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Gross Anatomical Based Whole Effluent Chronic Toxicity Testing of Noodles Processing Company Waste, Using Clariasgariepinus as a biomarker.

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ABSTRACT
Industrial effluents discharged into aquatic environments pose significant ecological and biological threats, particularly to aquatic species like Clariasgariepinus. This study investigates the chronic toxicity of effluents from a noodles food industry on selected environmental bio-indicator fish, Clariasgariepinus, focusing on the gross anatomical features of growth, survival, and condition factor (CF).The following known aquatic contaminants from food industries were selected as the effluent target chemical (TC) of concern: cadmium (Cd), copper (Cu), chromium (Cr), lead (Pb), nickel (Ni) and polyaromatic hydrocarbons (PAH). Effluent samples were analyzed for chemical constituents, and their effects on fish were assessed under varying concentrations (6.25%, 12.5%, 25%, 50%, and 100%).Results revealed that most effluent parameters, such as Cu, Cr and Ni, were within NESREA and USEPA limits, while Cu levels (0.037 mg/L) exceeded permissible thresholds, high-lighting significant contamination concerns. Survival rates and growth indices, including relative growth rate (RGR) and specific growth rate (SGR), declined markedly at higher effluent concentrations, with only 20% survival recorded at 100% effluent exposure.The findings highlight the toxicity and ecological risks posed by cadmium-contaminated effluents, emphasizing the need for improved effluent treatment and stricter regulatory enforcement. This study underscores the critical need for sustainable industrial practices to protect aquatic ecosystems and ensure biodiversity conservation.

Keywords: Gross anatomical, chronic toxicity testing,Clariasgariepinus, biomarkers

I. INTRODUCTION

Increased water consumption is a result of the evolution of numerous industrial sectors and expanded urbanization, as well as the formation of wastewater that is significantly diversified in nature, toxicity, and treatability. Food processing takes a huge amount of water, and the food industry/sector is the largest source of wastewater/effluent of all industrial activities. Because the strength of wastewater from food processing industries is generally high, it must be pre-treated before being discharged into the environment or reused [1].

Today, several countries face an acute shortage of water caused by pollution. According to Owa [2], water is considered polluted if there is a degradation in itsquality caused by the direct or indirect introduction of substances that alter its physicochemical and biological properties and interfere with its regular use. These substances having either a solid, liquid or gaseous origin have different effects on water depending on their amount, potential danger and fragility of the environmentwhere they are released [3]. The presence of compounds like lactose, whey proteins, nutrients, and fats in the wastewater contributes to unpleasant odours and also hampers the degradation process [4].

Industrial effluents are characterized by a diverse range of pollutants, including organic compounds, heavy metals, nutrients, suspended solids, chemical additives, and other toxic substances. The specific composition of effluents depends on the industrial processes and products involved. Effective management and treatment of these effluents are essential to mitigate their adverse effects on aquatic ecosystems and human health [5].Heavy metals are usually introduced into food processing plant effluent from the use of water in their waste treatment plants. Water is extensively used in industrial processes for various functions like cooling, heating, cleaning, dilution, processing, and product transport. However, much of this water is ultimately discharged as wastewater,

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often containing high levels of heavy metals. Discharging contaminated wastewater directly exposes aquatic life to the dangers of these heavy metals. Bioaccumulating these metals in the food chain can harm humans and animals who consume the water or contaminated fish. Mercury (Hg), lead (Pb), cadmium (Cd), arsenic (As), chromium (Cr), nickel (Ni), copper (Cu), zinc (Zn), iron (Fe), and manganese (Mn) are some of the major heavy metal pollutants from industrial effluents [6].

The effects of industrial effluents on aquatic life are diverse and severe. Chemical pollutants can cause immediate toxic effects, including fish kills and the death of other aquatic organisms. Long-term exposure to lower concentrations of pollutants can lead to chronic health issues, such as reproductive problems, endocrine disruption, and impaired growth. For instance, heavy metals can interfere with enzyme functions, disrupt hormone systems, and damage vital organs in fish [7]. The physical parameters of water relate to the appearance of water (colour, turbidity, taste, odour and temperature) [8]. In order to be fit for use, water must be free of all impurities that are offensive to the sense of smell, sight and taste [9]. Such physical parameters include colour, turbidity, taste and odour, temperature e.t.c.

The African catfish, Clariasgariepinus, is a prominent species in the Choba River and is widely used in toxicological studies due to its hardiness and high resistance to pollutants. However, prolonged exposure to sub-lethal concentrations of pollutants, such as those found in industrial effluents, can lead to chronic toxicity, which may not be immediately lethal but can cause significant physiological and anatomical features over time [10]. C. gariepinus has been used to assess the environmental impact of pollutants through standardized toxicity tests. These tests typically involve exposing the fish to different concentrations of a pollutant under controlled laboratory conditions and observing its survival, behavior, and physiological changes over time. By analyzing dose-response relationships, researchers can determine the lethal and sub-lethal concentrations of a pollutant and establish safety thresholds for aquatic life. The ability of C. gariepinus to tolerate a wide range of environmental conditions allows it to be used in both acute and chronic toxicity tests. For instance, studies have evaluated the impact of heavy metals, such as lead and cadmium, on the behavior and mortality of C. gariepinus. Chronic toxicity studies have also focused on sub-lethal effects, such as reduced growth rates, reproductive failure, and histopathological changes in vital organs [11]. These tests contribute to regulatory frameworks for managing pollutant levels in water bodies and protecting aquatic biodiversity.

While general studies on industrial effluents have documented their harmful effects on aquatic organisms, there is a lack of detailed research focused specifically on the effluents from noodles food industry. Previous studies have largely focused on broader categories of food processing industries or specific pollutants but have not extensively examined how the unique composition of noodles processing effluents affects local aquatic species such as Clariasgariepinus.

The study was conducted to provide insight into the potential long-term effects of industrial effluents on the reproductive health of fish, with a focus on understanding how chronic exposure to these contaminants affects the gross anatomical features of survival, growth and condition factors (CF) Clariasgariepinus. This research is crucial in developing strategies for monitoring and mitigating the impact of industrial activities on aquatic ecosystems, ensuring the sustainability of fish populations, and maintaining the ecological balance of rivers like the New Calabar River (NCR).

II. MATERIALS AND METHOD

Study Type

The study type is a short-term chronic whole effluent toxicity laboratory testing in receiving waters of freshwater organism for aduration of 30 days (USEPA, 2002). This guideline describes chronic toxicity tests for use in the National Pollutant Discharge Elimination System(NPDES) Permits Program to identify effluents and receiving waters containing toxic materials in chronically toxic concentrations.This Standard allows for the use of the following:

- **Static renewal:** The test organisms are exposed to a fresh solution of the same concentration of sample every 24 hours or other prescribed interval, either by transferring the test organisms from one test chamber to another, or by replacing all or a portion of solution in the test chambers.
- **Key Outcome**: The key outcomes measured in this study were gross anatomical changes of mortality, survival, growth and CF observed in C. gariepinus.
- **Validity of Test:** For the conditions of validity, ISO (1994) and USEPA (2002)

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conditions for the validity of test were adopted for this study: The mortality in the controls should not exceed 10% at the end of test. The dissolved oxygen concentration should be at least 60% of the air saturation value throughout the test. In semi-static procedures, aeration can be used, provided it does not lead to a significant loss of test substance. There should be evidence that the concentration of the substance being tested has been satisfactorily maintained (it should be at least 80% of the nominal concentration) over the test period. The results should be based on measured concentration if the deviation from the nominal concentration is greater than 20%

Study Design

The study employed an experimental design involving the exposure of Clariasgariepinus to different concentrations of industrial effluents. Fish were divided into experimental groups subjected to varying effluent concentrations (6.25%, 12.5%, 25%, 50%, and 100%) and a control group maintained in clean water (USEPA, 2002). The experiment was carried out for a 30-day duration, ensuring consistent environmental conditions such as temperature, pH, and aeration.

Sampling

Effluent Sampling: The industrial effluent used in this study was collected directly from the discharge point of the industry, ensuring that it represented the actual waste being released into the surrounding environment. Collection was done using clean, non-reactive containers to prevent contamination. The samples were then transported to the laboratory under controlled conditions, where they were stored at low temperatures to preserve their chemical integrity until further analysis and use in the experimental setup.

Fish Sample Collection: The fish used in this study were Clariasgariepinus fingerlings, obtained from African Regional Aquaculture Centre (ARAC). ARAC is a reputable government owned fish hatchery, located in Aluu, Port Harcourt, Rivers State. Ealy life stage (fingerlings) were used for this study. They had an average weight of 0.75g, making them suitable for the toxicological experiment. Upon collection, the fingerlings were acclimatized in clean, aerated water for a period of two weeks before being introduced to the experimental conditions. This acclimatization period allowed the fish to stabilize physiologically, reducing the impact of transport and handling stress on the study's outcomes. The fingerlings were then randomly assigned to the different experimental groups, each exposed to varying concentrations of the effluent as described in the research design.

Evaluation

Chemical Analysis: To understand the chemical composition of the effluents used in this study, samples were analyzed for target parameters: Cd, Cr, Cu, Pb, Ni and PAH. These parameters were compared against the standards set by NESREA (National Environmental Standards and Regulations Enforcement Agency) and USEPA (United States Environmental Protection Agency) effluent guidelines to assess compliance.

Fish Survival Rate: The survival rate of Clariasgariepinus exposed to different effluent concentrations was monitored throughout the study. Initial and final weights, mortality rates, and survival percentages were recorded. The formulas used include:

$$\text{Relative Growth Rate (RGR)} = \left(\frac{W_1 - W_0}{t}\right) \times \frac{100}{W_0}$$

$$\text{Specific Growth Rate (SGR)} = \frac{W_1 - W_0 \times 100}{t}$$

$$\text{SurvivalRate} = \left(1 - \frac{\text{Numberofdeaths}}{\text{FishstockConcentratio}}\right) \times 100$$

Where;

- **Wo:** Initial weight,
- **W1:** Final weight, **t:** duration of experiment.
- **SGR,** Specific Growth Rate is a coefficient that measures the percentage increase in fish weight per day. It was gotten by subtracting the initial weight from the final weight, multiplied by 100 and dividing it by the number of days of the experiment.

Condition Factor: The condition factor (CF) was used to assess the gross anatomical health of Clariasgariepinus. Total length and weight of the fish were measured, and CF was calculated using Fulton's formula:

$$CF = \frac{\text{weig ht (g)} \times 10^5}{\text{lengt h (cm}^3)}$$

Data Analysis

Data collected from the study were statistically analyzed using ANOVA to compare the effects of effluent concentrations on survival rates, growth parameters, and CF. Significance levels were set at $p<0.05$, and results were presented as mean ± standard

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III. RESULTS

Table 1 Chemical constituents of Effluent used for the study

S/N	Parameter(s) Mg/l	Conc(%)	National standard (NESREA)	International standard (USEPA)	Remarks
1	PAH	<0.001	0.10	0.03	Below the standard, hence it is safe.
2	COPPER, Cu	0.079	1.2	1.3	Below the standard, hence it is safe.
3	LEAD, Pb	<0.001	0.1	0.015	Below the standard, hence it is safe.
4	CADMIUM, Cd	0.037	0.003	0.015	Above the standard, hence it is toxic.
5	CHROMIUM, Cr	<0.001	0.1	0.1	Below the standard, hence it is safe.
6	NICKEL, Ni	<0.001	0.5	0.1	Below the standard, hence it is safe.

The table provides a comparative analysis of key chemical parameters in the effluent, measured against the NESREA and USEPA standards:

- **PAH (<0.001%)**: Well below the NESREA (0.10) and USEPA (0.30) standards, indicating safety.
- **Copper (0.079 mg/L)**: Within permissible limits of both NESREA (1.2 mg/L) and USEPA (1.3 mg/L), making it safe.
- **Lead (<0.001 mg/L)**: Safe under NESREA (0.1 mg/L) but exceeding the stricter USEPA standard (0.015 mg/L).
- **Cadmium (0.037 mg/L)**: Exceeds both NESREA (0.003 mg/L) and USEPA (0.015 mg/L) thresholds, indicating toxicity.
- **Chromium and Nickel (<0.001 mg/L each)**: Well within the limits for both NESREA (0.1 mg/L for Chromium, 0.5 mg/L for Nickel) and USEPA (0.1 mg/L for both), making them safe.

The results show cadmium levels as the primary concern for aquatic toxicity, necessitating stricter effluent management strategies.

Table2 ; Showing The Fish Survival Rate

EFFLUENT Conc.(%)	**W0(g) Initial weight**	**W1(g) Final weight**	**RGR**	**SGR PER DAY %**	**Number of deaths.**	**FSR (%) Survival rate**
100	**0.83**	**10.82**	**1302.4**	**1081.0**	**3**	**60**
50	**0.51**	**6.82**	**1335.2**	**681.4**	**3**	**60**
25	**0.75**	**8.23**	**1096.0**	**822.2**	**2**	**40**
12.5	**0.63**	**6.25**	**990.4**	**624.3**	**2**	**40**
6.25	**1.00**	**13.72**	**1370.0**	**1370.8**	**1**	**20**

Keys: TC: Tank conc; **Wo:** initial weight;**W1:** final weight;**RGR:** relative growth rate; **SGR:** specific growth rate; **FSR:** fish survival rate; **RGR,** Relative Growth Rate is the rate of growth with respect to the initial size.

This table presents survival data of Clariasgariepinus under varying effluent concentrations:

- **RGR (Relative Growth Rate)** and **SGR (Specific Growth Rate)** decline significantly with increasing effluent concentration.

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- The survival rate drops to **20% at 100% effluent concentration**, indicating severe toxicity.
- **Mortality rates** correlate with effluent toxicity, reinforcing the acute effects of heavy metal contamination, particularly cadmium.

Table1: Descriptive statistics in the measured parameters for Treated Tanks and Control

Variables	Tank 1 6.25% conc.	Tank 2 12.5% conc.	Tank 3 25% conc.	Tank 4 50% conc.	Tank 5 100% conc.	Control
LENGTH	0.31	0.33	0.13	0.14	0.30	0.15
WEIGHT	0.42	0.48	0.19	0.21	0.49	0.24
CF	1.40	1.50	1.50	1.50	1.60	1.60

Growth parameters and the condition factor (CF) demonstrate how effluent concentrations affect fish health:

- **Weight and length gains** are substantially reduced in higher effluent concentrations, consistent with impaired physiological functions.
- **Condition factor (CF)** shows minor variations (1.40–1.60), suggesting that while general body proportions remain stable, other growth indices like weight are adversely impacted in polluted tanks.

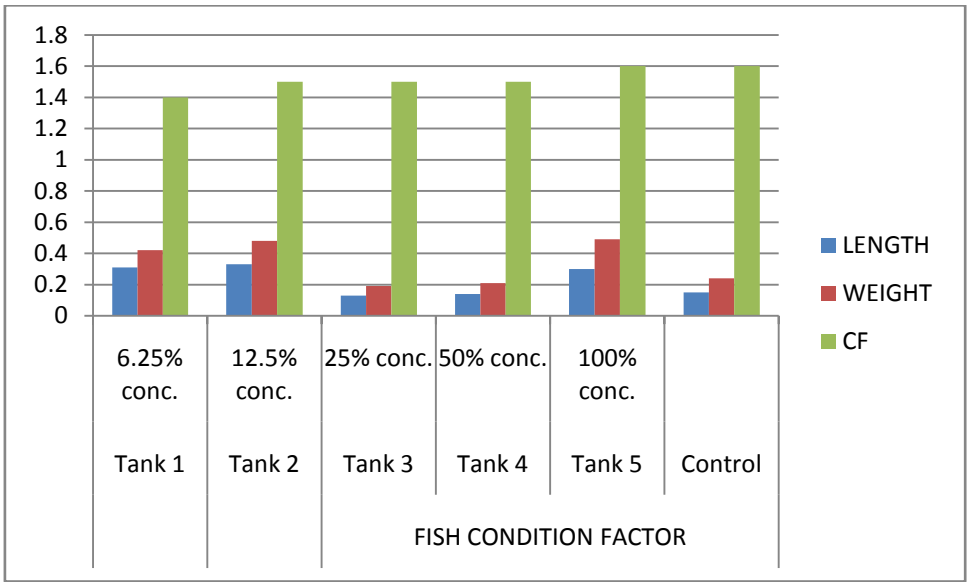

Figure1 Bar Chart showing the fish condition factor distribution of each tanks

The bar chart illustrates:

- A relatively stable CF across tanks, with slight declines in higher effluent concentrations.
- This indicates that while CF is less sensitive to effluent impacts, it alone cannot fully depict toxicity levels, which align better with mortality and histological data.

IV. DISCUSSION

This study investigated the chronic toxicity of effluents from the Indomie food industry on the male gonads of Clariasgariepinus, focusing on physiological, growth, survival, and CF.

Chemical profile of the effluent showed that Most of the tested TC; , Cu, Cr and Ni,, were

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within permissible limits under NESREA and USEPA standards. Cadmium levels (0.037 mg/L) exceeded both NESREA (0.003 mg/L) and USEPA (0.015 mg/L) standards, posing a significant toxicological risk.

The effluent impact ongrowth and survival showed that RGR and SGR declined as effluent concentration increased. A sharp decline in survival rate was observed at higher effluent concentrations, with only 20% survival at 100% effluent concentration. This data underscores the acute and chronic impacts of effluent toxicity on Clariasgariepinus. The steady decline in RGR and SGR, coupled with the high mortality rates at higher effluent concentrations, demonstrates the direct physiological stress imposed by the contaminants. This aligns with previous findings that industrial effluents, particularly those rich in heavy metals, impair metabolic activities and compromise survival (Akaninwor et al., 2007). The results indicate that sub-lethal concentrations also disrupt growth patterns, potentially impairing fish development and ecological stability over time.

CF remained relatively stable across all groups, its limitations as a sole toxicity indicator are evident. The minimal CF variations, even at higher concentrations, contrast sharply with the significant declines in survival and growth rates. This suggests that CF might mask underlying physiological and histological damage, reinforcing the need for more sensitive biomarkers, as shown in histopathological analytical studies.

The study would recommend the following:

- Effluent treatment and management:The noodles food industry should adopt advanced effluent treatment technologies, such as activated carbon filtration or chemical precipitation, to reduce cadmium levels below regulatory thresholds.
- Regular monitoring of effluent discharge should be enforced by regulatory bodies like NESREA and NOSDRA (National Oil Spill Detection and Response Agency) to ensure compliance with permissible standards.
- Aquatic ecosystem monitoring:Regular biomonitoring using sensitive tools like histological assessment should be conducted to assess the ecological impact of industrial effluents.
- Community awareness and engagement:Awareness programs should be conducted to educate local communities on the risks of water contamination and promote sustainable practices.Collaborations between industries and researchers should be encouraged to develop eco-friendly manufacturing practices.
- Policy implementation**:**Stricter penalties should be imposed for non-compliance with effluent standards to deter industrial pollution.
- Effluent discharge zones should be designated away from critical aquatic habitats to minimize ecological impact.

V. CONCLUSION

This study underscores the severe environmental and biological consequences of industrial effluent discharge into aquatic ecosystems. Elevated cadmium levels in the effluent from the noodles processing company were identified as a key contributor to physiological and gross anatomical health featuresofClariasgariepinus. These findings reveal that toxicity threats to growth and survival are posed by untreated or inadequately treated effluents.The study highlights the urgent need for enhanced effluent management practices, stricter regulatory standards, and continuous environmental monitoring to mitigate these impacts. Protecting aquatic biodiversity and ensuring sustainable industrial practices require a concerted effort from industries, regulatory bodies, and local communities. Implementing the recommended measures will not only safeguard aquatic life but also protect the livelihoods of communities relying on these ecosystems.

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