Gross Anatomical Based Ecotoxicolgy: Evaluation of the Ecological Health Status of Bodo River, Using the Condition Factor and Health Assessment Index of *Tympanotanus fuscatus*

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ABSTRACT

This ecotoxicological study is aimed to evaluate the ecological health status of Bodo River using *Tympanotonus fuscatus* as a biomarker. The study complements chemical environmental monitoring with bio-indicator based bio-monitoring to achieve a more holistic ecological health assessment. Based on literature review on hazardous chemical crude oil spill, the following Target Chemicals (TCs) where selected for the study: lead (Pb), cadmium (Cd), chromium (Cr), copper (Cu), nickel (Ni) and polyaromatic hydrocarbon (PAH). The experimental study area was Bodo River, while African Regional Aquaculture Centre (ARAC) was selected as the references site. In both sites, water, sediment and *T. fuscatus* were sampled for a comparative study. Water and sediment samples were used to evaluate Environmental Water Quality Index (EWQI) and sediment quality respectively. The sampled fish was used for gross anatomical assessment of condition factor (CF) and Health Assessment Index (HAI). EWQI of Creek Road River results showed worse outcome than ARAC, with dissolved oxygen (DO) and copper (Cu) failed to meet the Maximum Allowable Toxicant Concentration (MATC) guideline for marine water. Sediment quality showed that worse outcomes in ARAC, but with Bodo River having a high Ni value above the MATC for marine sediment in Bodo River. There was no significant difference in CF and HAI for experimental and reference sites. The study was able to show that there was an ongoing Cu contamination of Bodo River with resultant reduction in DO and a historic spectrum of Ni contamination profile. The CF and HAI results showed a good ecological health status of the river. Though, the finding was inconsistent with the expected environmental impact from the water and sediment quality results. It was therefore concluded that a more ecological health sensitive test tool is needed to make a more assertive statement on the ecological health status of Bodo River.

Keywords: Gross Anatomy; Ecotoxicology; Water Quality; Tympanotanus fuscatus; Biomarker.

INTRODUCTION

Human health and well-being are closely linked to the state of the environment. High-quality natural environments provide essential resources such as clean air, water, fertile land, and materials for production. The Ogoni region, which include Bodo, has suffered extensive environmental damage due to oil spills from multinational oil companies' extraction activities (Fentiman, and Zabbey, 2015). This has severely disrupted the livelihoods of local communities that depend on these resources for fishing, farming, and daily activities (Fentiman and Zabbey, 2015). These concerns necessitate a study to evaluate the post impact ecological status of the the Bodo River basin, which can also to be to assess the efficacy of the remediation efforts so far administered to that environment.

It is well known that various human activities, industrial and other anthropogenic waste have caused damage to land, air, water, and organisms that inhabit them resulting in environmental burdens that are contamination, pollution, and health risk. Contamination is the presence of foreign substances beyond the pristine or allowable level for that environment (CCME, 2010; Allison & Paul, 2018). Pollution is environmental contamination resulting in deleterious biological effects. (WHO, 2006; EEA, 2021; CCME, 2010).

Representative species of the environment must be studied to assess the effects of pollutants in a an environmental matrix. An environmental bio-indicator organism can therefore be used as a biomarker of the ecological health of a concerned environment. A biomonitor can be identified as an organism or group of organisms that are important for qualitative and quantitative environmental pollution determination (Allison and Ogoun, 2024). The degraded environment in turn poses a serious threat to affect human health and social well-being which are feasible through the use of health status, naturally distributed and edibility of periwinkle, ***Tympanotanus fuscatus,*** to evaluate the ecological health of the study area. Periwinkle is vastly consumed as a staple food source in the Bodo locality, and in the Niger Delta region of Nigeria. Human exposure to trace metal contaminants through the consumption of periwinkle may result in adverse health effects. These health effects with varying symptoms depend on the nature and quantity of the metal ingested (Adepoju and Alabi, 2003).

The health of wild fishes can be used as an indicator of aquatic ecosystem health. Gross anatomical-based fish health assessments provide documentation of visible lesions or abnormalities, data used to calculate condition indices as well as the opportunity to do more in-depth analyses (Blazer, et al, 2013). Gross anatomical-based fish assessments, which may include; Condition Factor (CF). Organosomatic Index (OI) and Health Assessment Index (HAI), are external and internal observations and allow for the measurement of gross anatomical indices. Observations made during the gross anatomical-based assessment can be used to compare the prevalence of individual lesion types or percentage of normal individuals. However, they can also be used in a more quantitative health assessment (Goede and Barton, 1990; Adams et al., 1993)

Current ecotoxicological evaluations focus mainly on chemical analysis of toxicants in environmental media; lithosphere, hydrosphere and atmosphere. However, these assessments often overlook the ecological health impact of environmental toxicants. Hence, this study complements chemical environmental monitoring with bio-indicator based bio monitoring to achieve a more holistic environmental assessment. Gross anatomical methods of condition factor (CF) and health assessment index (HAI) was applied in this study as community based techniques. These methods are quick, cheap and can be taught to locals to deduce the health status of their fisheries ecology, before the application of a more technical and invasive certify and stratify pollution and other serious ecological health outcomes.

MATERIALS AND METHOD

Study Species

Habitat

Tympanotonus fuscatus is one of the most widely distributed species of periwinkles in the estuarine mud flats of southern Nigeria. *T. fuscatus* is a deposit feeder, feeding on mud and digesting the detritus and other organic matter in highly productive and in most cases extremely polluted estuarine ecosystems. The organism is very sensitive to pollutants and has been previously used as a bio-indicator of aquatic pollution because of its ability to accumulate and bio-magnify contaminants like heavy metals, and polycyclic aromatic hydrocarbons in the environment. (Davies et al., 2006; Kalay et. al.,1999; Canli & Atli, 2003; Ayenimo et. al,2005; Burger & Gochfeld,2005 53; Chindah et. al,.2009).

External Anatomy

The shell of *Tympanotonus fuscatus* is sharply conical and divided into a pointed apex (or spire), a columella (central axis) and cephalic cavity. The sculpturing shell is spiral turrets or ridges that are granular with spines. The spiral ridges are less marked in young animals tend to become very conspicuous in older individuals, giving the shell a ridged appearance. The shell colour ranges from grey-black-brown-red but is generally black or dark grey-brown, often lighter towards the apex, and is usually patterned with spiral darker lines.

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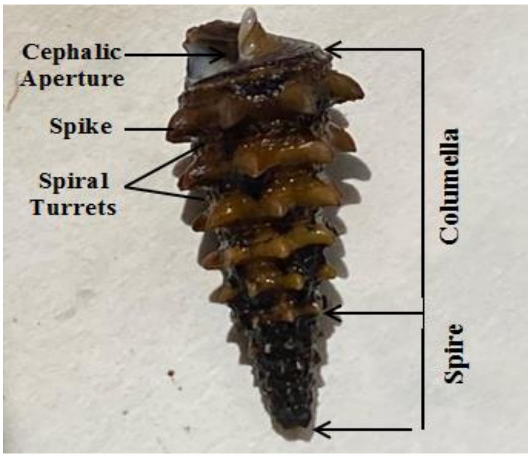

Figure 1: Photograph of the hard structure of *Tympanotanus fuscatus*

Soft Anatomy

The soft body of *T. fuscatus* is coiled in a series of complete turns, or whorls, of increasing diameter from the apex to the cephalic opening. The columella whorl is not visible externally. The body whorl, which is present at the cephalic openings is by far the largest whorl (Graham, 1988; Fretter and Graham, 1994). The proteinaceous operculum, which is attached to the thick muscular foot, serves as a door to close the cephalic aperture when the soft parts are withdrawn into the shell. The foot is attached to the columella by the powerful columellar muscle. The pale ventral sole makes contact with the substratum and is used for crawling. It may be folded on itself as it always is when retracted. The sole of the foot is located on the ventral.

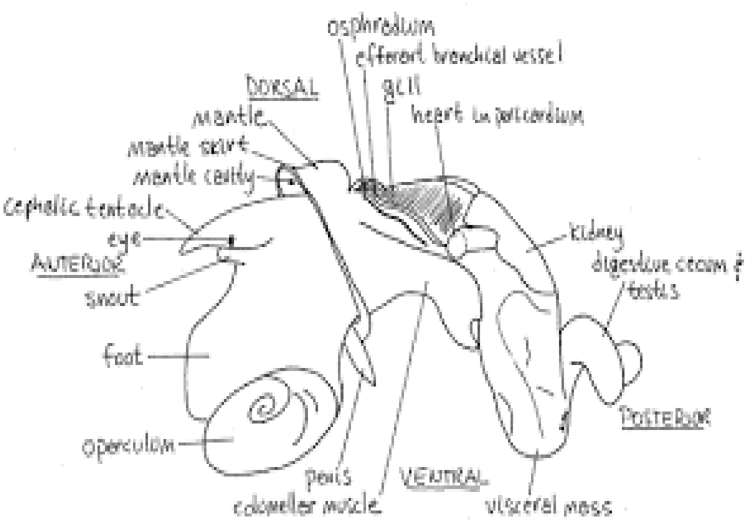

Figure 2: Soft Structure of *T. fuscatus*

Soft anatomy of *T. fuscatus:* The shell has been removed but the mantle is intact. Structures in the mantle cavity are viewed through the mantle (Fretter and Graham, 1994). The head is an anterior protuberance dorsal to the foot. It bears two sensory cephalic tentacles with a lateral eye on the base of each. A median snout protrudes anteriorly from between the bases of the two tentacles. The mouth is located at the anterior end of the snout. The snout is not retractile and is thus not a proboscis.

The mantle, which is the dorsal body wall, forms a fold that is the root and walls of the mantle cavity. The mantle cavity is a deep pocket lying dorsally above the anterior part of the visceral mass, posterior to the head. Its only opening lies above the head and is bordered by the mantle skirt. The anterior edge of this mantle fold is the skirt.

The gill is a large, long pale brown, oval organ lying medial to the osphradium. It extends from the osphradium to the midline and from the anterior to the posterior ends of the mantle cavity. Its left border bears faint white pigmentation that marks the location of the efferent branchial vessel which drains oxygenated blood from the gill to the heart. The lift gill is present in commogastropods and it is monopectinate.

The hypobranchial gland which secretes mucus into the mantle cavity is a long narrow secretory region of the mantle roof located on the right side of the gill. It is about the same size as the gill. The intestine exits in the stomach and winds through the visceral mass where it is hidden from view to the posterior end the rectum emerges from the mass and extends obliquely across the roof of the mantle cavity on the right border of the hypobranchial gland, is easily seen in males, but is hidden from view in reproductive females by the large yellowish egg capsule gland on the right edge of the mantle cavity. The prostate gland, which occupies the equivalent position in the male, is not as large and does not hide the rectum.

Study Area

Experimental Site

The experimental site for this study was Bodo River in Bodo town, Gokana local government area of Rivers State, Nigeria. This was where sampling of water, sediment and fish for experimentation was collected. Bodo is inhabited by Ogonis, it is located within Ogoniland. In 2010, the community had a population of around 69,000 people (Sampson, A., 2010). Bodo River forms long reticulated creeks, with some combining to form a common coastline water body at Bodo town, forming the Bodo waterside. The waterside plays host to a popular open market, the Bodo market and a boat transit point to other hinterland communities not accessible by road. The main occupations in Bodo are fishing and farming. A lot of the farming work is done by hand, and by women. Cassava is one of the main food items they farm there.

In 2003, a "relatively small" oil spill affected the mangroves in Bodo. In 2008 and 2009, two oil spills from the Trans-Niger pipeline operated by Shell Nigeria spilled at least 560,000 barrels of oil into the village's land, one of the biggest spills in decades of oil exploration in Nigeria. As a fishing town, the livelihoods of the majority of Bodo's inhabitants were destroyed. Fish populations were decimated, mangroves were destroyed, and water, fruits, and trees were all contaminated. People's health was also widely affected during the years following the oil spill (Vidal, 2012).

United Nations Environment Programme (UNEP) reports on the impacts of the oil spill on the environment of Bodo and the people living around it affirmed that the impact of the oil spill incidence increased the number of health-related issues in the area. It also resulted in the destruction of farmlands and loss of livelihood (UNEP, 2011).

Reference Site

The reference site for this is African Regional Aqua-culture Centre (ARAC), located at Buguma in Asari Toru Local Government Area of Rivers State. This was where the cultured periwinkle, of similar species and sizes as the feral fish was harvested and used as control for the study. ARAC was chosen for this study because it is an internationally recognized aquaculture Centre with good control of its periwinkle water quality.

Sampling

Sample Selection

Target Chemical (TC) Selection: Selection of target chemicals was based on findings from literature review of studies done on crude oil spill sites which includes lead (Pb),cadmium (Cd), copper (Cu),nickel (Ni),chromium (Cr) and PAH. (USEPA, 2005; Osuji et al., 2006; Fatoba et al., 2015; Mustafa et al., 2015; Aigberua and Tarawou, 2018).

Fish Selection: EROCIPS (Emergency Response to coastal Oil, Chemical and Inert Pollution from Shipping), (2006), ‘’Protocol for selection of sentinel Species’’ was basically used to select the appropriate sentinel specie for this study. The resident species, *Tympanotonus fuscatus* was consequently selected for this study.

Sample Collection

Water Sampling: Surface water was sampled from designated stations of experimental and reference site for water quality study. Standard protocol for water sampling method was adopted for physical and chemical parameters test in surface water and evaluation of result against Maximum Allowable Toxicant Contaminant (MATC) in the environment (USEPA, 2013; CCME, 2001). Four (4) samples of Surface water from each station, at two weeks interval was collected in vials and transported to the laboratory through iced tanks for further analysis.

Sediment Sampling: Sediment from the designated field stations were sampled for sediment quality. An Erkman Grab Sampler was used to collect underwater sediment of the experimental site and reference site for target chemical analysis. This was done in accordance with SOEPA (2001) sediment sampling guideline.

Fish Sampling: Test periwinkle was caught from Bodo River. Samples were taken at stations approximately at a transect of 100; 200; 500; 800; 1,500; 2,500; and 5,000 metres from the water front in the direction of the most persistent bottom current of the nearby river (EGASPIN, 2002). The control periwinkle was harvested from ARAC marine aqua culture at Buguma. A group of ten (10) periwinkle specimens of the target species, each from each study area, was randomly collected at the water's edge near the sampling stations. Periwinkle samples were meticulously gathered by hand picking at the intertidal flats of mangrove swamps. The collection occurred during the mid-tide level (MTL) and mean low tide level (MLTL). The specimen were subjected to a meticulous cleaning process and subsequently sealed in clean plastic bags containing samples of soil and water from their natural environment and transported to the laboratory for further evaluation. The shells of the periwinkle samples from each station were carefully cracked and removed to access the edible tissue inside. The tissue that was separated and underwent multiple rinses using distilled water before detailed anatomical and histopathological examinations

Evaluations

Chemical Environment Evaluation

Water Quality: The Bodo surface Water was analyzed for physico-chemical quality and use to evaluate the Environmental Water Quality Index (EWQI) applying the CCME (2001) formula to grade the surface water into the following categories:

- Excellent: (EWQI= Value 95-100) – water quality is not impaired; with conditions very close to natural or pristine levels.
- Good: (EWQI= Value 80-94) – water quality has a minor degree of threat or impairment; with conditions deviating from natural or desirable levels.
- Fair: (EWQI= Value 65-79) – water quality is occasionally threatened or impaired; with conditions deviating from natural or desirable levels.
- Marginal: (EWQI=Value 45-64) – water quality is frequently threatened or impaired; with conditions deviating from natural or desirable levels.
- Poor: (EWQI=Value 0-44) – water quality is almost always threatened or impaired; with conditions usually deviating from natural or desirable levels.

Sediment Quality: Target chemical (TCs) of sediment samples were analyzed using Atomic Absorption Spectrometer (AAS). The results were compared against the Maximum Allowable Toxicants Concentrations (MATC) standard for marine water sediment (CCME, 2001) in order to evaluate sediment quality.

Gross Anatomical Evaluation

Condition Factor (CF): The external gross anatomical features of weight and length relationship of the periwinkle was studied immediately after harvesting to ascertain their ecological health status (Fulton, 1902; Calendar, 1969; Allison & Ogoun, 2024).

The length-weight relationship was estimated using the equation: $W= a \times L^b$ and it logarithmic: In(W)= In() + bIn(L). Where W= Weight (g); L= Length (m^3); a' and 'b' = regression coefficient

The Fulton's Condition Factor (K_F) was calculated using the equation: $K_F = 100W/L^3$ .Where K_F = Fulton's Condition Factor (K_F); W = Weight of periwinkle (g); L = Length of periwinkle (m^3); and CF = Weight (g) x 100^{-5}/length (m^3)

The value of K_F is influenced by age of the periwinkle, sex, season, stage of maturation, fullness of gut, type of food consumed, amount of fat reserved and the degree of muscular development. The K_F value can be used to

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assist in determining the stocking rate of fishes in particular water. If the K value reaches an unacceptably low level in water which is totally or partly dependent on stocking, the stocking rate can be reduced accordingly until the K_F value improves and reaches an acceptable level. On the basis of comparism of the K_F value with general appearance, fat content etc. the following standards by Carlandar, (1969) were adopted:

- Excellence condition, trophy class (1.60).
- A good, well proportional (1.40).
- A fair condition (1.20).
- A poor condition, long and thin (1.00).
- Extremely poor condition (0.80).

Health Assessment Index (HAI): This is a community based field assessment tool for quantitative evaluation of periwinkle health (Adam et al., 1993). Immediately after catching, each periwinkle was assessed for external and internal gross anatomical body lesions using a checklist. The observed internal and external lesions were recorded and scored in terms of the severity of the lesion using a Health Assessment Index (HAI) protocol by Adams et al (1993). To evaluate HAI, the variable of the HAI were assigned values of 10, 20, 30, depending on the extent of abnormality or observed damage. To calculate an HAI for each periwinkle within the sample, numerical values for all variable were summed. The HAI for a sample population was then calculated by sunning all the individual periwinkle HAI values and dividing by the total number of periwinkle examined for the sample. A standard deviation for each sample was calculated

as: SD

Where N= number of periwinkle per site; X= average index for each periwinkle; Vi= index value for each periwinkle. The coefficient of variation (CV) is calculated as CV = 100. SD/X

RESULTS

Environmental Water Quality Index

Table 1: Descriptive statistic of Physical and chemical parameters used in evaluation of EWQI for Bodo

	BODO Surface Water Quality							
PARAMETERS	**DAY 1**	**DAY 2**	**DAY 3**	**DAY 4**	**Mean ± Std**	National Guideline (DPR) (mg/l)	MATC Int. Guideline (USEPA)	**Remarks**
Temp.	30.4	30.1	29.2	28.9	29.65 ± 0.71	Ambient (<31.0)	Ambient (<31.0)	NA
DO (mg/l)	5.51	5.37	5.22	5.17	5.32 ± 0.15	5.5 - 9.5	6.6-8	AP
pH	7.54	7.05	6.72	6.65	6.99 ± 0.41	6.0 - 9.5	6.5-8.5	AP
Salinity (ppt)	15.7	16.2	16.8	15.9	16.15 ± 0.48	25	NS	AP
Lead, Pb (mg/l)	<0.001	0.102	<0.001	<0.001	**0.001 ± 0.00**	1	0.008	AP
Cadmium Cd (mg/l)	<0.001	<0.001	0.019	<0.001	0.05 ± 0.02	0.05	0.008	AP
Copper Cu (mg/l)	0.076	0.130	0.249	0.098	0.21 ± 0.09	0.02	0.003	AP
Nickel Ni (mg/l)	<0.001	<0.001	<0.001	<0.001	0.001 ± 0.00	0.02	0.008	AP
Chromium Cr (mg/l)	0.054	0.167	<0.001	<0.001	**0.001 ± 0.00**	0.003	0.05	AP
PAH (mg/l)	<0.001	<0.001	<0.001	<0.001	0.001 ± 0.00	0.001	0.000005a	AP
SWQI					41.5		POOR	

Key: MATC = Maximum Allowable Toxicant Concentration; CCME= CCME (2001) MATC standard for Marine Water; DPR = DPR (2007) Guideline for marine water; USEPA = USEPA (2021) Saltwater chronic Criteria Maximum Concentration (CMC); Bold (Red) = failed tests; N/A = Not applicable for SWQI estimation; and AP = Applicable for SWQI estimation. This result showed that the following parameters failed to meet the Maximum Allowable Toxicant Concentration (MATC) guideline limits of Department Petroleum Regulation (DPR): Temp., pH, DO. Salinity, Cd, Cr, Cu, Ni and PAH.

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Using CCME (2001) guideline for mathematical evaluation of EWQI, BODO was estimated to be 41.5 (poor)

Table 2: Descriptive statistic of Physical and chemical parameters of ARAC

	ARAC Surface Water Quality							
PARAMETERS	**DAY 1**	**DAY 2**	**DAY 3**	**DAY 4**	**Mean ± Std**	National Guideline (DPR) (mg/l)	MATC Int. Guideline (USEPA)	**Remarks**
Temp.	30.4	30.1	29.2	28.9	29.65 ± 0.71	Ambient (<31.0)	Ambient (<31.0)	AP
DO (mg/l)	5.51	5.37	5.22	5.17	5.32 ± 0.15	5.5 - 9.5	6.6-8	AP
pH	7.54	7.05	6.72	6.65	6.99 ± 0.41	6.0 - 9.5	6.5-8.5	AP
Salinity (ppt)	15.7	16.2	16.8	15.9	16.15 ± 0.48	25	NS	AP
Lead, Pb (mg/l)	<0.001	0.102	<0.001	<0.001	0.03 ± 0.05	1	0.008	AP
Cadmium Cd (mg/l)	<0.001	<0.001	0.019	<0.001	0.01 ± 0.01	0.05	0.008	AP
Copper Cu (mg/l)	0.076	0.130	0.249	0.098	0.14 ± 0.08	0.02	0.003	AP
Nickel Ni (mg/l)	<0.001	<0.001	<0.001	<0.001	0.001 ± 0.00	0.02	0.008	AP
Chromium Cr (mg/l)	0.054	0.167	<0.001	<0.001	0.06 ± 0.08	0.003	0.05	AP
PAH (mg/l)	<0.001	<0.001	<0.001	<0.001	0.001 ± 0.00	0.001	0.000005a	AP
SWQI					66.5			FAIR

Key: MATC = Maximum Allowable Toxicant Concentration; CCME= CCME (2001) MATC standard for Marine Water; DPR = DPR (2007) Guideline for marine water; USEPA = USEPA (2021) Saltwater chronic Criteria Maximum Concentration (CMC); Bold (Red) = failed tests; N/A = Not applicable for SWQI estimation; and AP = Applicable for SWQI estimation. This result showed that the following parameters failed to meet the Maximum Allowable Toxicant Concentration (MATC) guideline limits of Department Petroleum Regulation (DPR):Temp., pH, DO. Salt, Cd, Cr, Cu, Ni.

Using CCME (2001) guideline for mathematical evaluation of EWQI, ARAC was estimated to be 66.5 (Fair)

Sediment Quality

Table 3: Descriptive statistics of heavy metals in sediment samples

parameters	ARAC [N = 3]			BODO [N = 3]			
	Min	**Max**	**MeanSD**	**Min**	**Max**	**MeanSD**	**National Guideline**
Lead Pb (mg/l)	1.45	3.01	2.23±0.78	0.00	0.00	0.001±0.00	**0.5**
Cadmium Cd (mg/l)	0.00	0.00	0.001±0.00	0.00	0.00	0.001±0.00	**0.1**
Copper Cu (mg/l)	4.89	5.43	5.18±0.27	0.67	1.95	1.46±0.69	**30**
Nickel Ni (mg/l)	2.87	3.98	3.47±0.56	5.08	6.12	5.47±0.57	**O.2**
Chromium Cr (mg/l)	0.00	0.00	.001±0.00	0.00	0.00	0.002±0.001	**12**
PAH (mg/l)	0.00	.13	0.08±0.07	0.00	0.01	0.01±0.00	**0.03**

Table 4: Post Hoc (ANOVA) multiple comparison of measured parameters in sediment

	sediments	MD	S.E.D	95% C.I of the Difference		P-value
Parameters				**Lower**	**Upper**	
Lead Pb (mg/l)	**BODO - ARAC**	-2.22900*	.51546	-3.7046	-.7534	.009*
Cadmium Cd (mg/l)	**BODO - ARAC**	0.00000	.01440	-.0412	.0412	1.000
Copper Cu (mg/l)	**BODO - ARAC**	-3.72000*	.99088	-6.5566	-.8834	.017*
Nickel Ni (mg/l)	**BODO - ARAC**	1.99667*	.37648	.9189	3.0744	.003*
Chromium Cr (mg/l)	**BODO - ARAC**	.00067	.00054	-.0009	.0022	.418
PAH (mg/l)	**BODO - ARAC**	-.07213	.03296	-.1665	.0222	.121

a. Post Hoc-Dunnett t-tests treat one group as a control, and compare all other groups against it.

Gross Anatomical Assessment

Table 5 Descriptive statistic of the Condition Factor of ARAC, Creek Road and Bodo

Measured parameters	ARAC [N = 10]			BODO [N = 10]		
	Min	**Max**	**MeanSD**	**Min**	**Max**	**MeanSD**
Weight	5.70	7.70	**6.89±0.67**	4.50	6.10	**5.13±0.56**
Length	45.00	54.00	**49.7±2.91**	29.00	44.00	**37.3±3.71**
Condition Factor	1.30	1.60	**1.40±0.11**	1.20	1.80	**1.39±0.17**

❖ Score = Excellent (1.60), good (1.40), fair (1.20), poor (1.00) & Very Poor (0.80). (Fulton, 1902)

Table 6: Inferential Statistic for Significant Difference Using ANOVA Condition Factor

		Sum of Squares	**df**	**Mean Square**	**F**	**Sig.**
Weight	Between Groups	15.57	2	7.784	19.854	**0.000***
	Within Groups	10.59	27	.392		
Length	Between Groups	778.40	2	389.200	47.271	**0.000***
	Within Groups	222.30	27	8.233		
Condition Factor	Between Groups	0.002	2	0.001	.051	0.950
	Within Groups	0.525	27	0.019		

Table 7: Post Hoc (ANOVA) multiple comparison of measured parameters

	Measured parameters	MD	S.E.D	95% C.I of the Difference		P-value
				Lower	**Upper**	
Weight	BODO - ARAC	-1.76	.28	-1.20	2.39	**.000***
Length	BODO - ARAC	-12.40	1.28	-9.84	1.42	**.000***
CF	BODO - ARAC	-.01	.06	0.11	1.19	.601

*The Mean Difference is Significant at the 0.05 level

a. Post Hoc-Dunnett t-tests treat one group as a control, and compare all other groups against it.

Table 8 Health assessment index (HAI)

Organ	SITE	Mean±SEM	F	R-square	p-value	Inference
	ARAC	3.00±1.53				
Crack shell	BODO	3.00±1.53				
	ARAC	6.00±4.00				
Gill	BODO	6.00±4.00				
	ARAC	3.00±3.00				
Kidney	BODO	6.00±4.00				

DISCUSSION

The EWQI has shown that the experimental site (Bodo River) has a worse EWQI (poor - 41.5) when compared to the reference site (ARAC), which was fair (66.5%). The failed water quality parameters were DO and Cu. Oxygen is important and essential for the survival and functioning of all aquatic organisms as it is required for respiration (DWAF, 1996). Fish need oxygen to breath while bacteria need oxygen for the oxidization of wastes. DO affects the solubility and availability of nutrients. Its low levels can result in damages to oxidation state of substance from oxidized to the reduced form thereby increasing the levels of toxic metabolites. This would no doubt reduce fish yield with the study river. Copper (Cu) occurs naturally in all plants and animals. It is an essential element for all known living organisms including humans and other animals at low levels of intake. At much higher levels, toxic effects can occur. Cu can enter the environment through waste dumps, domestic waste water, crude oil spill, refining of petroleum, combustion of fossil fuels and wastes (ATSDR, 2004). We can therefore surmise that, with the high levels of reported historic crude oil spills at the Bodo River basin over the years (UNEP, 2011; Vidal, 2012; Fentiman, and Zabbey, 2015), Cu would have still been leaching into the Bodo surface water from unremediated crude oil spill. It will damage a number of organs and systems, including the gills, liver, kidney, immune system, and nervous system (Cardeilhac and Whitaker 1988). Chronic Cu exposure will adversely affect fish health. Sublethal and toxic levels of Cu can damage the gills and other tissues of fish, and also are known to depress the immune system (Cardeilhac and Whitaker 1988). During toxicity, in addition to general signs of distress (e.g., increased respiration), fish may display darkening and behavioral abnormalities: lethargy, incoordination, problems with posture and balance, and, eventually, death (Cardeilhac and Whitaker 1988). This findings might be the reason why there was reported dwindling fish yield in the Bodo River (Fentiman, and Zabbey, 2015).

Sediment quality showed that ARAC had worse sediment quality than the experimental site, with Pb, Ni and PAH above the MATC levels for marine sediment. This needs would need further investigation since ARAC marine environment is a controlled. Nevertheless Bodo River has higher levels of Ni above the MATC for marine sediment. Sediment is a habitat and major nutrient source for aquatic organisms. Sediment analysis is important in evaluating qualities of total ecosystem of a body of water in addition to water sample analysis practiced for many years, because it reflects the long term quality situation independent of the current inputs and it is the ultimate sink of contaminants in the aquatic system. This indicates that there has been long-term Ni contamination of the Bodo River, which might be related former crude oil spills in the region (UNEP, 2011; Vidal, 2012; Fentiman, and Zabbey, 2015)

CF analysis showed that fish harvested from ARAC had better health conditioning than those caught from Bodo River. Nevertheless, fish from both sites had good CF score. This was further validated by the statistical study which showed no significant difference between both sites. But the comparison of length and weight showed significant difference between sites. CF is based on the hypothesis that heavier individuals of a given length are in better condition than less weightier fish. (Bagenal and Tesh, 1978). CF on this study is not consistent with the evaluated water and sediment quality ecological impact. Some fishes might be grossly anatomically observed to be in good condition but still have lesion that are causing serious organ dysfunction (Bernet et al., 1999; Marchand et al., 2009). Hence, histological analysis is needed to make a conclusive statement (Bernet et al., 1999: Allison and Paul, 2014).

HAI success as a gross anatomical ecological health status tool is based on the fact that if fish are in good condition, the vital organs and other easily observed body structures will be in good condition. The use of this method was based on the assumptions:

In fish under stress, tissue and organ function will change in order to maintain homeostasis;

If a change in function persists in response to continuing stress, there will be a gross change in the structure of organs;

If the appearance of all organs are normal according to the gross anatomical criteria, there is a good probability that the fish is normal; and

If the appearance of an organ or tissue system departs from the normal or from a control condition, the fish is responding to changes brought about by the environmental stressor (Goede and Barton, 1990; Allison and Paul, 2014).

Fish subjected to various stressors over an extended period, changes in organ appearance and morphology would become apparent and can be recognized as serious departures from normal. If these stressors are left unchecked, the fish becomes increasingly susceptible to disease, and mortality may result. HAI in this study showed that in all examined parameters, fish harvested from ARAC had a health status outcome than those caught from Bodo River. Nevertheless, statistical analysis showed that there was no significant difference between both sites. This finding did not correlate with the water and sediment quality impact results. Nevertheless, it should be noted that HAI was not intended to be diagnostic, but rather to provide a quick, simple means of judging the general health and condition of a group of fish (WRC, 2008; Allison and Paul, 2014). Hence, there is need apply a more invasive and diagnostic oriented too like histology to ascertain the extent of micromorphology alterations in order to evaluate the environmental pollution status of Bodo River basin.

CONCLUSION

Chemical monitoring protocols of this study has shown that there is an ongoing Cu contamination of Bodo River, with a resultant reduction in dissolved oxygen (DO) content of the surface water. There is proven long-term sedimentation of Ni, which was surmised to be due to previous crude oil spill in the Bodo River basin. Gross anatomical tools which were applied as an easy and quick method to ascertain the ecological health status of Bodo River, using *T. fuscatus* as a biomarker was not consistent with the potential impact that would have occurred from the water and sediment quality findings. It is therefore advised, that more diagnostically invasive and toxicity sensitive tools, like histological analysis, should be applied to get a more holistic results of the environmental health status of Bodo River.

DISCLAIMER (ARTIFICIAL INTELLIGENCE): Authors hereby declare that no generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

ETHICAL APPROVAL: All procedures performed in this study are in accordance with the ethical standards of the institutional and/or national research committee.

Competing Interests

Authors have declared that no competing interests exist.

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