Histopathology Of Three Common Economically Important Marine Fish Species from Selected Estuaries in The Gulf of Guinea for Sustainable Fisheries Management: A Baseline Study for Anthropogenic Censorship

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***Abstract*—This study assessed the histopathological condition of three commercially important marine fish species, *Ilisha africana*, *Cynoglossus senegalensis*, and *Scomberomorus tritor*, from the Benin, Brass, and Escravos estuaries of the Gulf of Guinea, Nigeria, and evaluated selected water quality parameters to establish baseline information for sustainable fisheries management and environmental monitoring. A total of eighteen (18) liver and gonad tissue samples were collected and examined using standard histological techniques, while water quality parameters including temperature, dissolved oxygen, pH, salinity, turbidity, total suspended solids, and nitrate concentration were measured. Histopathological analysis revealed normal liver and gonadal architecture in *I. africana* and *S. tritor* across all estuaries, with no detectable lesions and Histopathological Alteration Index (HAI) and Gonadal Alteration Index (GAI) values of zero. In contrast, *C. senegalensis* exhibited mild hepatic and gonadal alterations, including sinusoidal congestion, cytoplasmic vacuolation, and slight germ-cell degeneration. These lesions occurred at a frequency of 33.3%, with mean lesion scores of 1.33 ± 1.15 and HAI and GAI values of 4, indicating mild tissue stress without significant impairment of organ function. Water quality conditions were generally favourable for fish survival and growth, with temperature ranging from 26.83–27.42°C, dissolved oxygen from 6.08–6.13 mg L^{-1}, pH from 7.32–7.88, salinity from 21.75–24.92 ppt, turbidity from 12.67–17.50 NTU, total suspended solids from 35.67–46.67 mg L^{-1}, and nitrate concentrations from 1.60–1.85 mg L^{-1}. Although all parameters were within acceptable ecological limits, the Escravos estuary recorded relatively higher salinity levels, suggesting greater marine influence. The findings indicate that the three estuaries currently support healthy fish populations; however, the mild tissue alterations observed in *C. senegalensis* suggest exposure to low-level environmental stressors and highlight its potential as a bioindicator species. This study provides important baseline data for environmental assessment, fisheries conservation, and sustainable management of estuarine resources in the Gulf of Guinea.**

***Index Terms*—Histopathology, Fish Health, Water Quality, Estuaries, Bioindicator Species, Sustainable Fisheries Management, Gulf of Guinea.**

I. INTRODUCTION

Estuarine ecosystems, positioned at the confluence of freshwater rivers and marine coastal environments, represent some of the most dynamic and productive ecological zones on the planet (Blaber *et al*., 2022). These habitats provide vital nursery grounds, feeding sites, and migration pathways for a wide variety of aquatic species, including commercially and ecologically significant fish (Douglas *et al*., 2022). Within the West African region, the estuaries of the Gulf of Guinea are especially crucial for maintaining the biodiversity and productivity of fisheries that support millions of people with food, employment, and socio-economic benefits (Raoult *et al*., 2022; Cooke *et al*., 2023).

Three key fish species that dominate the estuarine fisheries of the Gulf of Guinea are *Ilisha africana* (African ilisha), *Cynoglossus senegalensis*

(Senegalese tongue sole), and *Scomberomorus tritor* (West African Spanish mackerel). These species are not only abundant but also form a significant part of the artisanal and industrial fisheries in countries like Nigeria, Ghana, and Ivory Coast (Essien-Ibok *et al*., 2024; Amponsah *et al*., 2023).

The sustainability of estuarine fisheries in West Africa is being undermined by multiple anthropogenic pressures, including pollution, habitat degradation, overexploitation, and climate-related changes in salinity, temperature, and dissolved oxygen (Broadley *et al*., 2022; Abd El-Hack *et al.*, 2022). These stressors not only reduce fish productivity but also affect the health and fish populations, potentially leading to population fragmentation, reduced resilience, and loss of adaptive potential (Galappaththi *et al*., 2022; Petit-Marty *et al*., 2022).

Furthermore, histological evaluations of fish organs such as gills, liver, and gonads offer insight into physiological responses to pollutants, disease, and other environmental stressors (Eissa et al., 2025; Dawood *et al*.,2022). These microscopic examinations can reveal cellular damage, inflammation, or reproductive anomalies that are not visible externally but may significantly impact fish health, reproductive success, and population viability (Guerrera *et al*., 2021; De Marco *et al*., 2023). Histopathological changes have been widely used as biomarkers for environmental monitoring, allowing researchers to link environmental quality to biological outcomes (Catteau *et al*., 2021; Tegomo *et al*., 2021).

This study is thus poised to contribute valuable insights into the population dynamics, water quality resilience and health status of three ecologically significant fish species. It provides a scientific foundation for improved fisheries governance, particularly in regions where biological data and monitoring systems remain limited (Gebremedhin *et al*., 2021; Edmondson and Fanning, 2022).

II. MATERIALS AND METHODS

2.1.Description of study area

This study was conducted in three major estuarine systems along the Nigerian Atlantic coast: the Benin Estuary, Escravos Estuary, and Brass Estuary. The selected estuaries were chosen for their ecological relevance, varying degrees of anthropogenic impact, and strategic locations across the Niger Delta. Each estuary exhibits unique features, water chemistry, and human influence, offering a comparative basis for understanding spatial variability and how it affects fish phenotype, health status, and morphological characteristics. The study area is located within the coastal Niger Delta region bordering the Gulf of Guinea and lies approximately between latitudes 1°00′00″ N and 3°00′00″ N and longitudes 1°00′00″ E and 3°00′00″ E

Figure 1: Geolocation of the Map showing Escravos, Benin and Brass Estuaries in the coastal Niger Delta region bordering the Gulf of Guinea

2.2.Biological Data Collection

Accurate collection of biological data is essential for evaluating water quality indicators and species health in fish ecology and fisheries management.

2.1.1.Samples Preservation and Identification

For histological assessments, organs of interest namely the liver, and gonads were surgically excised and placed in 10% neutral buffered formalin for fixation, before taking to Eliezer Analytical laboratory Asaba.for histological analysis. Each sample was labeled with species code, site code, sex, and date of collection to ensure traceability. The combination of standardized measurement, reliable preservation, and accurate identification enhances the validity of the biological data and allows meaningful interpretation of morphological, genetic, and histological patterns across the three estuarine environments.

2.3.Histological Assessment

Histological analysis is a crucial method in fish health assessment, offering direct insight into tissue-level changes that reflected exposure to environmental stressors such as pollutants, hypoxia, and nutritional deficits (Guerrera *et al.*, 2021; Eissa et al., 2025). This component of the study evaluated the microscopic structure of the liver, and gonads of *I. africana*, *C. senegalensis*, and *S. tritor* collected from the Benin, Escravos, and Brass estuaries. These organs were selected due to their essential physiological roles and their documented sensitivity to environmental alterations.

The goal of the histological assessment was to detect pathological alterations such as necrosis, hyperplasia, inflammation, cellular degeneration, and reproductive impairment conditions that can compromise fish survival, fitness, and reproductive output. These assessments also aimed to correlate histopathological changes with water quality parameters and genetic diversity patterns observed across the three estuarine ecosystems.

2.3.1.Tissue Sampling (Liver and Gonads)

Immediately after morphometric measurement and molecular tissue extraction, each selected fish samples were dissected under sterile conditions for internal organ retrieval. Dissections were performed using sterilized surgical instruments in a clean laboratory setting to prevent contamination and degradation of tissues. The target organs, liver, and gonads were carefully excised and placed individually in properly labeled containers.

2.3.1.1. Liver tissues were removed by sectioning a small piece (~1 cm^3) from the central hepatic lobe.

The liver plays a key role in detoxification, metabolism, and immune response, making it a primary target for histopathological studies (Guerrera *et al*., 2021; Catteau et al., 2021).

2.3.1.2. Gonads were sampled depending on the sex of the fish.

A segment from either testis or ovary were collected, as gonadal tissue reveals reproductive health, maturity stages, and endocrine disruption signals.

Tissue samples were promptly fixed in 10% neutral buffered formalin (NBF) for 48 hours to preserve cellular architecture and prevent autolysis. After fixation, tissues were transferred to 70% ethanol for long-term storage prior to processing. Each container was labeled with specimen ID, tissue type, estuary, and date of collection.

A total of 18 tissue sets (Liver and Gonad) were prepared across the three estuaries. Subsamples were stratified by species and location to ensure balanced representation in subsequent histological processing and analysis.

2.3.2.Fixation, Sectioning, and Staining Procedures

Fixed tissues were processed for histological examination using standard paraffin embedding techniques, following internationally accepted protocols. The procedure involved the following steps:

Dehydration:

Tissues were passed through a graded ethanol series (70%, 80%, 95%, and 100%) for 1 hour each to remove water content.

Clearing:

Ethanol was replaced with xylene to clear the tissue and prepare it for paraffin infiltration.

Embedding:

Cleared tissues were embedded in molten paraffin wax using steel molds and cooled to form blocks suitable for sectioning.

Sectioning:

Thin tissue sections (5–7 µm thick) were cut using a rotary microtome and mounted on glass slides pre-treated with poly-L-lysine to ensure adhesion.

For staining, the Hematoxylin and Eosin (H&E) protocol was employed, which highlights cellular morphology by staining nuclei dark blue and cytoplasmic components pink (Eissa *et al.,* 2025; Guerrera *et al.*, 2021). H&E is widely used in fish histology due to its contrast, versatility, and diagnostic reliability across tissues and lesion types.

Stained slides were examined under a light microscope (Leica DM500) at magnifications ranging from 40× to 400×. Photomicrographs were captured using a digital camera system (Leica ICC50 HD) and analyzed for histopathological lesions. Lesions were evaluated and scored using a semi-quantitative grading system adapted from previous studies (Abd El-Hack *et al*., 2022; Tumwesigye *et al*., 2022), 2.4. Water Quality Analysis

Water quality plays a fundamental role in shaping the health, physiological performance, and genetic diversity of aquatic organisms, especially in estuarine ecosystems that are characterized by high variability and vulnerability to anthropogenic disturbances (Guerrera *et al.*, 2021; Otogo *et al.*, 2022). Fish species such as *I. africana*, *C. senegalensis*, and *S. tritor* are particularly sensitive to fluctuations in salinity, dissolved oxygen (DO), temperature, and other physicochemical variables, which can induce stress, disrupt reproductive cycles, and alter population dynamics (Blaber *et al*., 2022; Tumwesigye *et al.*, 2022).

Therefore, a comprehensive water quality monitoring program was incorporated into this study to evaluate environmental conditions at each sampling site and to correlate habitat quality with morphological, genetic, and histological outcomes. Parameters measured included temperature, salinity, pH, dissolved oxygen (DO), conductivity, turbidity, nitrate and biochemical oxygen demand (BOD) all of which are essential indicators of ecological health in estuarine habitats (Essien-Ibok *et al*., 2024; Abd El-Hack *et al*., 2022).

2.4.1. Collection of Water samples Sampling Protocols

Water samples were collected concurrently with fish sampling to ensure temporal and spatial consistency. At each of the three estuaries Benin, Escravos, and Brass

Seasonal sampling was done during the dry season (November–March) and wet season (April–October) to capture the effects of rainfall, freshwater inflow, and tidal mixing.

Surface water was collected at approximately 0.5-metre depth using clean, acid-washed polyethylene bottles (1 liter) for laboratory analysis, while in-situ parameters were measured directly at the sampling sites. All sample containers were labeled with location, date, time, and environmental notes, and were transported in cool boxes to the laboratory within 6 hours of collection (Guerrera *et al.*, 2021; Fernandes *et al.*, 2021). To ensure accuracy, all equipment were calibrated daily before use, and each parameter were measured in triplicate.

2.4.2. In-situ Analysis (pH, DO, Temp, Salinity)

In-situ measurements were taken using a portable multi-parameter water quality probe HANNA HI98194 capable of simultaneously measuring temperature, pH, salinity, dissolved oxygen, and conductivity.

2.4.2.1. Temperature (°C)

It was recorded in real-time, as it affects metabolic rate, oxygen solubility, and reproductive timing in fish (Abd El-Hack *et al.*, 2022; Tumwesigye *et al.*, 2022).

2.4.2.2. Hydrogen Ion Concentration (pH)

Levels were measured to assess water acidity or alkalinity. Optimal ranges for estuarine fish are typically 6.5–8.5, with deviations indicating potential pollution or acidification events (Eissa *et al.*, 2025; Guerrera *et al*., 2021).

2.4.2.3. Salinity (ppt)

Was monitored using the conductivity function, and values were adjusted based on temperature and site-specific tidal influence (Blaber *et al.*, 2022; Petrou *et al.*, 2021).

2.4.2.4. Dissolved Oxygen (mg/L)

Was measured to assess the concentration of oxygen in the water water, a key factor influencing fish respiration, survival, and stress response (Essien-Ibok *et al.*, 2024; Galappaththi *et al.*, 2022).

2.4.2.5. Turbidity (NTU)

Was recorded using a handheld nephelometer, as increased turbidity reduces light penetration and can impair gill function or egg survival (Otogo *et al*., 2022; Cooke *et al.*, 2023).

2.4.3. Laboratory Analysis of Water Samples

Water samples collected from each sampling station were carefully transferred into pre-cleaned polyethylene bottles, properly labelled, and transported to the Microbiology Department laboratory DELSU. Laboratory analyses were conducted using standard procedures recommended by the American Public Health Association (APHA, 2023).

2.4.3.1. Total Suspended Solids (TSS) (mg/L)

Total suspended solids were determined using the gravimetric method. A measured volume of water was filtered through a pre-weighed glass fibre filter paper to retain suspended particles. The filter paper was then dried in an oven at 103–105°C until a constant weight was achieved. The difference in weight before and after filtration represented the amount of suspended material present in the sample and was expressed as milligrams per litre (mg/L).

2.4.3.2. Nitrate (NO_3^-) Concentration (mg/L)
Nitrate concentration was determined using a spectrophotometric method. Water samples were treated with appropriate reagents to produce a measurable colour reaction proportional to the nitrate concentration present. Absorbance readings were obtained using a UV-visible spectrophotometer, and nitrate concentrations were calculated from standard calibration curves. Nitrate is an important nutrient in aquatic systems, and elevated concentrations may indicate agricultural runoff, domestic waste inputs, or eutrophication.
All values were compared against Nigerian Federal Ministry of Environment (FMEnv) water quality standards and WHO guidelines to assess compliance and ecological risk (Essien-Ibok *et al*., 2024; Fernandes *et al.*, 2021).

2.5. Data Analysis
In this research, integrated data sets from histological, and environmental assessments were subjected to detailed statistical treatment using Software packages used such as SPSS v27, each selected for its appropriateness to specific data types (Jeena *et al.*, 2022; Petrou *et a*l., 2021).

2.5.1. Multivariate and Correlation Analyses
To integrate biological, genetic, histological, and environmental data, a series of multivariate and correlation analyses were conducted:

- Pearson’s or Spearman’s correlation coefficients were computed to assess relationships between water quality parameters and histopathological scores or morphometric indices (Eissa *et al*., 2025; Tumwesigye *et al.,* 2022).
- Canonical Correspondence Analysis (CCA) was used to identify environmental gradients influencing biological responses across species and estuaries (Galappaththi et al., 2022).
- Multivariate Analysis of Variance (MANOVA) tested the combined effect of estuary and season on multiple dependent variables including genetic indices, lesion scores, and morphometric traits.
- Hierarchical Cluster Analysis (HCA) grouped populations based on overall similarity in genetic, morphometric, and histopathological profiles.
- Generalized Linear Models (GLMs) were employed to determine predictors of tissue damage and genetic variation.

III. RESULTS

This section presents the findings from the study in a clear and organized way, based on the objectives set out earlier. It covers the water quality conditions of the Benin, Brass, and Escravos estuaries observed in the liver and gonads of the sampled fish ofn the histopathological changes in *I, africana*, *C. senegalensis*, and *S. tritor.*

3.1. Histological Assessment of Liver and Gonad Tissues
Microscopic examination of both the liver and gonad tissues (Tables 4.14 and 4.15) shows that, in general, the fish from the three estuaries Benin, Escravos, and Brass, were in good physiological condition. Most of the species maintained normal tissue structure, although *Cynoglossus senegalensis* consistently showed mild signs of stress compared to the other two species.

3.1.1. Liver Histology
In *I. africana* and *S. tritor*, the liver tissues appeared completely normal across all locations. The liver cells were well arranged, the nuclei were intact, and the sinusoidal spaces looked healthy and clear. There were no signs of damage such as inflammation, tissue breakdown, or degeneration. This suggests that the liver function of these fish was not negatively affected by their environment.
On the other hand, *C. senegalensis* showed mild changes in the liver in all the estuaries. These changes were mainly slight congestion in the sinusoids and small areas of vacuolation in the liver cells. Even with these changes, the cell nuclei remained intact, and there was no evidence of serious damage. The lesion score was low (1.33 ± 1.15), and the Histopathological Alteration Index (HAI) was 4 in all cases. Since this value is still within the normal range, it means the liver function was not seriously affected, although this

species appears more sensitive to environmental conditions.

3.1.2 Gonad Histology

A similar pattern was observed in the gonads. *Ilisha africana* and *Scomberomorus tritor* showed healthy reproductive tissues in all estuaries. Their gonads were well developed, with clearly visible and normal germ cells, showing no signs of damage. This indicates good reproductive health across the study area.

In contrast, *Cynoglossus senegalensis* again showed mild reproductive stress. There were slight signs of germ cell degeneration and minor disorganization of the gonadal structure in all estuaries. However, the overall structure of the gonads was still intact, and reproductive function does not appear to be severely affected. The GAI value of 4 also indicates only mild alteration.

When both liver and gonad results are considered together, a clear pattern emerges. *Ilisha africana* and *Scomberomorus tritor* show consistently healthy tissues with no visible damage across all estuaries, suggesting stable physiological conditions. However, *Cynoglossus senegalensis* shows mild but repeated signs of stress in both organs.

Importantly, the absence of severe tissue damage in all species suggests that the estuarine environments are not heavily polluted or degraded. The mild changes observed, especially in *C. senegalensis*, likely reflect slight environmental stress rather than serious ecosystem problems.

Overall, the fish populations across the Benin, Escravos, and Brass estuaries are generally healthy, with only minor physiological stress observed in one species, indicating that the ecosystems are still largely supportive of fish survival and reproduction.

Table1: Liver Histopathological Characteristics and Histopathological Alteration Index (HAI) of Fish Species from Benin, Escravos, and Brass Estuaries

Estuary	Species	n	Histological Characteristics	Lesions Observed (%)	Mean Lesion Score ± SD	HAI	Histological Interpretation
Benin	*I. africana*	1	Normal hepatocyte arrangement with intact nuclei and well-defined sinusoidal spaces	No lesions detected (0%)	0.00 ±0.00	0	Normal liver architecture
Benin	*C. senegalensis*	1	Mild sinusoidal congestion and focal cytoplasmic vacuolation with preserved nuclear integrity	Sinusoidal congestion (33.3%); cytoplasmic vacuolation (33.3%)	1.33 ±1.15	4	Mild hepatic alteration with preserved function
Benin	*S. tritor*	1	Normal liver structure with well-arranged hepatocytes and intact nuclei	No lesions detected (0%)	0.00 ±0.00	0	Normal liver condition
Escravos	*I. africana*	1	Well-preserved hepatic architecture with normal hepatocytes and sinusoidal spaces	No lesions detected (0%)	0.00 ±0.00	0	Normal liver structure
Escravos	*C. senegalensis*	1	Mild hepatic congestion and vacuolation observed in some hepatocytes	Sinusoidal congestion (33.3%); cytoplasmic vacuolation (33.3%)	1.33 ±1.15	4	Mild liver stress
Escravos	*S. tritor*	1	Normal hepatocyte organization with no structural abnormalities	No lesions detected (0%)	0.00 ±0.00	0	Normal hepatic condition

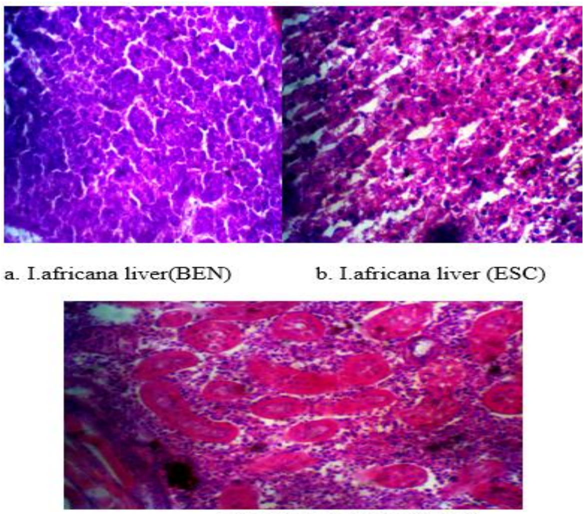
Estuary	Species	n	Histological Characteristics	Lesions Observed (%)	Mean Lesion Score ± SD	HAI	Histological Interpretation
Brass	*I. africana*	1	Normal liver histoarchitecture with intact cellular arrangement	No lesions detected (0%)	0.00 ±0.00	0	Healthy liver condition
Brass	*C. senegalensis*	1	Mild vacuolation and sinusoidal congestion observed	Sinusoidal congestion (33.3%); cytoplasmic vacuolation (33.3%)	1.33 ±1.15	4	Mild hepatic alteration
Brass	*S. tritor*	1	Normal liver tissue with intact hepatocytes and sinusoidal spaces	No lesions detected (0%)	0.00 ±0.00	0	Normal liver architecture

Note: Histopathological Alteration Index (HAI) was calculated as the sum of lesion scores recorded in liver tissues. HAI values between 0 and 10 indicate normal organ function with only slight tissue alterations.

Table2: Gonadal Histopathological Characteristics and Gonadal Alteration Index (GAI) of Fish Species from Benin, Escravos, and Brass Estuaries

Estuary	Species	n	Histological Characteristics	Lesions Observed (%)	Mean Lesion Score ± SD	GAI	Histological Interpretation
Benin	*I. africana*	1	Gonads well developed with normal germ cell arrangement and intact reproductive cells	No lesions detected (0%)	0.00 ± 0.00	0	Normal gonadal condition
Benin	*C. senegalensis*	1	Mild germ cell disorganization with slight degeneration of reproductive tissue	Germ cell degeneration (33.3%)	1.33 ± 1.15	4	Mild gonadal stress
Benin	*Scomberomorus tritor*	1	Normal gonadal structure with well-defined germ cells at different stages	No lesions detected (0%)	0.00 ± 0.00	0	Normal reproductive condition
Escravos	*I. africana*	1	Normal gonadal architecture with intact oocytes/spermatogenic cells	No lesions detected (0%)	0.00 ± 0.00	0	Normal gonadal condition
Escravos	*C. senegalensis*	1	Slight tissue disorganization and mild germ cell degeneration observed	Germ cell degeneration (33.3%)	1.33 ± 1.15	4	Mild gonadal alteration
Escravos	*S. tritor*	1	Normal gonads with intact reproductive cells and proper organization	No lesions detected (0%)	0.00 ± 0.00	0	Normal gonadal structure
Brass	*I. africana*	1	Healthy gonadal tissue with normal germ cell development	No lesions detected (0%)	0.00 ± 0.00	0	Normal reproductive status

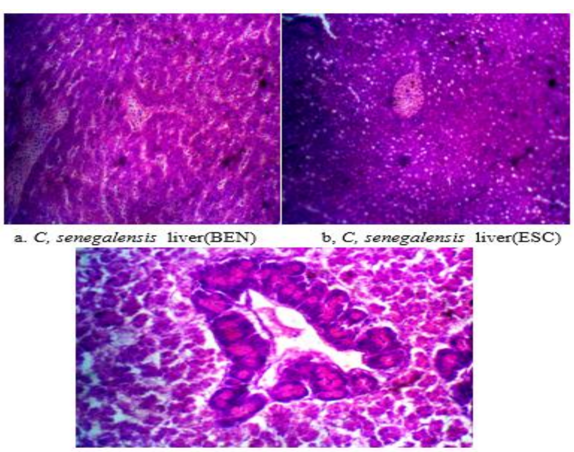
Estuary	Species	n	Histological Characteristics	Lesions Observed (%)	Mean Lesion Score ± SD	GAI	Histological Interpretation
Brass	*C/ senegalensis*	1	Mild gonadal changes including slight degeneration of germ cells	Germ cell degeneration (33.3%)	1.33 ± 1.15	4	Mild stress condition
Brass	*S. tritor*	1	Well-organized gonads with no visible abnormalities	No lesions detected (0%)	0.00 ± 0.00	0	Normal gonadal condition



c I. africana liver (BRAS)

There is no observable lesion. HE x400

Figure 2: Photomicrograph of Liver section in *I. africana from Benin Estuary, Escravos Estuary and Brass Estuary*



Liver- There is moderate congestion and accentuation of sinusoids and venules. HE x400

Figure 3: Photomicrograph of Liver section in C, senegalensis *from Benin Estuary, Escravos Estuary and Brass Estuary*

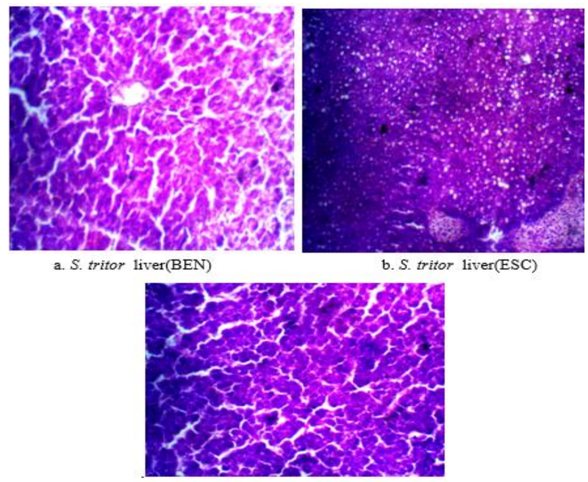


Figure 4: Photomicrograph of Liver section in *S. tritor* from Benin Estuary, Escravos Estuary and Brass Estuary

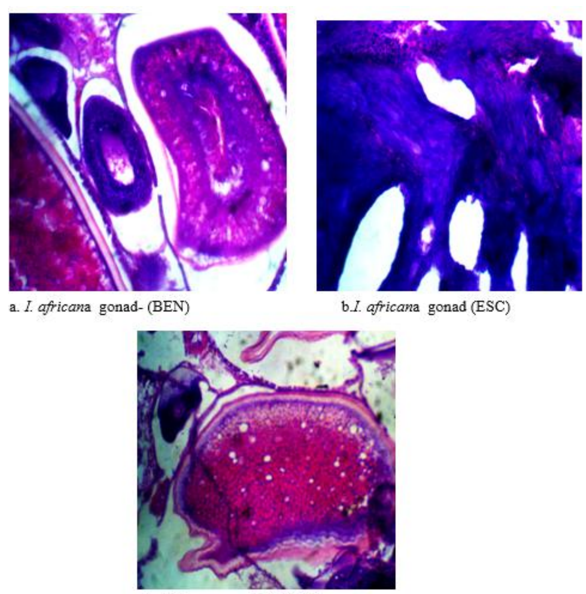


Figure 5: Photomicrograph of Gonad section in *I. africana* from Benin Estuary, Escravos Estuary and Brass Estuary

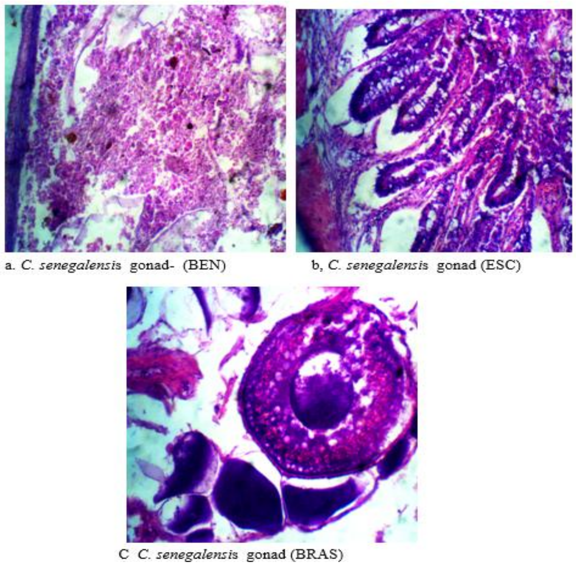
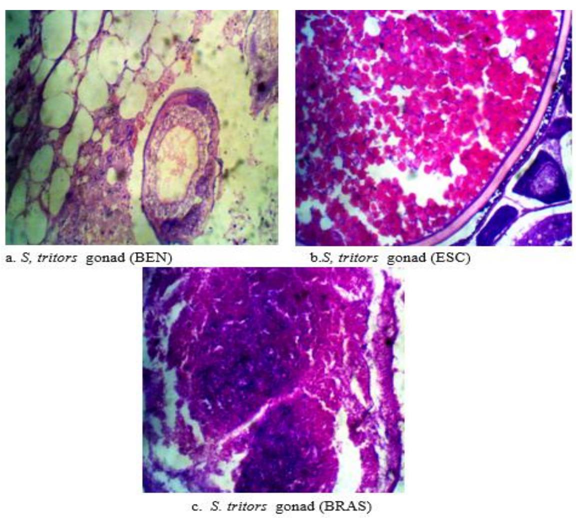


Figure 6: Photomicrograph of Gonad section in *C, senegalensis* from Benin Estuary, Escravos Estuary and Brass Estuary



Ovary- There is atrophy of follicles. HE x400

Figure 7: Photomicrograph of Gonad section in *S.tritor* from Benin Estuary, Escravos Estuary and Brass Estuary

3.2. Water Quality Analysis

Table 3: Comparison of Water Quality Parameters Across Sample Stations

Parameter	Location	Mean ± SD	N
Temperature (°C)	Brass	27.08 ± 1.44	12
	Escravos	27.42 ± 1.31	12
	Benin	26.83 ± 1.27	12
Dissolved Oxygen (mg/L)	Brass	6.13 ± 0.51	12
	Escravos	6.12 ± 0.79	12
	Benin	6.08 ± 0.69	12
pH	Brass	7.37 ± 0.54	12
	Escravos	7.32 ± 0.57	12
	Benin	7.88 ± 0.22	12
Salinity (ppt)	Brass	22.17 ± 6.60	12
	Escravos	24.92 ± 6.07	12
	Benin	21.75 ± 5.93	12
Turbidity (NTU)	Brass	17.17 ± 6.09	12
	Escravos	12.67 ± 7.11	12
	Benin	17.50 ± 6.69	12
TSS (mg/L)	Brass	45.83 ± 14.75	12
	Escravos	35.67 ± 13.04	12
	Benin	46.67 ± 12.31	12
Nitrate (mg/L)	Brass	1.85 ± 0.58	12
	Escravos	1.60 ± 0.59	12
	Benin	1.85 ± 0.58	12

Table 4: Spatial Variation on Water Quality Parameters

Parameter	F-value	p-value	Significance
Temperature	0.570	0.571	Not significant
Dissolved Oxygen	0.013	0.987	Not significant
pH	5.296	0.010	Significant
Salinity	0.923	0.408	Not significant
Turbidity	1.980	0.154	Not significant
TSS	2.505	0.097	Not significant
Nitrate	0.734	0.488	Not significant

Table 5: Tukey Post Hoc Test (Significant Only)

Parameter	Comparison	Mean Difference	p-value	Interpretation
pH	Benin vs Brass	+0.517	0.030	Significant
pH	Benin vs Escravos	+0.567	0.016	Significant

No other parameters showed significant pairwise differences.

3.2.1 Pearson Correlation Matrix

The Pearson correlation analysis reveals several important relationships among water quality parameters. Temperature showed a strong positive correlation with salinity ($r = 0.808$, $p < 0.01$), indicating that higher temperatures are associated with increased salinity, likely reflecting evaporation and marine influence. It also exhibited a moderate positive relationship with dissolved oxygen ($r = 0.604$, $p < 0.01$). Dissolved oxygen was positively correlated with salinity ($r = 0.774$, $p < 0.01$) and weakly correlated with pH ($r = 0.336$, $p < 0.05$), suggesting some linkage between oxygen availability and chemical conditions. Salinity demonstrated strong negative correlations with turbidity ($r = -0.973$), TSS ($r = -0.938$), and nitrate ($r = -0.968$), all significant at $p < 0.01$. This indicates that as salinity increases, water clarity improves and nutrient concentrations decrease, which is typical of marine-dominated conditions. Turbidity, TSS, and nitrate were strongly positively correlated with each other, suggesting that increased suspended solids are associated with higher nutrient loads, possibly due to runoff or sediment resuspension. Overall, the correlation results highlight strong interdependence among parameters and suggest that salinity plays a central role in structuring water quality dynamics in the estuarine system.

Table6: Pearson Correlation Matrix

Variables	Temp	DO	pH	Salinity	Turbidity	TSS	Nitrate
Temperature	1	0.604**	0.054	0.808**	-0.770**	-0.731**	-0.773**
Dissolved Oxygen	0.604**	1	0.336*	0.774**	-0.721**	-0.711**	-0.790**
pH	0.054	0.336*	1	0.232	-0.220	-0.212	-0.297
Salinity	0.808**	0.774**	0.232	1	-0.973**	-0.938**	-0.968**
Turbidity	-0.770**	-0.721**	-0.220	-0.973**	1	0.950**	0.960**
TSS	-0.731**	-0.711**	-0.212	-0.938**	0.950**	1	0.945**
Nitrate	-0.773**	-0.790**	-0.297	-0.968**	0.960**	0.945**	1

Legend: * = Significant at $p < 0.05$ ** = Significant at $p < 0.01$

Final Scientific Summary

Most parameters show no spatial variation, indicating environmental uniformity pH is the only parameter significantly influenced by location Strong correlations suggest salinity-driven ecosystem control

The figure illustrates temporal fluctuations and spatial similarities in temperature patterns. Values represent monthly observations; absence of replication precludes statistical inference.

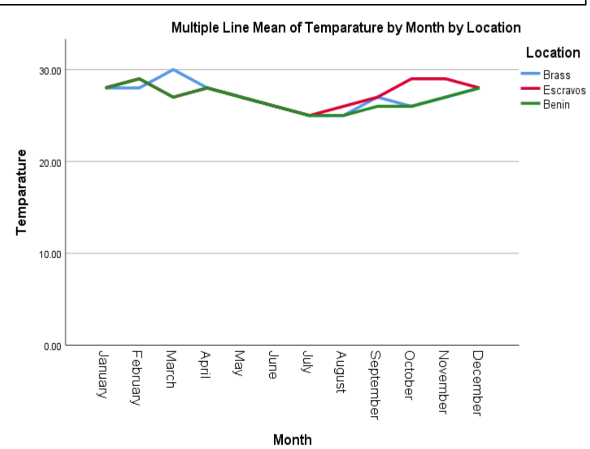


Figure8: Monthly variation in water temperature (°C) across Brass, Escravos, and Benin estuaries.

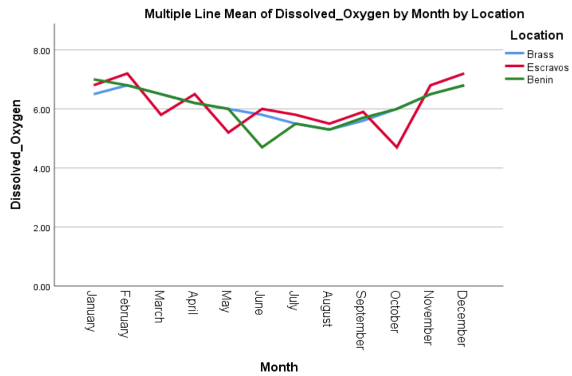


Figure 9: Monthly variation in dissolved oxygen concentration (mg/L) across the three estuaries. The plot shows seasonal trends and relative consistency among locations under varying environmental conditions.

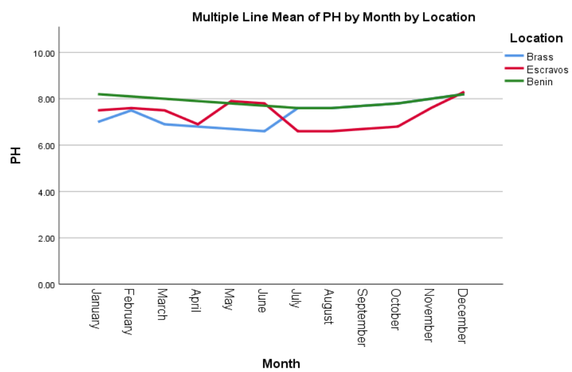


Figure 10: Monthly variation in pH across Brass, Escravos, and Benin estuaries.

The figure highlights spatial differences, particularly the relatively elevated pH values observed in the Benin estuary.

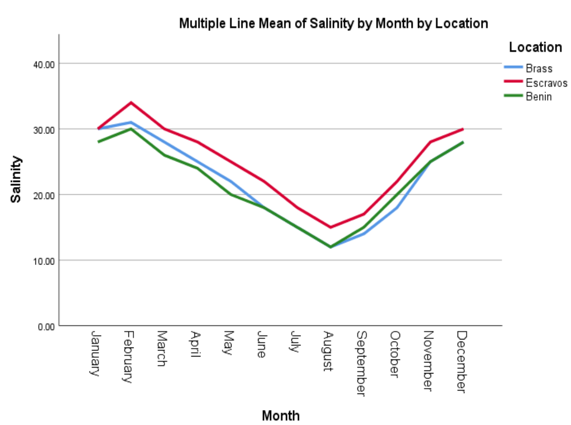


Figure 11: Monthly variation in salinity (ppt) across the study locations.

The figure demonstrates seasonal fluctuations likely associated with freshwater inflow and marine influence.

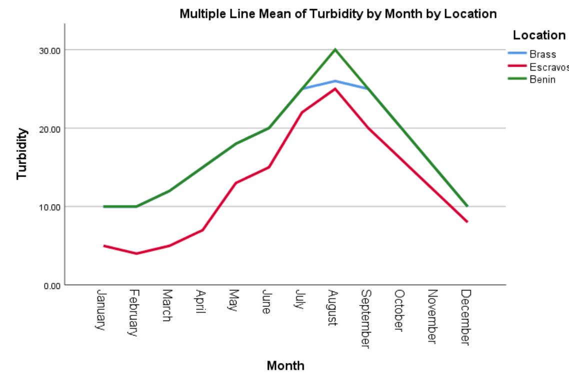


Figure10: Monthly variation in turbidity (NTU) across the estuaries.

The observed patterns indicate periods of increased suspended particles, potentially linked to runoff and sediment disturbance.

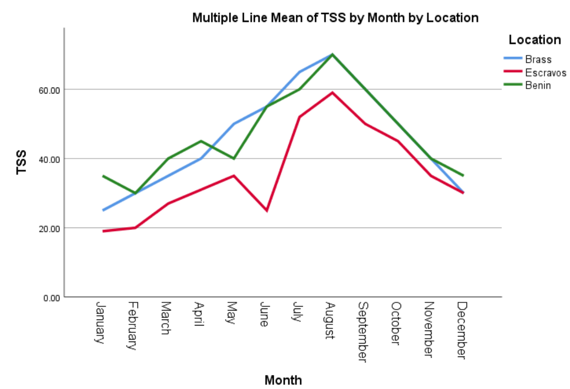


Figure 12. Monthly variation in total suspended solids (mg/L) across Brass, Escravos, and Benin estuaries. The figure reflects changes in particulate matter concentrations over time.

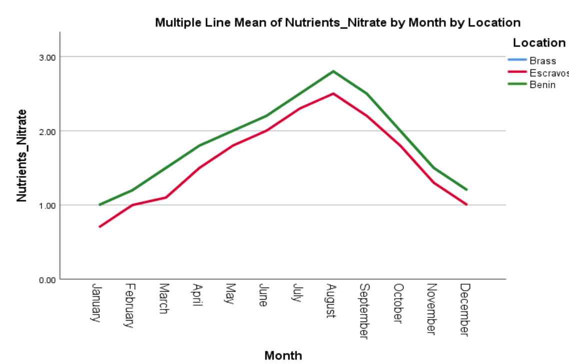


Figure 13: Monthly variation in nitrate concentration (mg/L) across the study locations.

The figure shows gradual temporal changes consistent with nutrient cycling processes.

IV. DISCUSSION

4.1. Histopathological Responses to Environmental Stress

The liver is among the most responsive organs to environmental perturbations in aquatic organisms because it plays a central role in detoxification, energy metabolism, nutrient storage, and the biotransformation of xenobiotic compounds. Consequently, histopathological alterations in hepatic tissues are widely regarded as reliable indicators of exposure to environmental stressors and provide valuable insight into the physiological condition of fish populations inhabiting aquatic ecosystems (Sonone *et al.*, 2020; Sharma *et al.*, 2023). The histological findings obtained in the present study revealed a generally high degree of hepatic integrity among the examined fish species, although species specific differences in tissue responses were evident.

The complete absence of observable liver lesions in *Ilisha africana* and *Scomberomorus tritor* suggests that hepatic function was maintained under conditions that did not induce measurable structural injury. Normal hepatocyte arrangement, intact nuclear morphology, and well-defined sinusoidal spaces are characteristic features of healthy fish livers and reflect the preservation of cellular homeostasis and metabolic competence (Korkmaz *et al.*, 2022). The lack of necrosis, fibrosis, inflammatory infiltration, or hepatocellular degeneration further indicates that these species were not subjected to contaminant burdens sufficient to trigger pathological responses in hepatic tissues. Similar observations have been reported in fish populations inhabiting relatively stable aquatic environments where contaminant concentrations remain below thresholds capable of inducing cellular damage (Soliman *et al.,* 2020).

In contrast, *Cynoglossus senegalensis* exhibited mild histopathological alterations characterized by sinusoidal congestion and focal cytoplasmic vacuolation. Although these lesions occurred at relatively low frequency, their presence is ecologically meaningful because both alterations are recognized as early manifestations of physiological stress in fish exposed to changing environmental conditions (Bhat *et al.*, 2023). Cytoplasmic vacuolation has been associated with disruptions in intracellular metabolism, altered lipid accumulation, and impaired cellular regulation arising from contaminant exposure or physiological stress (Gautam et al., 2024). Likewise, sinusoidal congestion reflects circulatory disturbances within hepatic tissues and may develop when environmental stressors interfere with normal vascular function and tissue perfusion (Korkmaz *et al.,* 2020). The simultaneous occurrence of these lesions in *C. senegalensis* therefore suggests that some individuals experienced low intensity hepatic stress despite the absence of severe tissue injury.

An important feature of the observed hepatic alterations is that they were not accompanied by nuclear abnormalities, necrosis, fibrosis, or inflammatory cell infiltration. This distinction is critical because severe toxicological effects typically involve progressive deterioration of cellular architecture, including nuclear degeneration and irreversible tissue damage (Bashir et al., 2022). The preservation of nuclear integrity in all examined specimens indicates that the lesions recorded in *C. senegalensis* were likely reversible and had not progressed to advanced pathological stages. Experimental investigations have demonstrated that vacuolar degeneration and mild vascular alterations often represent the earliest detectable responses to environmental contaminants and may precede more severe histological manifestations when exposure intensifies or persists over time (Bhat *et al.*, 2023; Gautam *et al*., 2024). Consequently, the lesions observed in the present study should be interpreted as indicators of mild physiological disturbance rather than evidence of substantial hepatic dysfunction.

The Histopathological Alteration Index provided quantitative support for this interpretation. The HAI value of four recorded for *C. senegalensis* remained considerably low and reflected only slight tissue alteration, while the values recorded for *I. africana* and *S. tritor* indicated complete absence of detectable histological damage. These findings collectively suggest that the overall health status of the examined fish populations remained satisfactory despite the occurrence of localized hepatic responses in *C. senegalensis*. The relatively low lesion scores also indicate that environmental pressures within the study area were insufficient to induce widespread pathological impairment of liver tissues.

Differences in the histological responses of the examined species may be linked to ecological characteristics that influence exposure pathways. Species occupying different trophic positions often encounter varying contaminant loads because pollutants can be transferred through aquatic food webs and accumulate differentially among consumers (Gao et al., 2021). Variations in feeding behaviour, habitat preference, and sediment association can therefore produce marked differences in physiological responses even among species inhabiting the same ecosystem (Whitfield et al., 2022). This explanation is particularly plausible for estuarine environments where contaminant distribution is spatially heterogeneous and where benthic associated species may experience greater exposure to sediment bound pollutants than pelagic species (Otogo *et al.,* 2022). The exclusive occurrence of hepatic lesions in *C. senegalensis* may therefore reflect species specific ecological interactions rather than a uniform environmental effect across the fish assemblage.

The generally favourable histological condition observed in this study may also be related to the ecological functions performed by mangrove dominated estuarine systems. Mangrove ecosystems contribute substantially to nutrient retention, sediment stabilization, organic matter processing, and water quality regulation, thereby enhancing ecological resilience and supporting healthy aquatic communities (Akram *et al.*, 2023). These ecosystems also function as biologically productive habitats that sustain diverse fish populations through the provision of feeding grounds, shelter, and nursery areas (Arceo Carranza *et al.*, 2021; Whitfield, 2020). Nevertheless, the Niger Delta mangrove region remains subject to multiple anthropogenic pressures, particularly oil exploration, habitat degradation, and resource exploitation, which continue to alter ecological processes and threaten aquatic biodiversity (Onyena & Sam, 2020; Aransiola *et al*., 2024). The mild hepatic alterations detected in *C. senegalensis* may therefore represent an early biological response to such pressures, even though severe tissue damage was not evident during the period of investigation.

Histopathological responses can vary substantially among individuals as a consequence of age, reproductive condition, nutritional status, and temporal fluctuations in environmental quality (Dane & Şişman, 2023). The relatively small sample size restricts the extent to which population level inferences can be made and may underestimate the true variability of hepatic responses within each species. Future investigations incorporating larger sample sizes, seasonal sampling regimes, and complementary biomarker analyses would provide a more comprehensive understanding of fish health and environmental quality within the study area.

Overall, the liver histopathology results indicate that the examined fish populations maintained largely normal hepatic structure and function. The absence of severe lesions such as necrosis, fibrosis, and inflammatory infiltration demonstrates that environmental conditions within the study area did not impose substantial hepatic stress during the sampling period. Nevertheless, the occurrence of sinusoidal congestion and cytoplasmic vacuolation in *Cynoglossus senegalensis* highlights the value of histopathological assessment as a sensitive monitoring tool capable of detecting subtle biological responses before the onset of overt tissue damage. Such early warning indicators are particularly important in Niger Delta estuarine ecosystems where continuing anthropogenic activities have the potential to influence fish health and ecosystem integrity over time (Onyena & Sam, 2020; Aransiola *et al.*, 2024).

4.2. Water Quality Characteristics and Ecological Implications

The physicochemical assessment showed that most water quality parameters remained within acceptable ranges for tropical estuarine ecosystems. However, significant spatial variation in pH and notable trends in salinity, turbidity, and suspended solids indicate environmental heterogeneity among the estuaries.

The relatively stable temperature and dissolved oxygen concentrations suggest favorable environmental conditions for fish growth and survival. Similar temperature ranges have been reported for tropical estuaries in West Africa and are considered suitable for estuarine fish communities (Gomes *et al.*, 2024).

Salinity emerged as one of the most influential environmental variables. The higher salinity recorded in Escravos reflects stronger marine influence and reduced freshwater input. Salinity is widely regarded as a primary ecological driver in estuarine ecosystems because it affects osmoregulation, species distribution, metabolism, and physiological performance (Whitfield, 2021; Elliott & Whitfield, 2023).

The strong negative correlations observed between salinity and turbidity, TSS, and nitrate concentrations indicate that marine-dominated conditions are associated with clearer waters and lower nutrient concentrations. Similar relationships have been documented in estuarine systems worldwide, where tidal exchange regulates nutrient dynamics and suspended sediment transport (Kennish, 2022).

The significant pH differences observed between Benin and the other estuaries may also have biological implications. Variations in pH influence nutrient availability, contaminant toxicity, and fish physiological processes. According to Boyd and Tucker (2023), even moderate pH differences can alter metabolic performance and stress responses in estuarine fishes.

Importantly, the environmental patterns correspond closely with the biological findings. The higher salinity regime and greater environmental distinctiveness of Escravos coincided with stronger morphometric differentiation, greater stock separation, and more evident histopathological alterations. This supports the growing consensus that environmental gradients play a central role in shaping fish population structure in estuarine ecosystems (Elliott & Whitfield, 2023; Fruciano, 2024)..

V. CONCLUSION

This study examined the histology assessment and environmental quality of *I. africana*, *C. senegalensis* and *S. tritor* from the Benin, Escravos, and Brass estuaries.

The histological results showed that most fish had healthy liver tissues, although mild stress-related changes were observed mainly in *C. senegalensis*. This suggests that environmental conditions are generally acceptable but may still cause slight stress in more sensitive species.

Water quality parameters were mostly within safe ranges, although variations in salinity and pH were observed. These environmental differences likely play a major role in shaping fish distribution and biological differences across the estuaries.

These findings highlight the importance of treating each estuary as a distinct ecological unit when planning conservation and fisheries management in the region.

Contributions to Knowledge

This study makes significant contributions to fisheries, aquatic ecology, and estuarine resource management, particularly in the Gulf of Guinea region. Based on the specific objectives and findings, the following new knowledge has been generated:

1) The study established new insights into the histopathological responses of estuarine fishes to environmental stress, showing how liver and gonadal tissues exhibit varying degrees of alteration such as vacuolation, congestion, and follicular atresia. These findings confirm that tissue-level changes can serve as sensitive biomarkers of environmental stress and ecosystem health.
2) It established a clear relationship between water quality variability and biological responses, showing that spatial differences in parameters such as salinity, pH, turbidity, and dissolved oxygen significantly influence histological integrity, and of estuarine fish species.
3) The study further established by demonstrating that environmental stressors (Dissolved Oxygen Variations, pH Alterations, Agricultural Runoff, Increased Turbidity and Suspended Solids etc.), particularly in the Escravos estuary.

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