied dilution percentiles of effluents

	Duplicate 1							Duplicate 2						
Effluent type	Dilution percentile (effluent in water)	Mortality 12 h	24 h	48 h	72 h	96 h	Mortality ratio	Dilution percentile (effluent in water)	Mortality 12 h	24 h	48 h	72 h	96 h	Mortality ratio
Non-alcoholic	0% (control)	0	0	0	0	0	0/8	0% (control)	0	0	0	1	0	1/8
	5%	0	1	1	0	0	2/8	5%	0	0	0	1	1	2/8
	25%	0	1	1	0	1	3/8	25%	0	1	2	0	1	4/8
	50%	0	2	3	1	0	6/8	50%	0	1	1	2	0	4/8
	100%	0	2	2	1	1	6/8	100%	0	2	3	1	1	7/8

Table 3: Mortality rate of African catfish (*C. gariepinus)* juveniles exposed to varied dilution percentiles of effluents

	Duplicate 1							Duplicate 2						
Effluent type	**Dilution percentile (effluent in water)**	**Mortality**						**Dilution percentile (effluent in water)**	**Mortality**					
		12 h	**24 h**	**48 h**	**72 h**	**96 h**	**Mortality ratio**		**12 h**	**24 h**	**48 h**	**72 h**	**96 h**	**Mortality ratio**
Alcoholic	0% (control)	0	0	0	0	0	0/8	0% (control)	0	0	0	0	0	0/8
	5%	0	2	1	1	0	4/8	5%	0	1	2	0	2	5/8
	25%	1	0	2	1	2	6/8	25%	1	0	3	1	1	6/8
	50%	0	3	1	0	3	7/8	50%	2	2	1	0	1	6/8
	100%	0	2	1	1	2	6/8	100%	1	0	3	1	3	8/8
Abattoir	0% (control)	0	1	0	0	0	1/8	0% (control)	0	0	0	0	0	0/8
	5%	0	0	0	0	1	1/8	5%	0	0	0	3	0	3/8
	25%	0	0	1	3	1	5/8	25%	0	0	0	2	1	3/8
	50%	0	0	1	2	1	4/8	50%	0	0	3	1	2	6/8
	100%	0	0	1	1	2	4/8	100%	0	0	1	3	3	7/8

Number of fish in the effluent (n) = 8

Physicochemical evaluation versus acute toxicity assessment of the industrial effluents: The comparison of the quality of the effluents using the results of physicochemical parameters is quite at variance with the acute toxicity results. Scrutiny of the recorded values for the physicochemical parameters showed that the effluent from the alcohol-producing facility alone had a pH level (of 7.17±0.01) within the recommended set limits. The alcoholic industrial effluent equally recorded the lowest levels for EC, hardness, TSS, turbidity, ammonia, chloride, nitrates, phosphates, sulphates, as well as heavy metals, which may connote better quality in comparison to the non-alcoholic industry and abattoir effluents. However, the results from the acute toxicity evaluation of the water quality were in contrast with this assumption because more mortality (death) of the fish was recorded from the alcoholic industrial effluent. The highest mortality of 60% with the LC_{50} of 8.71 mg/L observed for the alcoholic industrial effluent denotes more environmental implications from the effluent.

The physicochemical results indicate that the alcoholic industrial effluent had the least DO concentration (2.50±0.10 mg/L), which was below the set lower limit of 4.0 mg/L recommended for the sustainability of aquatic organisms by NESREA (NESREA, 2011). Insufficient levels of DO in an aqueous system affect the breathing and behavioural responses and may lead to suffocation of the fish (Knight *et al*., 2013). The behavioural responses of fishes to low DO levels in aquatic ecosystems, as posited by Kramer (1987), usually entail "changes in activity, increased use of air-breathing, increased use of aquatic surface respiration, and vertical or horizontal habitat changes". These responses may translate to the restlessness and erratic movements, and gasping for breath that was observed in the acute toxicity assessment, which was more intense for the fish subjected to the alcoholic industrial effluent. Overtime time, the fish may grow weak from the overt movements leading to "sluggishness" in movement and eventual death.

Comparatively, the non-alcoholic and abattoir effluents were not too distinct from each other, as the recorded physicochemical values were quite close. However, based on the observed values, the non-alcoholic effluent may be adjudged to be of better quality as indicated by the Duncan Multiple Range Test. The non-alcoholic industrial effluent recorded a slightly higher mortality of 42.50% as compared to the 41.25% recorded for the abattoir effluent. In this scenario, the non-alcoholic industrial effluent had a higher DO level (6.19±0.08 mg/L) than the abattoir effluent (2.77±0.03 mg/L). This suggests that the DO levels may not be the sole reason for the recorded mortality in industrial effluents. Perhaps the complex mix of pollutants in the industrial effluents exerted the observed toxicological effects on the fish (Ahmed *et al*., 2022). More so, the possible presence of several other pollutants (or micropollutants) not assayed in this study can contribute to the overall observations and mortality of the fish in the different industrial effluents. These observations buttress the possible limitation of using the physicochemical and/or the elemental analysis as the criteria for effluent quality assessment. In this regard, a more holistic approach, which will encompass x-raying the physical, chemical, biological, as well as ecotoxicological properties of effluents, may be the best way to assess effluent quality. Hence, the incorporation of ecotoxicity assays into the effluent quality assessment regime will help to ascertain the possible impact of the effluents on the receiving water bodies (Akharame *et al*., 2022; Pereao *et al*., 2021).

Conclusion

A comparative acute toxicity assessment of industrial effluents from alcoholic, non-alcoholic, and abattoir facilities in Benin City, Nigeria, was carried out using the African catfish (*C. gariepinus*). The physicochemical results (78.79%) showed non-conformation with the standards set by the National Environmental Standards and Regulations Enforcement Agency (NESREA) for effluent discharge, while the levels of the heavy metals were majorly (76.67%) below the maximum permissible limits. The acute toxicity assessment carried out using the African catfish recorded the highest mortality for the alcoholic industrial effluent with a 96-h LC_{50} of 8.71 mg/L. The observed mortality showed alcoholic (60%) > non-alcoholic (42.50%) > abattoir (41.25%) effluents, whereas the effluent quality assessment result showed alcoholic > non-alcoholic > abattoir industrial effluents. Dissolved oxygen levels and the overall effect of the complex mix of pollutants in the industrial effluents may be major contributors to their toxicity to the fish. It is recommended that the toxicity assays be incorporated into routine effluent quality monitoring regimes to complement the physicochemical, elemental, and microbial evaluations for better assessment. The regulatory agency (NESREA) should intensify efforts in its monitoring and enforcement responsibilities to ensure that industrial facilities comply with guidelines set for effluent discharges.

Conflict of interest

The authors declare that they have no conflict of interest.

Ethical approval

All applicable international, national, and/or institutional guidelines for the care and use of animals were followed. This study followed the principles in the Declaration of Helsinki on the humane treatment of animals used in research (http://www.wma.net/en/30publications/10policies/a18/) and the principles in the AVMA guidelines for the euthanasia of animals (AVMA 2013).

Data availability statement

The authors confirm that the data supporting the findings of this study are available within the article.

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