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Journal of Fisheries Science

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## ARTICLE Impact of Commercial Floodplain Aquaculture on Common-pool Resource Dependent Community

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#### ABSTRACT

Aquaculture in pond and floodplain was accelerated in Bangladesh in the 1990s as a means of better production and income which was backed by the donor agencies, NGOs, and the government. Currently, the commercial actors are involved in the aquaculture systems due to the availability of production technologies and inputs. This paper aims to explore how the commercialization and privatization of floodplain aquaculture become the cause of the sufferings of the natural resource-dependent people and biodiversity loss in the floodplains. Now, Influential people hold control of the common pool floodplains and restricted the access of the Small-Scale Fishers (SSF) to manage the aquaculture. Our findings suggest that the SSF, for whom the seasonal floodplains were an important source of livelihood, their livelihood has been destroyed and overall wellbeing have been negatively affected. Besides that, lending enough evidence to the increased inequality, a new group of poor has emerged. Because instead of ensuring the welfare of SSF, Bangladesh government has leased the floodplain lands to the powerful rich people. In addition to growing inequalities, natural resource degradation has welcomed social vulnerabilities. However, no development initiative will ever be sustainable and effective if the existing socio-ecological setting is not considered. Bangladesh government should take robust attempts to revisit fisheries policies to ensure livelihood resilience of fisheries resource-dependent community by managing the access rights of the common pool resources.

#### 1. Introduction

Bangladesh holds the world's richest and most complex aquatic food systems <sup>[1]</sup> where fish and fishery resources are derived from mainly two sources- capture and culture <sup>[2]</sup>. Though capture fisheries cover 89% of the total freshwater areas of Bangladesh, production from capture fisheries plummeted during the past three decades <sup>[3,4]</sup>. In contrast, aquaculture has shown spectacular growth, which led Bangladesh to become one of the self-sufficient

countries in terms of fish production <sup>[2,4,6]</sup>. This remarkable development of the aquaculture sector is known as the 'Blue Revolution', with the belief that aquaculture can contribute to fight against hunger, malnutrition, and unemployment problems <sup>[3,6]</sup>. Whilst this blue revolution in Bangladesh has impressed many and inspired much research on biological, economic, and governance aspects of the fishery, less attention has been paid to the socio-cultural issues around fishers' livelihoods. The

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blue revolution changed and transformed the aquatic food system to ensure the intensification of aquaculture production in a similar fashion as the green revolution did to the agricultural sector. To understand the overall process and impact of this transformation, it is needed to understand the local and global discourses supporting this course of action.

Privatization and market-friendly development policies in poor and third world countries were promoted following the discourse of development economics in the 1980s and 1990s [8]. Pioneers of these models didn't consider socioeconomic, cultural, political, and environmental factors associated with these in Latin America and most of the third world. As a result, some strategies prescribed by the World Bank to deal with hunger and malnutrition in some third world countries throughout the 1970s and 1980s led to aggravation and contradictory results <sup>[8]</sup>. For example, the World Bank Development Report in the 1970s portrayed Lesotho as a poor; agriculturally and commercially backward country with a homogenized entity prepared for benefits of development interventions <sup>[8]</sup>. Ferguson <sup>[8]</sup> has questioned the desirability, effectiveness, and beneficial value of the western development discourse in the context of rural Lesotho by analyzing the actual social effects of the development projects. Following this trend known as Green Revolution (GR), farming for subsistence was transformed enormously in favor of the intensification of food production systems in developing countries <sup>[9]</sup>. To ensure food security, employment, and income generation for the growing population, Bangladesh has also adopted GR strategies and technologies like high yield varieties of rice, fertilizers, irrigation, and chemical pesticides since the 1970s<sup>[2,6,9]</sup>. Though rice yield tripled by 2013 from 17 million tons to 52 million tons [11] in Bangladesh, still GR has been condemned for not giving the required attention to the social, ecological, and agricultural context which in turn has believed to be responsible for growing social and environmental inequalities, negative socio-economic transformation <sup>[6,8,9]</sup>.

Aquaculture as a means of better production and income was generated as an outcome of the blue revolution and was backed by the donor agencies, NGOs, and the government <sup>[12]</sup>. As the leading authority, the Department of Fisheries (DoF), Bangladesh has always supported and changed policies in support of aquaculture development, including contribution to private sector development. Historically, seasonal floodplains of Bangladesh are common pool resources that are managed collaboratively and used by local communities <sup>[4,12]</sup>. But these floodplains have been under commercial aquaculture for the last two decades. Numerous evidence shows that the conversion of seasonal floodplains that were previously occupied by small-scale capture fishing into commercial aquaculture often comes with adverse social and environmental consequences <sup>[4,13,14]</sup>. The commercialization of flood plain aquaculture (FPA) coupled with new ways of privatization of land through the current leasing system and fisheries policies. This paper is based on a small-scale qualitative research among common pool resource users and commercial aquaculture in four seasonal floodplains in Bangladesh. This paper focuses on to what extent commercialization and privatization of FPA impact the traditional common-pool resource using system and how it has changed aquatic resource-dependent community's livelihood and social structures. Finally, it will briefly discuss the importance of understanding the long-term dynamics of the socio-ecological dynamics of flood plain resource use systems for designing and implementing sustainable fishery management.

## **2.** Transformation from Traditional SSF to Commercial FPA

Geographically, Bangladesh is graced by large amount of open water resources known as rivers, canals, floodplains like haors, beels, and lakes. Historically rural people harvest fishes for subsistence and livelihood by using traditional techniques and equipment from these water bodies <sup>[12,15]</sup>. According to FAO (2005), these rural fishers are included in Small Scale Fishers (SSF). Basically, SSF is associated with different interchangeable terms, such as 'artisanal', 'local', 'coastal', 'traditional', 'small', 'subsistence', 'inshore', 'nonindustrial', 'lowtech', and 'poor' fishing [17]. SSFs play a pivotal role in the livelihoods and well-being of rural people. Besides, more than ninety percent of fishers are included under SSF who contribute to more than half of the total global fishery production<sup>[15]</sup>. Compared to the commercial fishery, SSF is considered ecologically resilient; sustainable in securing local resource users' livelihoods; accessible to more people and equitable in sharing socio-economic benefits derived from the aquatic resources <sup>[4,15,16]</sup>. Though SSFs play a pivotal role in developing countries' economies and fish production, the management and sustainability of SSF is a growing concern. Currently, many SSFs including Bangladesh's are facing numerous social and environmental challenges. Among them, transformations from artisanal fishing to aquaculture are now ubiquitous in developing and third-world countries. As part of the blue revolution, aquaculture has been introduced and promoted in Bangladesh, with the objective of enhancing food security directly by the production of fish for household consumption and by improving the supply and reducing the price of fish in the market <sup>[1,2,14]</sup>. Along with NGOs, both national and international research organizations, Bangladesh government has made tremendous transformation and improvements in pond aquaculture, floodplain aquaculture, communitybased fisheries management and ecosystem approach to fisheries management by exercising technological innovation. The major shifts can be discussed under three broad categories- pond aquaculture, community-based fisheries management, and community-based flood plain aquaculture.

#### 2.1 Pond Aquaculture Extension

In 1990, the DANIDA funded MAEP- 1 project was introduced in Mymensingh district, which continued up to 1994<sup>[12]</sup>. This project focused on semi-intensive pond aquaculture through knowledge and input support with the help of a network of skilled extension workers. Their extension approach included demonstration ponds, credit farmers, and contact farmers. "Credit farmers" were poor pond owners having no experience of aquaculture who received training and input support on credit for doing aquaculture. On the other hand, "Contact farmers" were selected to widen extension support among interested pond owners who only received training followed by a visit by an extension worker. In this way, MAEP-1 supported 823 credit farmers and 2,594 contact farmers <sup>[12]</sup>. After that, the Thana Level Fisheries Extension Project (TLFEP) was a Bangladesh Government-funded DoF project, which started implementation in 1994<sup>[12]</sup>. It directly supported 12,000 demonstration model fish farmers and adopted a trickle-down approach to diffuse aquaculture technologies to 60,000 neighboring "Fellow farmers". This project supported not only pond owners but also people who were interested in cultivating fish and motivated them to lease ponds <sup>[12]</sup>. But this leasing process didn't remain confined in leasing ponds, rather lead to further leasing of government-owned water bodies.

#### **2.2 Community Based Fisheries Management**

DoF is the responsible authority for formulating policies, strategies, and preparing rules to conserve fisheries and enhancing fish production, but they do not hold the sole control over the using rights on the water bodies <sup>[15]</sup>. Instead, According to East Bengal State Acquisition and Tenancy Act, 1950, state-owned water bodies are under the control of the Ministry of Land, which leases out fishing rights to the highest bidder for

generating revenue <sup>[1,14]</sup>. Though fisher cooperatives, consisting of existing artisanal fishers were supposed to get priority, researchers presented evidence that fishers have failed to gain exclusive fishing rights under the current leasing system <sup>[1,4]</sup>. In reality, though fishers received the lease of close water bodies, often by taking sublease or by renting the water bodies, wealthy and politically powerful people gained control over these <sup>[1,17]</sup>. This short-term tenure right leads to over-exploitation of aquatic resources, declining biodiversity, and a lack of conservation measures <sup>[15]</sup>.

From the 1990s, DoF undertook proactive measures for capture fisheries management by adopting co-management approaches through a number of donor-supported projects (CBFM-1, CBFM-2 and MACH) and demonstrated good practices at different ecological settings with higher fish production, increased biodiversity, increased fish consumption, and increased incomes <sup>[12]</sup>. These projects mostly promoted group stocking of carp in closed water bodies and supported government initiatives to restore fish habitats <sup>[1,16]</sup>. In 2000, 300 among 12000 state water bodies were transferred to the DOF for 10 years to secure fishing rights for genuine small-scale fishers and to ensure sustainable community-based fisheries management<sup>[1]</sup>. Though, this initiative restored fishery productivity and biodiversity; improved the livelihoods and fish consumption of local communities, the independent functionality of these CBOs was not continued after the project is phased out <sup>[14,16,17]</sup>.

#### 2.3 Community Based Flood Plain Aquaculture

On one hand, the success of pond aquaculture motivated people to be involved in aquaculture, and on the other hand, community-based fisheries management pointed out the option and potential of leasing in government-owned water bodies. When these possibilities interlocked with each other, it resulted in Flood Plain Aquaculture (FPA). Typically, floodplains are deep depressions that become inundated in the monsoon during which they are naturally recruited by indigenous fishes, aquatic flora and fauna [1,4,18,20]. Most of the seasonal floodplains become the habitat of aquatic resources during the wet season and are used for rice cultivation during the dry season, which is widely known as the rice-fish system <sup>[18,20]</sup>. Historically, these public, public-private, privately owned floodplains were the major source of natural fish production and during monsoon, rural people had common and open access fishing rights over there <sup>[1,16]</sup>. However, inspired by the blue revolution to boost fish production and income generation; some seasonal floodplains were brought under Community Based Fish Culture (CBFC) and Flood Plain Aquaculture (FPA) project <sup>[4,16]</sup>. These projects combined public waterbodies with pooled private land and then transformed the natural hydrology of floodplains by building enclosures <sup>[4,18]</sup>. They adopted productivity-enhancing techniques, such as stocking exotic carp fingerlings, adding a supplementary feed, and fertilizing the pond, which resulted in high fish production <sup>[16,18]</sup>. Very few study showed that aquaculture in these flood plains had created some negative impacts, such as destruction of natural fishery and biodiversity by enclosure of floodplains, restriction on small scale fishers' fishing rights during the wet season, social exclusion of the poorest part of the community, and unequal distribution of profit <sup>[4,16,18,19]</sup>. Findings pointed that these negative impacts have significant further impacts on the livelihoods of community people, including small-scale fishers [4,16,18,19]

Despite these negative environmental and socioeconomic effects, aquaculture practices in floodplains continued expanding. After the project phase-out period, wealthy influential people accepted the learning of the projects and gave it a private and commercial shape. Sometimes they indirectly lease in the flood plains from the government through the help of registered fisher groups and sometimes they rent the floodplain from the owners or CBO. Thus, commercialization and privatization of flood plain aquaculture are happening in many parts of Bangladesh. But no evidence-based literature has been found regarding this commercial flood plain aquaculture (FPA) during this study.

#### 3. Study Location and Data Source

Four adjacent floodplains named Bajail Beel, Cheera Beel, Boyrakuri Beel, and Dura Beel was selected for this study. These flood plains are located under the Brahmaputra river basin in Mymensingh and have similar agro-ecological and socio-economic characteristics.

Data for this study have been gathered from January 2020 to December 2020 by adopting qualitative tools such as in-depth-interview and focus group discussions. Empirical data have been collected from people who used to depend on aquatic food systems for their livelihood and current owners of commercial fisheries projects in respective study areas. To have a clear understanding of the changing social and economic pattern, some experienced and aged local people have been interviewed who have seen the changes over time. Respondents have been selected following the purposive sampling protocol. The total number of IDIs was 20 (12 with resource dependents, 4 with landowners, and 4 with commercial aquaculturists) and the total number of FGDs was 5

(3 with resource dependents and 2 with aquaculturists and service providers). Before data collection, consent was taken from the respondents. Finally, findings were recorded, transcribed, coded, and analyzed in a meaningful way.

#### 4. Characteristics of Selected Beels and Their Resource Use System

Bajail Beel, Cheera Beel, Boyrakuri Beel and Dura Beel are located in Berunia Union under Bhaluka Upazila. They cover 100, 22, 18, and 6 acres of land respectively whereas all three beels except Boyrakuri comprises both public and private land. Geographically these beels were large deep depressions, which used to turn into floodplains during the monsoon by retaining rainwater and floodwater. They used to be seasonally flooded and remained submerged for 6 to 8 months. During the monsoon, indigenous wild fish and other aquatic flora and fauna used to enter and reproduce in these flood plains. No feeding or fertilization was added, the natural life cycle was favored by the siltation and organic decomposition of aquatic resources. In these flood plains, land boundaries were relatively clear and fixed during the dry season, whilst during the wet season these boundaries remained indistinguishable. As a result, in the wet season aquatic resources were considered as common-pool resources granting open access for both owners and non-owners of land. Different types of native species of fish, shellfish, wetland birds, and aquatic plants used to be found. Local communities used to secure their livelihoods through collaboratively managing and using these resources. Local people had inherited knowledge and adaptation techniques for making their livelihoods from these aquatic resources. They were heavily dependent on flood plains and a remarkable part of their livelihoods and the social structure used to receive shape by the common use of these aquatic resources. Many Small Scale Fishers used to fish using artisanal gears for subsistence and run their families. Small indigenous fishes captured from the floodplains were the main source of their animal protein intake and contributed to maintain nutrition standards. The common-pool resource distribution and its interdependence with the livelihood patterns of dependent communities was a good example of communal harmony.

At the end of the wet season, the landowners used to restrict the open-access resource use system by installing temporary fences in the open sides. The remaining fish then used to be considered as private property, harvested jointly by the landowners, and distributed based on the amount of land ownership. During the dry season, the land boundaries were very clear. Due to the presence of water for a longer period, people used to produce 'Boro' rice once a year during the dry season in the same land. After the final harvesting of fish, the sheltered water used to be open accessed again, when water was irrigated from here to the surrounding croplands for agricultural production. However, common-pool resource dependency was a source of livelihood for the community people that can be characterized by a long tradition of adaptation to the dynamics of the social and natural environment.

Bajail Beel, Cheera Beel, Boyrakuri Beel, Dura Beel, and surrounding areas have been under commercial aquaculture for more than two decades. The landowners have excavated the beels and raised the peripheral dike of the beels, closed connecting canals, and installed enclosures to regulate water retention. Through this intervention, they have changed the seasonal flooding characteristics of these beels and stopped the natural recruitment of aquatic flora and fauna. Nowadays, via a mutual understanding process, the landowners are renting these beels. The landowners' association holds the exclusive power to decide the renting process, price, and duration. If none of the landowners is interested then the beels usually rented to people who are powerful either economically or politically. Though public land is included in Bajail Beel (25 acres), Cheera Beel (3 acres), and Dura Beel (0.5 acres), only the government land under Bajail Beel has been leased in through a formal procedure. As all four beels have gone under private ownership, so nowadays small-scale fishing and access of common people are not allowed. This change in resource allocation system has created threats to both the livelihood of resource-dependent people and to the community harmony. A snapshot of overall characteristics of studied beels has been presented below.

Table 1. Characteristics and status of selected Flood Plain

Name of Flood Plain	Land Ownership	Area (Acre)	Leasing Status	Rent/year (BDT)	Duration of Aquaculture
Bajail Beel	Private- Public	100	Leased	48,00,000	1996
Cheera Beel	Private-Public	22	No	10,56,000	1998
Boyrakuri Beel	Private	18	N/A	10,08,000	2013
Dura Beel	Private-Public	6	No	3,84,000	2013

Data Source: This study

#### 5. Findings

The findings focus on the livelihood patterns and strategies of the four study beels in terms of resource

access rights, social relations, gender norms and roles and practices, poverty and inequality, vulnerabilities and risks in pursuing livelihoods, and social wellbeing.

#### 5.1 Impact on Small Scale Fishing (SSF)

Historically people of Mahmudpur and Chandorati village had inherited knowledge and adaptation techniques for making their livelihoods from locally available aquatic resources. Community people had open fishing rights during monsoon and were allowed to use and collect all sorts of natural resources around the year from Bajail Beel, Cheera Beel, Boyrakuri Beel, and Dura Beel. According to field data, around 500 families currently live around these four beels, among which 80% families were involved in full-time or part-time fishing previously. These Small Scale Fishers used to depend on native species of fish for food and earn a living by selling fish in the past. Subsistence fishing from these beels used to create significant contributions to meet protein intake and good health. Fishing was not only a source of livelihood for the researched community but also an 'art of living' which has portrayed as a style of adaptation to the dynamics of the social and natural environment <sup>[16]</sup>. These small-scale fishers used to depend on different types of local crafts, gears, technologies, and indigenous fishing knowledge <sup>[16]</sup>. But currently the beels have been rented to aquaculture businesspersons who have invested a large amount of capital. As consequence, small-scale fishers are no longer allowed to capture fish from the Flood plains. One fisher said-

"I along with other community people used to go for fishing in the beel at night. We used to spend the whole night and capture a good amount of fish. Income from one night was sufficient to meet the weekly expense of my family. It was fun and life was relaxed then. Now I can't go to the beel and catch fish. So, I work as a day laborer for income-earning which I don't enjoy at all."

In this circumstance, small-scale fishers and beel dependent communities no longer capture natural fish for their livelihood. They are forced to change their income generation sources, which has a broader socio-economic impact on their life. Many people were involved with it and at the same time, many native species of fish and aquatic resources were available. Since the beel was open, water irrigation took place from here to the surrounding croplands. In other words, community people had open tenure rights and many people had employment opportunities. People used to fish in this beel and earn a living by selling fish. Now, these native fishers have changed their occupation, as they do not have the opportunity to do so. There is increasing evidence that inland aquatic resources are drastically declining in both quantity and quality, and there is a severe loss of livelihood, depending on these resources. The causes behind this are the conversion of more and more wetlands into commercial aquaculture and agriculture to meet the demands of a rapidly growing population; overexploitation of aquatic resources and siltation <sup>[13,19]</sup>. As a result most of the small-scale fishers have changed their occupation as they do not have the opportunity to do so.

#### 5.2 Impact on Access Right and Social Cohesion

Floodplains of Bangladesh are under different types of complex ownership such as completely public, public land surrounded by private lands, and some are completely private <sup>[19]</sup>. Public floodplains are normally leased out by the Department of Land (DoL) in the auction. In most cases, the wealthy and politically- influential people who can afford to pay the lease take control over the floodplains for fish culture. Under the current leasing system, revenue collection is the main target that does not consider the biological impact, and local livelihood <sup>[1,4]</sup>. Three (Bajail Beel, Cheera Beel, and Dura Beel) out of four selected beels comprise both public and private land. Only public land of Bajail Beel has been leased following the formal process from DoL. At the same time, all the beels have multiple landowners (Bajail Beel- 40, Cheera Beel- 20, Boyrakuri Beel- 7, and Dura Beel- 5), who have rented the beels to aquaculture businesspersons. As a result, the external businesspersons have gained complete control over the respective beels. As part of the aquaculture intensification process, they have restricted the entry and allocation of resources of the community people. The fishing rights were not well established and the small-scale fishers were not able to defend their fishing rights in any of the two non-leased Beels (Cheera Beel and Dura Beel). The landowners of these two Beels are socially and economically powerful, who are enjoying the overall benefit through exercising threats and social pressure. Poor small-scale fishers have failed to gain fishing rights over the beels, mainly because of high rent value. In this circumstance, poor fishers and beel dependent communities no longer capture natural fish for their livelihoods. Due to enclosures built by aquaculture businesspersons around all four beels, most local residents lost access to their source of income, food security during scarcity, fodder of livestock, drainage, and irrigation for agriculture. According to one respondent-

"Because of renting the Beel for aquaculture, we are in trouble. We can't catch fish for subsistance, cultivate rice by using Beel water, raise ducks depending snails, raise cattle depending natural fodder coming from the Beel. Current tenant came to our home and told us that he is the owner of the Beel now and warned us to not go to the Beel."

Common-pool resources, resource users, landowners, and all relevant stakeholders exist in webs of power and meaning <sup>[19,20]</sup>. Individuals and communities located in or near our selected beels had mutual understanding and arrangement of this resource utilization. But because of the commercialization and privatization of these beels, profit is being driven to some people over a mass community who was dependent on these resources. As common people's access has been limited, they have fallen into immense suffering. This has led to extremes of inequality in terms of access, wealth, and income. As a result, this increased gap between rich and poor is affecting community cohesion and resilience through increased social and economic disparities. Similar social resilience issue in the shrimp culture sector in Bangladesh has been observed where local elites and urban-based entrepreneurs built fortunes based on shrimp culture, leaving the coastal population to be worsen off<sup>[25]</sup>.

#### 5.3 Impact on Food Security

Both positive and negative impact of flood plain aquaculture have been observed in terms of household food security. Around 80% of rural households used to catch fish for subsistence and for selling, and the contribution of fish to the animal protein food basket was about 60% <sup>[1]</sup>. However, the poor people used to catch many "miscellaneous" small fishes from the floodplains, which have been neglected in official statistics. Small fishes were the accessible and affordable food of poor people and were good sources of micronutrients <sup>[26]</sup>. Three decades ago, unemployment was common and community people reported an annual great hardship, starting from August to November. During that time, subsistence fishing was the most important source of food and income to the people of Mahmudpur and Chandorati village. According to our data, beels were closely associated with their lives in the past. These were open where poor community people regardless of age and gender (around 80%) used to fish for their subsistence. But commercial FPA has changed the abundance of wild fish and the open fishing right, which resulted in a severe food security issue in the community. One of the respondent cited:

"Native fish are no longer available in beels. Previously, whenever we didn't have anything to eat, I used to set nets in the beel and collect fish for our dinner. We always have a variety of fish in our food baskets. We have to buy fish from the market nowadays. As the price of fish is very high, sometimes I can't manage fish once a month."

In contrast, fish production has increased a lot due to commercial FPA. Numerous studies have depicted the success of aquaculture in terms of increased fish production and contribution to animal protein consumption <sup>[2,4,20]</sup>. As a result, the supply of fish at the market has increased and the price of fish has decreased. Therefore, many argue that aquaculture contributes to improve the food security of the local population <sup>[2,4,20]</sup>. Our data also supports that more people can buy fish from the market and ensure household food security nowadays. In practice, people have to buy fishes produced by aquaculture from the market. Therefore, people's accessibility of these fish depends on their affordability, which is determined by their income. Thus, fish supply and cheap price in the market don't necessarily ensure household food security and protein intake.

#### 5.4 Impact on Gender Relation

In the context of Mahmudpur and Chandorati village, maintaining household food security during scarcity is considered a domain of women. Their restricted access to beels has negatively affected their household food security and nutrition. Previously many snails were available in the beels which was a good source of Duck feed. Women of Mahmudpur and Chandorati village used to raise ducks depending on these types of natural feed. Duck meat and eggs were a vital source of their protein consumption and income. These aquatic snails have declined significantly due to FPA. According to one respondent-

"I used to raise ducks and feed them snails collected from beel. Now snails are not available in our locality and I cannot afford commercial feed. Therefore, I can't raise ducks nowadays. Previously, if we had nothing to eat, I used to boil eggs and feed my children. Now I have to buy eggs from the shop."

As a result, women's traditional source of income has been hampered. Besides, women used to collect stems and roots of water lilies, water chestnut, water spinach, Hydra fluctuant, and many other aquatic fauna for consumption during food scarcity in monsoon. Due to FPA, women's access to beels has been restricted and the abundance of the fauna also has been reduced significantly. This has an indirect impact on traditional gender relations of the community. According to female respondents, they are no longer able to maintain household food security and to contribute significantly to the household food security. As a result, their position in the household has become vulnerable and participation in decision-making has been affected negatively.

#### 5.5 Impact on Biodiversity

No sustainable plan was required for leasing in Bajail Beel and renting all four selected flood plains. The current renting process gives the tenant's exclusive right to use the beels according to his or her very own need. Currently, all the tenants are doing mixed aquaculture in their respective rented floodplain. Based on the cost-benefit analysis and market demand, they are producing Rui, Katla, Mrigal, Sor puti, Tilapia in the flood plains. Their pond preparation process includes cleaning, applying lime, and eradicating all predators from the pond. Through the cleaning and liming process, they destroy all sorts of natural flora and through the predator eradication process they destroy the native fish species and aquatic animals. One respondent has rightly pointed out-

"Indigenous fish abundance is zero now. After renting the beels for aquaculture all of our small fishes are gone. How could they survive? Aquaculturists apply lime in the pond which destroys all sorts of eggs and fingerlings of fish."

There is no administration system to determine if their aquaculture practices are sustainable in the water bodies. In addition to fish, numerous flora, snails, reptiles, snakes and other wetland resources were abundant in these beels. Overfishing, enclosure of beels, and road construction were commonly cited causes of declining natural productivity and biodiversity of the beels. By negatively affecting the breeding and growth of flood plain resources, current aquaculture practice has accelerated the destruction of aquatic resources of selected beels.

According to this research, fish production has increased significantly in aquaculture, compared to capture fishery. But no specific data of capture fishery production from selected flood plains were found. According to the current aquaculturist of Cheera Beel and Dura Beel who also possessed land ownership in both the beels-

"Production of our target fish species has increased which contributes to income-earning. In contrast, we lost most of our native fish species, which were easily captured from the beel and taste good. Now I have to buy those neglected species at a high price from the market."

So, loss of biodiversity has been observed as an impact of commercial aquaculture in research areas. We have asked the status of current and previous abundance of different fish species. The findings are as such:

 Table 2. Status of aquatic biodiversity in selected flood

 plains

Local Name	Scientific Name	Previous Status	Current Status
Rui	Labeo rohita	Common	Common
Catla	Catla catla	Common	Common
Calibaus	Labeo calbasu	Common	Common
Koi	Anabas testudineus	Common	Common
Titputi	Puntius ticto	Common	Vulnerable
Sarpunti	Puntius sarana	Common	Common
Shing	Heteropneustes fossilis	Common	Nearly threatened
Magur	Clarias batrachus	Common	Nearly threatened
Tengra	Mystus tengara	Common	Vulnerable
Gulsha	Mystus cavasius	Common	Vulnerable
Chital	Chitala chitala	Common	Nearly threatened
Boal	Wallago attu	Common	Nearly threatened
Gutum	Lepidocephalus guntea	Common	Vulnerable
Boro baim	Mastacembelus armatus	Common	Nearly threatened
Taki	Chana punctatus	Common	Common
Gojar	Channa marulius	Common	Vulnerable
Rani mach	Botia dario	Common	Vulnerable
Madhu pabda	Ompok pabda	Common	Nearly threatened
Ketchki	Corica soborna	Common	Nearly threatened
Kani pabda	Ompok bimaculatus	Common	Vulnerable
Foli	Notopterus notopterus	Common	Nearly threatened
Bheda	Nandus nandus	Common	Nearly threatened
Kholisa	Colisa fasciatus	Common	Nearly threatened
Lomba chanda	Chanda nama	Common	Nearly threatened
Kakila	Xenentodon cancila	Common	Vulnerable
Dhela	Osteobrama cotio	Common	Vulnerable
Bacha	Eutropiichthys vacha	Common	Common
Baila	Glossogobius giuris	Common	Vulnerable
Darkina	Esomus danricus	Common	Nearly threatened
Shol	Channa striatus	Common	Common
Mola	Amblypharyngodon mola	Common	Common
Tara baim	Macrognathus aculeatus	Common	Common
Ghonia	Labeo gonius	Common	Nearly threatened

Data Source: This study

The destruction of aquatic biodiversity is affecting common pool resource-dependent people through the loss of nature's services. Conservation scientists are emphasizing biodiversity conservation including fish in a reversal of past trends. However, current aquaculture businesspersons of selected beels are not interested to concentrate on biodiversity conservation rather their focus is on profit maximization from FPA.

#### 5.6 Impact on Agriculture

During the last few decades Bajail Beel, Boyrakuri Beel, and Dura Beel were very important parts of the agricultural production of Chandorati village, whereas the people of Mahmudpur village's dependency was on Cheera Beel. As these beels are in deep depressions, during monsoon these were usually utilized as agricultural water drainage systems through small connecting canals. As a result, people were able to save the agricultural land and crops from being flooded. On the other hand, in the dry season, people used to grow crops by irrigating water from the sheltered flood plain water. People used to grow rice in the flood plains during the dry season. But because of aquaculture in these beels, landowners have built embankments, which has changed the natural hydrology. As the natural water drainage system has been disturbed and blocked, the adjacent agricultural land becomes easily flooded during the rainy season now. It disrupts the cultivation of Aman rice according to some respondents. In contrast, people used to depend on beel water to irrigate their Boro rice production during the dry season. As beels are under aquaculture projects, people are no longer allowed to use the water for irrigation. Therefore, they have to depend on groundwater using deep tube wells, which is costly. One respondent has pointed out-

"How can I cultivate without water. The project owner does not give us irrigation water. If I use deep tube well water, then the production cost increases a lot and I cannot make a profit from agriculture. Therefore, now I depend on rainwater, if it comes, I will cultivate, otherwise, I will work as a day laborer."

Similar effects have been pointed out in the rice-fish culture study and findings show that how this system has altered the indigenous agricultural system and lead to technological intensification of agriculture <sup>[27]</sup>.

#### 6. Discussion

Growing commercialization and privatization of FPA in Bajail Beel, Cheera Beel, Boyrakuri Beel, and Dura Beel has marginalized community people whose livelihood was dependent on aquatic resources. Our analysis showed that aquaculture in four selected beels has increased targeted fish production, which has improved the supply of proteinrich food to the local rural and nearby semi-urban areas. All four commercial FPA produce low-value fish, which coupled with increased fish supply, is contributing to improved protein intake for poor and market-dependent consumers. In contrast, these productive aquaculture methods also have some potentially negative impacts at the community level, although these could not be formally demonstrated or quantified in this research.

All four beels have excluded a large number of subsistence fishers from getting the benefit of improved fish production and affect them adversely through the deprivation of their common property rights over the floodplains. For example, Cheera Beel and Dura Beel from our research area have some public land in it, which is being illegally pulled in and used by the current aquaculture businessperson. Though open conflicts specifically related to traditional open access to rights over public waterbody didn't occur on the beels, tension and jealousy among the community people have been found. However, common-pool resource-dependent people have already been used to with the profit-driven aquaculture practice. Where the role of government was supposed to ensure well being of common people, the government has leased out the land to some rich people. As a result, privatization of these FPA has taken the profit to a wealthy group of people, and restricted access for others especially for the commons.

Severe depletion of aquatic biodiversity has been observed because of changing the hydrology of flood plain by building enclosures and applying intensive aquaculture techniques. As poor households were dependent on small and capture fishes for their animal protein consumption, depletion of these stocks has seriously affected their food consumption pattern and household food security. Besides, these seasonal floodplains were an important source of livelihood for poor people, especially small-scale fishers who have fallen into immense sufferings. So they were bound to change their traditional profession and migration which has transformed their overall socio-ecological interdependency in a negative manner. Though FPA in selected beels has multidimensional actors (fingerling, feeding, fertilizing, fishing labor etc.) working with them all the aquaculturists depend on the market for their needs. As a result, though employment opportunities have been generated, those are not targeted at local poor people. In most of the cases, flood plain resource-dependent communities are marginally benefited or are negatively affected by commercial FPA in researched beels.

Eventually, a new group of poor has emerged and their sufferings have worsened while some people are making a profit and becoming rich. Together with growing inequalities, natural resource degradation is creating social vulnerability. However, there were certain types and levels of social dependency, community cohesion, and socio-political stability around these flood plain centered common-pool resources. Now individualism has been created which has affected the social cohesion and overall well-being of the community, if we look from the development perspective.

While conflict between the protection of biodiversity and the development of rural livelihoods is well known, findings of this study show that the extension of commercial FPA has a dual effect in terms of loss in livelihood opportunities and decreased biodiversity. If any sort of conversion of natural resources occurs as part of a development initiative such as the blue revolution, then considering the existing socio-ecological setting is a prerequisite. Otherwise, the overall impact will neither be sustainable nor be entirely effective. It has also been argued by Toufique and Gregory that <sup>[9]</sup>, conjointly, the 'Blue Revolution' that boosted the development of the aquaculture sector in Asia is still struggling with the reminiscences of the severe environmental disasters that it engendered two or three decades ago <sup>[4]</sup>.

This consideration is pivotal in understanding the true costs and benefits of management decisions. It is important to develop locally appropriate livelihood enhancement programs for individuals who are expected to suffer during the period under consideration, particularly those who are in disadvantaged positions <sup>[28]</sup>.

#### 7. Conclusions

Social scientists often point out the trickle-down effects of the commercialization of natural resources on the poorest and resource-dependent part of the community, because of the potential risk of restricted access to rights, economic exclusion, and overall well-being consequences. In this context, this research has been conducted using commercial FPA in Bajail Beel, Boyrakuri Beel, Cheera Beel, and Dura Beel as an empirical field study. This paper was designed to understand socioeconomic and environmental aspects related to the transformation of flood plains for aquaculture inspired by the blue revolution. The primary objectives of the blue revolution were to contribute to poverty reduction, income generation, supply vital nutrition to poor households, and improvements in the overall welfare of low-income households<sup>[2]</sup>. However, in practice, commercialization and privatization of FPA is now a well-established practice in Bangladesh, which is excluding a poor part of the community from getting the service of natural resources <sup>[4]</sup>.

Most of the empirical researches have depicted the gains from FPA <sup>[4,18,19]</sup>. But its negative impact on the livelihoods of the common pool resource-dependent people, their social structure, and biodiversity haven't been assessed properly in contrast to the success. In reality, commercialization and privatization of FPA in our researched area has come with adverse social and environmental consequences including limited access to

common fishing grounds, changed traditional profession, increased social inequality, environmental pollution, and loss of biodiversity. All this evidence indicates the classic pitfall of developing commercial flood plain aquaculture. Similarly, the negative consequences of the World Bank proposed daudkandi FPA project has been portrayed by Toufique and Gregory<sup>[4]</sup>. They questioned the desirability and effectiveness of FPA by and for the common pool resource-dependent community through analyzing the distributional consequences in Daudkandi FPA.

Considering the success, aquaculture is important for increased fish production, but aquaculturists should find alternative and sustainable ways for its extension. Changing the hydrology, destroying the natural biodiversity of flood plains, and bringing them under commercial aquaculture should not be inspired and allowed as it has severe negative consequences. In Bangladesh, the distributional consequences of floodplain stocking has been ignored continuously in the design and implementation of fisheries policies <sup>[4]</sup>. There are knowledge gaps and consequent inconsistency in formulating policy for FPA in particular and fisheries in general. It has been admitted that large-scale enclosures of floodplains are destroying natural fishery and biodiversity and are preventing SSF from gaining access to fishery during the monsoon <sup>[21]</sup>. A genuine urge and recommendation have been observed in different literature to focus on policy measures and regulatory frameworks that facilitate common pool resource-dependent people's access to publicly owned land, water, and aquaculture extension services <sup>[2,4,8]</sup>. Besides, we have reliable and enough evidence (CBFM-1, CBFM-2, CREL, ECOFISH, and ECOFISH-2) which shows that increased fisheries production, income, and biodiversity conservation could be achieved by co-management of floodplains, wetlands and rivers. It has been argued that the economic value of conserving natural resources could be 100 times more than destroying and converting them to other commercial uses <sup>[29]</sup>. So not only conversion and degradation of floodplains for short-term private gain should be discouraged; but also sustainable use, conservation of natural resources should be followed. In extreme cases, compensation or payment for ecosystem services (PES) could be provided to the excluded part of the resource-dependent community. In all stages of designing appropriate fishery management and adaptation policies to navigate the effects of such transformations, it is required to understand and consider the traditional resource use system and dynamics of community livelihood <sup>[9]</sup>. Based on this small-scale qualitative study, no meta narrative was intended to be provided, rather this paper tried to focus on some empirical livelihood dynamics and wellbeing conditions of flood plain resource-dependent community that is being overlooked by the aquaculturists and policymakers because of the hype generated by the blue revolution following the dominant economic development discourse.

Researchers have examined how international development projects are conceived, researched, and put into practice <sup>[7]</sup>. It also looks at what these projects actually achieve. The idea of externally directed 'development' has been criticized for not taking proper account of the daily realities of the communities it is intended to benefit <sup>[8]</sup>. Instead, they often prioritize technical solutions for addressing poverty and ignoring its social and political dimensions, That's why the structures that these projects put in place often have unintended consequences. Development projects will continue to fail until the process becomes more reflective <sup>[8]</sup>.

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## ARTICLE Investigation and Protection of Fishery Resources in the Middle of Bohai Sea

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ARTICLE INFO	ABSTRACT		
Article history Received: 1 December 2020 Accepted: 16 March 2021 Published Online: 13 September 2021	In May and October 2017, 12 stations were set up in the Central Bohai Sea for fishery resources investigation. The results show that there are many dominant species in this area, and the inshore fishery resources are higher than those in the open sea because of the abundant nutrients from land, the high density of zooplankton and the food of swimming animals. In order		
<i>Keywords</i> : Fishery resources Investigation Central Bohai sea	to effectively protect the fishery resources in the Central Bohai Sea, thi paper puts forward some suggestions, such as strengthening the protectio propaganda, scientific and reasonable fishing, and strengthening th management of marine environment.		

#### 1. Introduction

The purpose of the investigation of fishery resources and fishery production status is to understand the fishery production environment, composition, distribution and quantity of fishery resources in the Central Bohai Sea area, analyze and master the distribution of fish eggs, larvae and juveniles, and the status quo of fishery production. In order to provide basic data for the possible impact of engineering construction period, operation period and unexpected accidents on biological resources <sup>[1]</sup>, and put forward reasonable suggestions for the protection of marine fishery resources.

#### 2. Materials and Methods

#### 2.1 Survey Methods

It is carried out in accordance with the relevant

methods such as the specification for marine monitoring, the specification for marine investigation, the Handbook for marine fishery resources survey and the technical specification for the assessment of the impact of construction projects on marine living resources (SC/T 9110-2007). Make a good record of the marine survey, and record the sampling, testing and analysis of all stations.

#### 2.1.1 Fish Eggs, Larvae and Juveniles

The survey of fish eggs and larvae shall be carried out in accordance with the relevant requirements of gb12763.6 marine survey specification Part 6: marine biological survey. A shallow water plankton net (50 cm in diameter and 145 cm in length) was used for quantitative sampling from bottom to surface. Qualitative samples were collected using a large plankton net (80 cm in diameter and 280 cm in length) with a horizontal trawl for 10 min at a speed of 2 n miles/h. The collected samples

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Figure 1. Survey station location diagram

**Table 1.** Longitude and latitude of survey stations

STANCE	Latitude(N)	Longitude(E)	investigation item
1	38° 52' 12''	118° 50' 05''	
2	38° 53' 00"	119° 21' 10"	
3	38° 50' 05''	119° 53' 12"	
4	38°39' 25"	118° 32' 05''	
5	38° 30' 00''	118° 53' 05"	
6	38° 31' 10"	119° 22' 08''	Tick and and
7	38° 30' 00"	120° 00' 15''	young fish fishery
8	38° 15' 17"	118° 32' 15"	resources
9	38° 10' 10''	119° 05' 16"	
10	38° 10' 14''	119° 38' 45"	
11	37° 52' 16"	119° 21' 18"	
12	37° 52' 14"	119° 54' 14"	

were fixed in 5% formaldehyde seawater solution, and then classified, identified and counted in the laboratory.

#### 2.1.2 Swimming Animals

The trawling survey of swimming animals shall be carried out in accordance with the relevant provisions of "gb12763.6 marine survey specification Part 6 marine biological survey", "marine fishery resources survey manual" and "national coastal zone and tidal flat resources comprehensive survey concise regulations". The single bottom trawl is used in the trawl survey of fishery resources. The mesh size is 5.6 cm, the perimeter of net mouth is 80 m, the mesh size of bag net is 20 mm, the width of net opening is 10.2 m and the height of net opening is 5.5 M. The trawl speed is 3.0 n mile/h, and the average sweeping area of each station is 0.05667 km<sup>2</sup>. The species of the catch were identified on board, and the weight and mantissa were recorded according to the species. The samples were frozen and brought back to the laboratory for detailed determination of biological data.

#### 2.3 Evaluation Method

#### 2.3.1 Swimming Animals

The method of sweeping area is used to calculate the density of swimming animal resources. The basic principle is to calculate the existing absolute resource density of unit area through the number of swimming animals captured in unit area swept by the net when trawling. The formula is as follows:

 $\rho=D/(p\bullet a)$ 

Where:  $\rho$  is the existing resources; D is the relative resource density, i.e. the average catch; a is the sweeping area of the net times; P is the catch rate of the net. The capture rate is calculated as 50%.

#### 2.3.2 fish eggs and juveniles

The formula for calculating the density of eggs and juveniles is as follows

G=N/V

Where G is the individual number of fish eggs, larvae and juveniles per unit volume of seawater (ind./m<sup>3</sup>); n is the individual number of eggs, larvae and juveniles in the whole net, the unit is ind.; and V is the filtering water volume, in cubic meters ( $M^3$ ).

#### 3. Results

#### **3.1 Fish Resources**

#### 3.1.1 Species Composition

A total of 39 species of fish belonging to 20 families and 7 orders were captured in the survey area. The list of fish is shown in Table 2. A total of 29 species of fish belonging to 17 families and 5 orders were captured in the survey area in spring. A total of 28 species of fish were captured in autumn, belonging to 13 families and 5 orders.

#### 3.1.2 catch

(1) spring

The average catch of fish in spring was 904 fish/h, 12.414 kg/h. According to the analysis of catches, the number of juveniles accounted for 30.20% of the total number, 273 fish/h, and the biomass was 1.082 kg/h. The average catch of adult fishery resources was 631 fish/h, 11.332 kg/h.

(2) autumn

The average catch was 303.9 kg/h in autumn. According to the analysis of catches, the number of juveniles in this survey accounted for 26.73% of the total number, which was 81 fish/h and the biomass was 0.265

kg/h. The average catch of adult fishery resources was 222 tails/h, 3.664 kg/h.

#### 3.1.3 Resource Density Assessment

#### (1) spring

In spring, the average catch of fish was 904 fish/h, 12.414 kg/h; the average catch of young fish was 273 fish/h, and the biomass was 1.082 kg/h; the average catch of adult fish was 631 fish/h, 11.332 kg/h. After conversion, the average stock density of adult fish is 399.75 kg/km<sup>2</sup>, and that of juvenile fish is 9630 fish/km<sup>2</sup>.

(2) autumn

In autumn, the average catch of fish was 303 fish/h, 3.929 kg/h; the average catch of young fish was 81 fish/h, the biomass was 0.265 kg/h; the average catch of adult fish was 222 fish/h, 3.664 kg/h. After conversion, the average resource density of adult fish is 129.25 kg/km<sup>2</sup> in autumn, and the average resource density of juvenile fish is 2857 fish/km<sup>2</sup>. According to the above results, the average stock density of adults and juveniles is 171.86 kg/km<sup>2</sup> in spring and autumn, and 3924.5 fish/km<sup>2</sup> in juveniles.

#### **3.2 Cephalopod Resources**

#### 3.2.1 Species Composition

There are mainly two types of cephalopods in the investigated sea area. One is coastal species, which mostly inhabit in the coastal shallow waters, with small individuals, slow swimming speed and only short-distance movement. This type of squid, octopus octopus and octopus Octopus <sup>[2]</sup>. The other type is inshore species, which mostly inhabit in the coastal waters where the coastal water and the outer sea water meet. They have larger individuals, faster swimming speed and longer migration distance. They have better adaptability to the environment and have a wide spatial distribution range.

According to the investigation results in spring and autumn, three species of cephalopods were captured in the survey area. Among them, 2 species of cephalopods were captured in spring, including sepia japonicus and Octopus ocellatus, and 3 species of cephalopods were captured in autumn (Table 3).

In spring, sepia japonicus is the dominant species. Octopus ocellatus is an important species. In autumn, sepia japonicus is the dominant species, Octopus ocellatus is an important species, and Octopus ocellatus is a common species.

#### 3.2.2 catch

#### (1) spring

Two species of cephalopods, sepia japonicus and

Table 2.	list of	f fish	species	in	spring	and	Autumn
			speeres	***	001110		

Serial number	name	order	section	spring	autumn
1	Harengula zunasi		Clumaidaa	+	+
2	Clupanodon punctatus		Ciupeidae		+
3	Thrissa kammalensis			+	+
4	Engraulis japonicus	Clupeiformes		+	+
5	Thrissa mystax		Engraulidae	+	+
6	Setipinna taty			+	+
7	Pseudosciaena polyactis				+
8	Argyrosomus argentatus		Sciaenidae	+	+
9	Johnius belengerii			+	+
10	Sillago sihama		Sillaginidae	+	+
11	Enedrias fangi		pholidae	+	+
12	Lateolabrax maculatus		serranidae	+	
13	Liza haematocheila		Sphyraenidae	+	
14	Chaeturichthys hexanema			+	+
15	Cryptocentrus filifer				+
16	Chaeturichthys stigmatias			+	+
17	Parachaeturichthys polynema		Gobiidae		+
18	Tridentiger barbatus	Perciformes		+	
19	Triaenopogon barbatus				+
20	Odontamblyopus lacepedii			+	
21	Odontamblyopus rubicundus				+
22	Amoya pflaumi			+	
23	Ctenotrypauchen chinensis			+	+
24	Callionymus beniteguri		Perchidae	+	+
25	Trichiurus lepturus				+
26	Eupleurogrammus muticus		trichiuridae	+	+
27	Acanthopagrus schlegelii		sparidae		+
28	Sawara niphonia		CYBIIDAE	+	
29	Platycephalus indicus		0 1	+	+
30	Sebastods schlegelii	Scorpaeniformes	Scorpaenidae	+	+
31	Hexagrammos otakii		Hexapodae	+	
32	Cynoglossus joyneri			+	+
33	Cynoglossus joyneri		Cynoglossidae	+	
34	Paralichthys olivaceus	Flathsh	Paralichthyidae		+
35	Kareius bicoloratus		pleuronectidae	+	
36	akifugu vermicularis	T . 1			+
37	Fugu pseudommus	letraodontiformes	Tetraodontidae	+	+
38	Saurida elongata	Lantern fishes	Dactylogynidae	+	
39	Syngnathus acus Linnaeus	Gillinidae	Sauridae	+	

## Table 3. list of cephalopods

Serial number	Chinese name	Latin name	Latin name
1	Loligo japonica	Loligo japonica	Sepieidae
2	Octopus ocellatus	Octopus ocellatus	Octopodidae
3	Octopus ocellatus	Octopus variabilis	Octopodidae

octopus octopus, were captured in spring. The average density was 855 tail/h, 9.61kg/h. The highest is station 10, followed by station 8, and the lowest is station 12. According to the analysis of catches, the mantissa of cephalopods accounted for 24.18% of the total mantissa, 207 tails/h, biomass of 0.87 kg/h, and the average catch of cephalopod adults was 8.74 kg/h, 648 tails/h.

#### (2) autumn

Three species of cephalopods were captured in autumn, which were sepia japonicus, Octopus ocellatus and Octopus ocellatus. The average resource density was 113 tails/h, 1.723 kg/h. The biomass of cephalopods ranged from 0.112 to 6.900 kg/h, with the highest at station 10, followed by station 12 and station 1 with the lowest.

According to the analysis of catches, the mantissa of cephalopods accounted for 22.81% of the total mantissa, 26 tails/h, and the biomass was 0.093 kg/h. The average catch of adult cephalopods was 1.63 kg/h and 87 tails/h.

#### 3.2.3 Resource Density Assessment

#### (1) spring

In spring, the average catch of cephalopods was 855 tails/h, 9.61 kg/h; the average catch of juveniles was 207 tails/h, and the biomass was 0.87 kg/h; the average catch of adults was 648 tails/h, 8.74 kg/h. After conversion, the average resource density of cephalopods was 308.35 kg/ km<sup>2</sup> for adults and 7292 tails/km<sup>2</sup> for juveniles.

(2) autumn

In autumn, the average catches of cephalopods were 113 tails/h, 1.723 kg/h; the average catches of juveniles were 26 tails/h and the biomass was 0.093 kg/h; the average catches of adults were 87 tails/h, 1.630 kg/h. After conversion, the average resource density of cephalopod adults was 57.91 kg/km<sup>2</sup>, and that of juveniles was 909 tails/km<sup>2</sup>. According to the above results of cephalopod survey, the average resource density of cephalopods in spring and autumn is 183.13 kg/km<sup>2</sup> for adults and 4100 tails/km<sup>2</sup> for juveniles.

#### 3.3 crustacean Resources

#### 3.3.1 Species Composition

#### (1) spring

In spring, 15 species of crustaceans were captured, including 9 species of shrimps, 5 species of crabs and 1 species of Stomatopoda. See Table 4 for details. In terms of economic value, there are 5 species with higher economic value, accounting for 33.3% of the total species, 4 species with general economic value, accounting for 26.7% of the total species, and 6 species with low economic value, accounting for 40.0% of the total species.

The dominant species of crustaceans were Oratosquilla in spring, Portunus trituberculatus and Litopenaeus ternatus. The common species were Charybdis japonica, drum shrimp and drum shrimp. The others were common species and rare species.

(2) autumn

In autumn, 13 species of crustaceans were captured, including 6 species of shrimps, 6 species of crabs and 1 species of Stomatopoda, as shown in Table 5. In terms of economic value, there are 5 species with higher economic

Social number	Chinaga nama	castion		economic value	
Serial number	Chinese name	section	higher	Commonalty	Lower
1	Fenneropenaeus chinensis	Danaaidaa	$\checkmark$		
2	Trachypenaeus curvirostris	Penaeidae	$\checkmark$		
3	Alpheus heterocarpus	Dalaamanidaa		$\checkmark$	
4	Alpheus japonicus	Palaemonidae		$\checkmark$	
5	Palaemon gravieri	Palaemonidae		$\checkmark$	
6	C rangon crangon	Crangonidae		$\checkmark$	
7	Lysmata vittata	III an alasti da a			
8	Latreutes planirostris	Hippolylidae			√
9	Leptochela gracilis	Hyalinidae			$\checkmark$
10	Portunus trituberculatus	Dortumulidoo	$\checkmark$		
11	Charybdis japonica	Portunundae	$\checkmark$		
12	Carcinoplax vestita	Comentaridae			$\checkmark$
13	Eucrate crenata	Goneplacidae			$\checkmark$
14	Dorippe japonica	Dorippidae			1
15	Oratosquilla oratoria	Squillidae			

Table 4. list of crustacean species in Spring

value, accounting for 30.8% of the total species, 3 species with general economic value, accounting for 23.1% of the total species, and 6 species with low economic value, accounting for 46.2% of the total species.

Among the swimming animal communities surveyed, the dominant species were gobia japonica and crayfish crayfish; the important species were Portunus trituberculatus, Charybdis japonica, carinata nipponensis; the common species were Brachionus kryptonii; the others were common species and rare species.

#### 3.3.2 catch

#### (1)spring

In spring, 15 species of crustaceans were captured, including 9 species of shrimps, 5 species of crabs and 1 species of Stomatopoda. The average resource density of crustaceans was 1536 individuals/h and the average biomass of crustaceans was 18.28 kg/h. According to the catch analysis, the mantissa of shrimps accounted for 23.64% of the total number of shrimps, 355 tails/h, biomass of 1.256 kg/h, adult shrimps of 1147 tails/h, biomass of 16.014 kg/h; the mantissa of crabs accounted for 26.47% of the total number of crabs, 9 tails/h, biomass of 0.102 kg/h, and 25 tails/h of crabs, with biomass of 0.91 kg/h.

#### (2) autumn

In autumn, 13 species of crustaceans were captured, including 6 species of shrimps, 6 species of crabs and 1 species of Stomatopoda. The average catch of crustaceans was 1483 fish/h, 11.835 kg/h. According to the survey, the total biomass of the shrimp is 92.6 kg/h, which is 92.6 kg/ h of the adult shrimp.

#### 3.3.3 Resource Density Assessment

#### (1) spring

In spring, 15 species of crustaceans were captured. Among them, the larvae of shrimps were 355 individuals/ h, the biomass was 1.256 kg/h, the adult shrimp was 1147 tails/h, the biomass was 16.014 kg/h, the crab larvae were 9 tails/h, the biomass was 0.102 kg/h, and the adult crabs were 25 tails/h, and the biomass was 0.91 kg/h. After conversion, the average resource density of shrimp adults is 564.96 kg/km<sup>2</sup>, 40456 tails/km<sup>2</sup>; larva is 12525 tails/km<sup>2</sup>; crab adult resource density is 32.10 kg/km<sup>2</sup>, 2882 tails/km<sup>2</sup>, larva is 317 tails/km<sup>2</sup>.

(2) autumn

In autumn, 13 species of crustaceans were captured. Among them, the larvae of shrimps were 308 tails/h, the biomass was 0.986 kg/h, the adult shrimp was 1134 tails/ h, the biomass was 8.927 kg/h, and the crabs were all adults with the biomass of 1.922 kg/h. After conversion, the average resource density of shrimp adults is 314.90 kg/km<sup>2</sup>, 3997 ind/km<sup>2</sup>, larva is 10865 ind/km<sup>2</sup>, and crab adult resource density is 67.80 kg/km<sup>2</sup>, 1446 tail/km<sup>2</sup>.

According to the above results of spring and autumn survey, the average resource density of shrimp in spring and autumn is 439.93 kg / km<sup>2</sup> for adult and 11695 tail / km<sup>2</sup> for larva; the average resource density for crab is 49.95 kg / km<sup>2</sup> for adult and 159 / km<sup>2</sup> for larva.

#### 4. Discussion

According to the survey results in spring and autumn, the average density of fish eggs is 0.143 ind/M  $\sim$  3, and

Sarial numbera	Chinasa nama	soction	economic value		
Serial numbere	Chinese name	section	higher	Commonalty	Lower
1	Trachypenaeus curvirostris	Penaeidae	$\checkmark$		
2	Alpheus heterocarpus	Palaemonidae		√	
3	Alpheus japonicus			N	
4	Palaemon gravieri	Palaemonidae		N	
5	Lysmata vittata	II in a la ti da a			√
6	Latreutes planirostris	прротупаае			$\checkmark$
7	Portunus trituberculatus	Derterrulidee	$\checkmark$		
8	Charybdis japonica	Portunundae	$\checkmark$		
9	Carcinoplax vestita	Concellacidae			√
10	Eucrate crenata	Goneplacidae			$\checkmark$
11	Pinnothere.sp	Leguminosae			√
12	Paguridae	Hermit crabs			$\checkmark$
13	Oratosquilla oratoria	Squillidae	$\checkmark$		

Table 5. list of crustacean species in autumn

the average density of larvae and juveniles is 0.065 ind/ M ~ 3. The average stock density of adult fish is 264.50 kg/km<sup>2</sup>, and that of juvenile fish is 6244/km<sup>2</sup>. The average resource density of cephalopods was 183.13 kg/km<sup>2</sup> for adults and 4100 tails/km<sup>2</sup> for juveniles. The average adult resource density of shrimp is 439.93 kg/km<sup>2</sup>, larva is 11695 tail/km<sup>2</sup>; the average adult resource density of crab is 49.95 kg/km<sup>2</sup>, larva is 159 tail/km<sup>2</sup>.

#### 5. Conclusions

#### 5.1 Current Situation of Fishery Resources

It is clearly defined in the supplementary provisions of Chapter 10 and Article 20 of Chapter 3 of China's marine environmental protection law that spawning grounds, feeding grounds, wintering grounds, migration channels and breeding grounds of fish, shrimp and shellfish are all "fishery waters", and the fishery waters are the objects of marine ecological protection, especially the shallow waters of Bohai Bay, which are the spawning grounds and nursery grounds of main economic fishery organisms In particular, larval stage is more sensitive to environmental pollution than adult stage. Once these waters are seriously polluted, it will bring heavy consequences to fishery resources. The sea area around the assessment area is the spawning ground and feeding ground for many kinds of fishes in Bohai Sea, and there are many national and local aquatic germplasm resources protection areas, which should be the primary sensitive protection target.

According to the relevant research results <sup>[3]</sup>, according to the ecological types, the main economic fish resources in the evaluation area basically belong to two ecological types.

The first is the warm temperature widely distributed population, which has strong adaptability to the changeable hydrological environment, does not carry out long-distance migration, winters in the Bohai Sea, inhabits in estuaries, reefs and shallow waters, and moves seasonally in deep and shallow water with the change of environment. Generally, they swim to the shore to lay eggs in spring and summer, and swim to deeper waters in autumn and winter. The other is that the species with longdistance migration are mostly warm temperate and warm water species, with a large distribution range and obvious migration routes, while a few species migrate for a long distance.

#### **5.2 Protection Suggestions**

(1) The importance of strengthening publicity on the protection of fish resources

Fishery resources are precious natural wealth, and also the important material basis for human survival, with important scientific, ecological and economic value. Bohai Sea is known as the "cradle of hundreds of fish", but in recent years, the fishery resources have shown a trend of exhaustion, so it is imperative to protect fish <sup>[4]</sup>.

(2) The fishing structure of Bohai Sea should be adjusted scientifically and reasonably

We should appropriately reduce the number of offshore vessels, actively and steadily develop offshore operations, and gradually stabilize and restore the fishery resources in the Bohai Sea.

(3) Strengthen the management of marine environment

In recent years, the pollution sources of the Bohai Sea are increasing. As the Bohai Sea is a closed inner bay, the marine environment is deteriorating and the pollution is aggravating <sup>[5]</sup>. The competent state departments should strengthen the management of the marine environment, focusing on routine monitoring of fishing areas, trend monitoring of important sea areas, emergency monitoring of red tide prone areas and pollution accidents, and monitoring of marine nature reserves.

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## ARTICLE Zooplankton Diversity of a Soft-water and Highly De-mineralized Reservoir of Meghalaya (Northeast India): The Spatio-temporal Variations and Influence of Abiotic Factors

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#### 1. Introduction

Though zooplankton have attracted attention in several hydro-biological surveys from distant parts of India, the limnology literature indicates proliferation of casual reports with incomplete species inventories and inadequate data-analysis vis-a-vis the paucity of the diversity studies based on the detailed analysis of

#### ABSTRACT

Hydrobiological survey of a 'soft-water' and 'highly de-mineralized' reservoir of Meghalaya state of northeast India is undertaken to analyze zooplankton diversity with reference to the spatio-temporal variations and influence of abiotic factors. The littoral and limnetic zooplankton assemblages of this subtropical reservoir without aquatic vegetation reveal total 36 species, and record lower abundance, quantitative dominance of Rotifera, sub-dominance of Cladocera and Copepoda and moderate species diversity. Keratella cochlearis, Bosmina longirostris, Polyarthra vulgaris, Mesocyclops leuckarti, Conochilus unicornis and Asplanchna priodonta influence abundance, species diversity, dominance and equitability of zooplankton. We report differential spatial influence of individual abiotic factors with the relatively more importance at the limnetic region, and the canonical correspondence analysis registers 72.5% and 78.8% cumulative influence of 10 abiotic factors on the littoral and limnetic assemblages, respectively. The spatial differences of various diversity aspects and the influence of abiotic factors suggest habitat heterogeneity amongst the two regions. This study is a useful contribution to zooplankton diversity of the subtropical environs, and soft and de-mineralized waters in particular. Our results mark a distinct contrast to the lowest richness and abundance of zooplankton noted from India vide the preliminary 1990-91 survey of this reservoir.

zooplankton assemblages of the subtropical lacustrine environs of this country and north India<sup>[1,2]</sup>, and soft and de-mineralized waters of the Indian sub-region in particular. Referring to north India, the useful limited works of zooplankton diversity interest from northwest India (NWI) deal with the selected lakes and reservoirs of Jammu & Kashmir<sup>[3,4]</sup>, Himachal Pradesh<sup>[5]</sup> and Uttarakhand<sup>[6-8]</sup>. On the other hand, notable contributions

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<sup>[1,2,9]</sup> extend this lacuna to zooplankton diversity of the subtropical lacustrine environs of northeast India (NEI), while other surveys relate to a sub-tropical rice-field of Arunachal Pradesh<sup>[10]</sup> and a subtropical urban wetland of Meghalaya<sup>[11]</sup>. In addition, certain related works from NEI deal with the tropical floodplain lakes of Assam<sup>[12-14]</sup> and Manipur<sup>[15,16]</sup>.

Our study on zooplankton diversity of 'highly demineralized' subtropical reservoir of Meghalaya state of NEI, a follow-up of an earlier limited 1990-91 survey <sup>[17]</sup>, thus merits limnology interest for India and the Indian sub-region in light of the stated remarks. We analyze the littoral and limnetic zooplankton assemblages of this 'very soft-water' reservoir with reference to the spatiotemporal variations of species composition, richness, community similarities, abundance, species diversity, dominance and equitability as well as both the individual and cumulative influence of abiotic factors. Remarks are made on zooplankton diversity of the sampled reservoir in comparison with the related studies from the northwest India and NEI in particular, and the reports elsewhere from India as well as adjoining countries of the Indian sub-region. Besides, we indicate the salient temporal differences of zooplankton diversity vis-a-vis the preliminary survey <sup>[17]</sup> limited to the limnetic region of this reservoir.

#### 2. Material and Methods

#### 2.1 The Study Site

The present study is based on the limnological survey of a rainwater-fed reservoir (Figure 1, A-C; Lat. 25°34'N; Long.91°56'E, area ~10 ha; max. depth: 15 m; refereed as 'Shillong reservoir') conducted during January-December 2014. This reservoir is located at a distance of about 10 km from Shillong city, the capital of Meghalaya stats of northeast India; it serves as drinking water storage basin and lacks any aquatic vegetation and fish fauna.



Figure 1 A-C. A, map of India showing Meghalaya state of northeast India (red color); B, map of Meghalaya indicating location of the capital city of Shillong; C, map of Shillong reservoir indicating the littoral (blue color) and limnetic (red color) regions

#### 2.2 Methods of Study

Water samples were collected at monthly interval from the littoral and the limnetic regions. Water temperature, pH and specific conductivity were noted with the field probes, transparency was recorded with a Secchi disc, dissolved oxygen (DO) was estimated by Winkler's method, and alkalinity, hardness, calcium (Ca), magnesium (Mg), chloride (Cl), phosphate (PO<sub>4</sub>) and nitrate (NO<sub>3</sub>) were analyzed following APHA <sup>[18]</sup>. The rainfall data were collected from the local meteorological station. The qualitative and quantitative plankton samples were collected monthly from the two regions and were preserved in 5% formalin. The former were obtained by towing nylobolt plankton net (#40 µm), and were screened with a Wild Stereoscopic binocular microscope for isolation of zooplankton. Various zooplankton, mounted in polyvinyl alcohol-lactophenol mixture, were observed with Leica stereoscopic microscope (DM 1000); the different species were identified following the standard literature <sup>[19-27]</sup>. The quantitative plankton samples were obtained by filtering 25 L of water each through nylobolt plankton net (#40 µm). The quantitative enumeration of zooplankton was done by using a Sedgewick-Rafter counting cell and abundance was expressed as ind. 1<sup>-1</sup>.

#### 2.3 Data Analysis

The community similarities between monthly zooplankton assemblages were calculated vide Sørensen's index and the hierarchical cluster analysis was done using SPSS (version 20). Species diversity, dominance and evenness were computed vides Shannon-Weiner index, Berger-Parker index and  $E_1$  index, respectively <sup>[28,29]</sup>. The significance of the spatial and temporal variations of abiotic and biotic parameters was ascertained by twoway ANOVA. Pearson correlation coefficients for the littoral and limnetic regions ( $r_1$  and  $r_2$ , respectively) were calculated between abiotic and biotic parameters; p values (2-tailed) were calculated and their significance was noted after applying Bonferroni corrections. The canonical correspondence analysis was done (using XLSTAT 2015) to register cumulative influence of ten abiotic parameters (water temperature, rainfall, transparency, specific conductivity, pH, alkalinity, hardness, Cl, PO<sub>4</sub>, and NO<sub>3</sub>) on the littoral and limnetic zooplankton.

#### 3. Results

Water temperature, rainfall, transparency, pH, specific conductivity, DO, alkalinity, hardness, Ca, Mg, Cl, PO<sub>4</sub>, and NO<sub>3</sub> range between 11.0-21.0 °C, 0.6-780.5 mm, 1.6-2.2 m, 5.64-6.67, 11.5-19.2  $\mu$ S/cm<sup>-1</sup>, 7.0-8.8 mg l<sup>-1</sup>, 9.0-16.8 mg l<sup>-1</sup>, 6.0-13.2 mg l<sup>-1</sup>; 3.6-7.6 mg l<sup>-1</sup>, 0.2-0.9 mg l<sup>-1</sup>, 18.0-42.0 mg l<sup>-1</sup>, 0.072-0.190 mg l<sup>-1</sup> and 0.066-0.196 mg l<sup>-1</sup>, respectively during the study period (Table 1). The significance of spatio-temporal of variations of abiotic factors is indicated in Table 2.

The variations of richness and abundance of zooplankton, abundance of different groups and important species, and diversity indices are indicated in Table 3. The significance of spatio-temporal variations of biotic parameters is indicated in Table 4. The littoral and limnetic zooplankton reveal total 36 species, and record

Factors↓ Regions-	<b>→</b>	Litt	toral	Liı	nnetic
		Range	Average ± S.D	Range	Average ± S.D
Water temperature	<sup>0</sup> C	11.0-21.0	17.1±3.5	11.0-20.5	16.8±3.3
Rainfall	mm	0.6-780.5	211.6±223.7	0.6-780.5	211.6±223.7
Transparency	m	1.6-2.2	1.88±0.16	1.6-2.2	1.93±0.16
рН		5.65-6.67	6.21±0.22	5.64-6.55	6.16±0.26
Specific conductivity	µScm <sup>-1</sup>	11.5-19.2	15.8±2.5	12.0-19.0	15.8±2.2
DO	mg l <sup>-1</sup>	7.0-8.6	7.8±0.4	7.2-8.8	7.9±0.4
Alkalinity	mg l <sup>-1</sup>	9.0-16.8	11.8±2.3	9.2-16.4	11.7±2.1
Hardness	mg l <sup>-1</sup>	6.2-13.2	8.6±2.2	6.0-13.0	8.7±2.2
Ca	mg l⁻¹	3.8-7.6	5.3±1.2	3.6-7.0	5.0±1.3
Mg	mg l <sup>-1</sup>	0.2-0.9	0.2±0.3	0.2-0.8	0.4±0.2
Cl	mg l <sup>-1</sup>	19.0-42.0	30.4±6.7	18.0-40.0	29.4±6.4
PO <sub>4</sub>	mg l⁻¹	0.072-0.190	$0.128 \pm 0.035$	0.080-0.190	0.128±0.031
NO <sub>3</sub>	mg l <sup>-1</sup>	0.066-0.196	$0.108 \pm 0.040$	0.070-0.188	0.110±0.036

Table 1. The spatio-temporal variations of abiotic factors

	1 1 6	
Parameters	Regions	Months
Water temperature	-	F <sub>11,23</sub> =244.629, P=1.62E-11
Transparency	$F_{1,23} = 17.742, P = 0.001$	$F_{11,23} = 9.069, P = 0.0003$
pH	-	F <sub>11,23</sub> =196.986, P=5.52E-11
Specific conductivity	-	F <sub>11,23</sub> = 66.715, P=1.94E-08
DO	F <sub>11,23</sub> = 10.632, P=0.0076	F <sub>11,23</sub> = 30.779, P=1.17E-06
Alkalinity	-	$F_{11,23} = 129223, P = 5.47E-10$
Hardness	-	$F_{11,23}$ = 342.936, P = 2.67E-12
Ca	F <sub>1,23</sub> = 27.770, P=0.0002	$F_{11,23} = 78.814, P = 7.93E-09$
Mg	-	$F_{11,23} = 17.551, P = 2.06E-05$
Cl	F <sub>1,23</sub> = 15.531, P=0.0023	$F_{11,23} = 220.202, P = 3.01E-11$
$PO_4$	-	F <sub>11,23</sub> = 157.459, P = 1.87E-10
NO <sub>3</sub>	-	$F_{11,23} = 195.429, P = 5.96E-11$

Table 2. The spatio-temporal significance of abiotic factors

(-) indicates insignificant variations

	1 1		1	
QUALITATIVE	Litte	oral region	Lir	nnetic region
Zooplankton richness Community similarities	36; 15-22 43	2 (18±2) species .7-86.5%	34; 15-	25 (19±3) species 45.2-82.3%
Rotifera richness	22; 7-13	(11±2) species	20; 10-	15 (11±2) species
	QUAN	TITATIVE		
Net Plankton ind. 1 <sup>-1</sup>	281-1194	647±234	275-560	394±99
Zooplankton ind. 1 <sup>-1</sup> Percentage of net plankton	113-307 18.5-67.0	218±70 37.8±17.3	74-238 28.6-73.1	177±51 46.6±15.0
Species Diversity	1.822-2.535	$2.076 \pm 0.194$	1.885-2.706	2.133±0.209
Dominance	0.186-0.411	$0.302 \pm 0.057$	0.200-0.398	$0.298 \pm 0.062$
Evenness	0.646-0.861	$0.719 \pm 0.053$	0.680-0.841	0.733±0.043
	Differ	ent Groups		
Rotifera ind. l <sup>-1</sup> Percentage of zooplankton	64-228 51.1-76.2	132±54 59.0±7.9	50-163 51.7-70.9	110±33 62.6±5.2
Cladocera ind. l <sup>-1</sup> Percentage of zooplankton	23-84 12.4-28.1	44±19 20.4±4.6	13-56 14.2-25.4	33±13 18.4±3.2
Copepoda ind. l <sup>-1</sup> Percentage of zooplankton	19-57 9.04-29.6	38±12 18.2±5.7	12-50 12.2-26.3	30±11 16.6±3.7
Rhizopoda ind. l <sup>-1</sup>	1-12	4±3	1-9	4±2
	Import	tant Species		
<i>Keratella cochlearis</i> ind. l <sup>-1</sup>	21-123	62±27	22-78	51±18
Bosmina longirostris ind. l <sup>-1</sup>	20-79	41±18	10-54	32±13
Polyarthra vulgaris ind. l <sup>-1</sup>	10-82	29±23	3-42	22±12
Mesocyclops leuckarti ind. l <sup>-1</sup>	12-51	30±13	9-38	22±9
Conochilus unicornis ind. l-1	2-31	12±10	2-32	12±9
Asplanchna priodonta ind. l <sup>-1</sup>	5-31	16±8	4-22	13±5

Table 3.	The spatio-	-temporal	variations	of zoop	lankton
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36 and 34 species at the two regions, respectively. The monthly zooplankton richness ranges between 15-22 and 15-25 species (Figure 2) and register 43.7-86.5% and 45.2-82.3% community similarities, and Rotifera record

richness between 7-13 and 10-15 species, at the two regions, respectively. Zooplankton hierarchical cluster analysis (Figures 3-4) exhibits differences in the cluster groupings at the two regions.



Figure 2. Monthly variations of zooplankton species richness

Parameters	Regions	Months
Zooplankton richness	-	$F_{11,23} = 3.162, P = 0.0344$
Rotifera richness	-	$F_{11,23} = 3.453, P = 0.0255$
	Abundance	
Zooplankton	$F_{1,23} = 15.849, P = 0.002$	$F_{11,23} = 11.904, P = 0.0001$
Rotifera	$F_{1,23} = 6.895, P = 0.015$	$F_{11,23} = 9.859, P = 0.0003$
Cladocera	-	-
Copepoda	F <sub>1,23</sub> = 49.107, P = 2.25E-05	$F_{11,23} = 34.348, P = 6.68E-07$
Zooplankton species diversity	-	$F_{11,23} = 17.253, P = 2.25E-05$
Dominance	-	$F_{11,23} = 3.807, P = 0.0181$
Evenness	-	$F_{11,23} = 9.657, P = 0.0004$
	Important species	
Keratella cochlearis	$F_{1,23} = 6.037, P = 0.032$	$F_{11,23} = 8.087, P = 0.0008$
Bosmina longirostris	-	-
Polyarthra vulgaris	-	$F_{11,23} = 4.354, P = 0.011$
Mesocyclops leuckarti	$F_{1,23} = 25.361, P = 0.0004$	$F_{11,23} = 16.132, P = 3.14E-05$
Conochilus unicornis	-	$F_{11,23} = 17.803, P = 1.92E-05$
Asplanchna priodonta	-	$F_{11,23} = 6.203, P = 0.003$

Table 4.	The spatio-	-temporal	significance	of zoop	lankton
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(-) insignificant variations

Zooplankton record abundance ranging between 113-307 and 74-238 ind.  $I^{-1}$  (Figure 5), comprise between 18.5-67.0% and 28.6-73.1% of net plankton, indicate species diversity ranging between 1.822-2.535 and 1.885-2.706 (Figure 6), and dominance, and evenness range between 0.186-0.411 and 0.200-0.398 (Figure 7) and 0.646-0.861 and 0.680-0.841 (Figure 8) at the two regions, respectively. *Keratella cochlearis* (62±27, 51±18 ind.  $I^{-1}$ ), *Bosmina longirostris* (41±18, 32±13 ind.  $I^{-1}$ ), *Polyarthra vulgaris* (29±23, 22±12 ind.  $I^{-1}$ ), *Mesocyclops leuckarti* (30±13, 22±9 ind.  $I^{-1}$ ), *Conochilus unicornis* (12±10, 12±9 ind.  $I^{-1}$ ) and *Asplanchna priodonta* (16±8, 13±5 ind. 1<sup>-1</sup>) indicate quantitative importance at the two sampled regions (Figures 9-10).

Rotifera record abundance ranging between 71-285 and 57-212 ind.  $I^{-1}$  (Figure 11) and comprise between 51.1-76.2 and 51.7-70.9% of zooplankton; Cladocera (23-84 and 13-56 ind.  $I^{-1}$ ) and Copepoda (19-57 and 12-50 ind.  $I^{-1}$ ) and comprise between 12.4-28.1 and 14.2-25.4% and 9.04-29.6 and 12.2-26.3% of zooplankton abundance at the littoral and limnetic regions, respectively (Figures 12-13). Rhizopoda and Gastrotricha record poor abundance at the two regions.



#### Dendrogram using Average Linkage (Between Groups)

Figure 3. Hierarchical cluster analysis of zooplankton assemblages (Littoral region)



Dendrogram using Average Linkage (Between Groups)

Figure 4. Hierarchical cluster analysis of zooplankton assemblages (Limnetic region)



Figure 5. Monthly variations of zooplankton abundance



Figure 6. Monthly variations of zooplankton species diversity



Figure 7. Monthly variations of zooplankton dominance



Figure 8. Monthly variations of zooplankton evenness



Figure 9. Monthly variations of abundance of important species (Littoral region)



Figure 10. Monthly variations of abundance of important species (Limnetic region)



Figure 11. Monthly variations of Rotifera abundance



Figure 12. Monthly variations of Cladocera abundance



Figure 13. Monthly variations of Copepoda abundance



Figure 14. CCA coordination biplot of zooplankton and abiotic factors (Littoral region)

#### Abbreviations

Abiotic factors: Alk (alkalinity), Cl (chloride), Hard (hardness), NO<sub>3</sub> (nitrate), pH (hydrogen-ion concentration), PO<sub>4</sub> (phosphate), Rain (rainfall), Scon (specific conductivity), Trans (transparency), Wt (water temperature).

**Biotic factors:** A pr (*Asplanchna priodonta* abundance), B lon (*Bosmina longirostris* abundance), C un (*Conochilus unicornis* abundance), Cld (Cladocera abundance), Cop (Copepoda abundance), Dom (dominance), Evn (evenness), K ch (*Keratella cochlearis* abundance), M leu (*Mesocyclops leuckarti* abundance), P vul (*Polyarthra vulgaris* abundance) Rot (Rotifera abundance), RR (Rotifera richness), SD (species diversity), Zoo (Zooplankton abundance), ZR (Zooplankton richness).

Of the recorded abiotic factors, water temperature exerts an inverse influence on zooplankton ( $r_2$ = -0.674, p = 0.0162), and Rotifera ( $r_2$ = -0.708, p = 0.0100) richness at the limnetic region. It records a positive influence on abundance of zooplankton ( $r_1$ =0.734, p = 0.0066;  $r_2$ = 0.912, p < 0.0001), Copepoda ( $r_1$ =0.833, p = 0.0008;  $r_2$ = 0.912, p < 0.0001) and *Mesocyclops leuckarti* ( $r_1$ =0.890, p = 0.0001;  $r_2$ = 0.901, p < 0.0001) at the two regions; an inverse influence on zooplankton evenness at the littoral region ( $r_1$ = -0.714, p = 0.0091); and positive influence on

abundance of Rotifera ( $r_2 = 0.779$ , p = 0.0028), Cladocera ( $r_2 = 0.836$ , p = 0.0007), *Bosmina longirostris* ( $r_2 = 0.855$ , p = 0.0004), and *Polyarthra vulgaris* ( $r_2 = 0.866$ , p = 0.0003) abundance at the limnetic region. Rainfall registers positive correlation on abundance of *P. vulgaris* ( $r_1 = 0.814$ , p = 0.0013), *M.leuckarti* ( $r_1 = 0.694$ , p = 0.0123) and *Conochilus unicornis* ( $r_1 = 0.832$ , p = 0.0008) at the littoral region, and on abundance of Cladocera ( $r_2 = 0.887$ , p = 0.0001), *B.longirostris* ( $r_2 = 0.874$ , p = 0.0002), and *P. vulgaris* ( $r_2 = 0.872$ , p = 0.0002) at the limnetic region.



Figure 15. CCA coordination biplot of zooplankton and abiotic factors (Limnetic region)

#### Abbreviations

Abiotic factors: Alk (alkalinity), Cl (chloride), Hard (hardness), NO<sub>3</sub> (nitrate), pH (hydrogen-ion concentration), PO<sub>4</sub> (phosphate), Rain (rainfall), Scon (specific conductivity), Trans (transparency), Wt (water temperature).

**Biotic factors:** A pr (*Asplanchna priodonta* abundance), B lon (*Bosmina longirostris* abundance), C un (*Conochilus unicornis* abundance), Cld (Cladocera abundance), Cop (Copepoda abundance), Dom (dominance), Evn (evenness), K ch (*Keratella cochlearis* abundance), M leu (*Mesocyclops leuckarti* abundance), P vul (*Polyarthra vulgaris* abundance) Rot (Rotifera abundance), RR (Rotifera richness), SD (species diversity), Zoo (Zooplankton abundance), ZR (Zooplankton richness).

pH affirms positive correlation with abundance of Copepoda ( $r_1 = 0.879$ , p = 0.0002;  $r_2 = 0.856$ , p = 0.0001) and *M. leuckarti* ( $r_1 = 0.860$ , p = 0.0001;  $r_2 = 0.816$ , p = 0.0012) at the two regions, while it indicates a positive correlation with abundance zooplankton ( $r_2 = 0.703$ , p = 0.0108), Cladocera ( $r_2 = 0.755$ , p = 0.0045), *B. longirostris* ( $r_2 = 0.763$ , p = 0.0039), and *P. vulgaris* ( $r_2 = 0.788$ , p = 0.0023) at the limnetic region. Specific conductivity registers positive correlation on abundance of *K. cochlearis* ( $r_1 = 0.769$ , p = 0.0035;  $r_2 = 0.864$ , p = 0.0035)

0. 000312) at the two region and that of *A. priodonta* ( $r_1$  =0.861, p = 0.00013) at the littoral region. Transparency records an inverse influence on abundance of Copepoda ( $r_2$  = -0.804, p = 0.0016), *P. vulgaris* ( $r_2$  = -0.788, p = 0.0002), *M. leuckarti* ( $r_2$ =-0.772, p = 0.0033) and positive influence on zooplankton dominance ( $r_2$ =0.675, p = 0.0160) only at the limnetic region.

Cl registers positive correlation on abundance of zooplankton ( $r_1 = 0.731$ , p = 0. 0067;  $r_2 = 0.731$ , p = 0. 0067), Copepoda ( $r_1 = 0.767$ , p = 0. 0036;  $r_2 = 0.785$ , p = 0.

0025), *M. leuckarti* ( $r_1$ =0.845, p = 0.0005;  $r_2 = 0.749$ , p =0.0051) and P. vulgaris ( $r_1 = 0.681$ , p = 0.0148;  $r_2 = 0.803$ , p = 0.0017) at the littoral and limnetic regions. Besides, it records a positive influence on abundance of C. unicornis  $(r_1 = 0.744, p = 0.0055)$  at the littoral region and that of Cladocera ( $r_2 = 0.785$ , p = 0.00245) and *B. longirostris*  $(r_2 = 0.861, p = 0.0003)$  at the limit region. PO<sub>4</sub> exerts positive correlations with abundance of *P. vulgaris*  $(r_1$ = 0.802, p = 0. 0017;  $r_2$  = 0.815, p = 0. 0012) at both the regions. It records an inverse influence on Rotifera richness ( $r_2$ = -0.697, p = 0. 0152), and records positive correlations with abundance of zooplankton ( $r_2 = 0.747$ , p = 0.0052), Cladocera ( $r_2 = 0.918$ , p < 0.0001) and B. *longirostris* ( $r_2 = 0.909$ , p < 0.0001) at the limit region. Besides, PO<sub>4</sub> exerts positive correlations with abundance *M.leuckarti* ( $r_1$  =0.706, p = 0.0103) and *C. unicornis* ( $r_1$ = 0.744 p = 0.0055) at the littoral region. NO<sub>3</sub> shows limited importance at the limnetic region with positive correlations on abundance of Cladocera ( $r_2 = 0.703$ , p =0. 0108) and B. *longirostris* ( $r_2 = 0.681$ , p = 0.0148). The canonical correspondence analysis (CCA) registers the differential cumulative influence of 10 abiotic factors on the littoral (72.5%) and limnetic (78.8%) zooplankton assemblages (Figures 14-15).

#### 4. Discussion

The present study records one of the lowest specific conductivity known from any aquatic environ of the Indian sub-continent <sup>[1,2,11,17,30,31]</sup>. This notable feature, indicating highly de-mineralized nature of the subtropical Shillong reservoir, is attributed to the weathered and leached nature of rocks and soils of the catchment area due to high rainfall <sup>[2]</sup>. Besides, this rainwater-fed reservoir indicates very soft, acidic, calcium poor and oxygenated waters, and lower concentrations of PO<sub>4</sub>, NO<sub>3</sub>, Mg and Cl. ANOVA registers significant spatio-temporal variations of transparency, DO, Ca and Cl, while water temperature, pH, specific conductivity, alkalinity, hardness, Mg, Cl, PO<sub>4</sub> and NO<sub>3</sub> record significant temporal variations.

Total 36 species observed vide our study reveal the relatively less diverse zooplankton of Shillong reservoir devoid of any aquatic vegetation; the richness marks a distinct threefold increase than earlier survey <sup>[17]</sup>. The species tally is higher as compared with the reports from the lacustrine environs of Jammu & Kashmir <sup>[3]</sup>, Himachal Pradesh <sup>[8]</sup> and Uttarakhand <sup>[6,32-34]</sup> from NWI; elsewhere from India from Karnataka <sup>[35]</sup>, Tamil Nadu <sup>[36-38]</sup> and Telangana <sup>[39]</sup>; and the reports from Bhutan <sup>[31]</sup> and Nepal <sup>[40]</sup>. Our study, however, indicates lower richness than observed from certain subtropical lakes and reservoirs of NEI <sup>[1,2,9]</sup>. Total 36 and 34 species examined from the

littoral and limnetic regions, respectively register  $\sim 97\%$  community similarity and depict overall homogeneity of zooplankton composition amongst the two regions.

Zooplankton richness follows oscillating monthly variations; it records peaks during (post-monsoon) and winter (January) at the littoral and limnetic regions respectively, and registers (vide ANOVA) insignificant spatial and significant temporal variations. Rotifera significantly influence the littoral ( $r_1 = 0.975$ , p < 0.0001) and limnetic ( $r_2 = 0.918$ , p=0.0002) zooplankton monthly richness, and register (vide ANOVA) insignificant spatial and significant temporal variations. The notable paucity of Brachionus species amongst Rotifera is attributed to soft and acidic waters of the sampled reservoir concurrent with the reports from NEI [1,2,9,11,41,42]. The littoral and limnetic zooplankton assemblages record 43.7-86.5% and 45.2-82.3% community similarities (vide Sørensen's index), respectively. Peak similarities between February-July and May-August collections, the differential monthly cluster groupings noted vide the hierarchical cluster analysis, and 72.9% and 60.6% instances indicating similarity values between 51-70% at the two regions suggest heterogeneity of zooplankton composition within the two regions individually. This generalization is supported by the closer affinities between February-July-December and again between January-October collections at the littoral region while zooplankton indicate closer affinity during May-August at the limnetic region, while March > August, and March assemblages record maximum divergence at the two regions, respectively.

Our results highlight lower zooplankton abundance with significant spatio-temporal variations (vide ANOVA). Lower abundance, attributed to the 'de-mineralized' nature of the sampled reservoir, concurs with the reports from aquatic environs of NEI [1,2,9,11,13,14], and Bhutan [31] but marks a distinct contrast to the lowest density  $(13\pm6 \text{ ind. } l^{-1})$ reported from India vide earlier survey <sup>[17]</sup>. Zooplanktons comprise notable quantitative component of net plankton in contrast to insignificant role noted earlier <sup>[17]</sup>. The present study records bimodal density variations at the littoral region, and registers maxima during pre-monsoon and post-monsoon); this pattern is less distinctive at the limnetic regions due to limited density differences during May-October in particular. Nevertheless, the stated trends affirm positive correlation of zooplankton abundance with water temperature at the both the regions concurrent with the results of NEI<sup>[1]</sup>, Himachal Pradesh<sup>[5]</sup>, Uttarakhand<sup>[34]</sup> and West Bengal<sup>[43]</sup>. Besides, abundance registers positive correlation with Cl at the two regions and with rainfall, pH and PO<sub>4</sub> at the limnetic region. Individual abiotic factors thus depict limited and differential spatial influence on

zooplankton abundance. The periods of higher abundance concur with the reports from Arunachal Pradesh <sup>[10]</sup>, and Uttarakhand <sup>[5]</sup> but differ from winter maxima known from Himachal Pradesh <sup>[8]</sup> and Uttarakhand <sup>[7,34]</sup>,while premonsoon peaks correspond with the report from Uttar Pradesh <sup>[44]</sup>.

Zooplankton depicts quantitative dominance of Rotifera, and sub-dominance of Cladocera and Copepoda; the significance pattern differs from the collective quantitative importance of Rotifera, Cladocera and Copepoda<sup>[2,44]</sup> and that of Rotifera and Copepoda<sup>[1]</sup>, and dominance of Copepoda<sup>[7,9]</sup>. Rotifera predominance is attributed to the short turn-over over enabling these microinvertebrates to dominate over other zooplankton groups, and life history r-strategies and the opportunistic character <sup>[45-47]</sup>. Rotifera importance concurs with the different reports from India<sup>[4,11-16,26,36,37,48-51]</sup>. Keratella cochlearis, Bosmina longirostris, Polyarthra vulgaris, Mesocyclops leuckarti, Conochilus unicornis and Asplanchna priodonta collectively (74.7±24.9% and 83.5±7%) exert significant influence ( $r_1 = 0.995$ , p < 0.0001;  $r_2 = 0.995$ , p <0.0001) on zooplankton abundance at the two regions. K.cochlearis ( $r_1 = 0.793$ , p =0.0062), B. longirostris ( $r_1$ = 0.797, p =0.0058), A. priodonta (r<sub>1</sub>=0.871, p=0.0010) and *P. vulgaris* ( $r_1 = 0.818$ , p =0.0038) individually influence zooplankton abundance at the littoral region, while K. cochlearis ( $r_2 = 0.693$ , p =0.0115), B. longirostris  $(r_2 = 0.883, p = 0.0001), C. unicornis (r_2 = 0.754, p$ =0.0046) and *P. vulgaris* ( $r_2 = 0.890$ , p =0.0001) influence abundance at the limnetic region. We categorize the stated species as 'specialist' in contrast to 'generalist' nature of rest of the species with lower densities; the former differ from lack of such species reported vide earlier survey <sup>[17]</sup>.

Rotifera follows bimodal monthly density variations concurrent with that of zooplankton, significantly influence abundance of the latter at the littoral ( $r_1 = 0.941$ , p < 0.0001) and limnetic (r<sub>2</sub>=0.940, p < 0.0001) regions, and ANOVA registers significant spatio-temporal variations. The rotifers indicate higher abundance at the littoral > limnetic region during April-June and October, and record less spatial differences during the rest of the study period. This group indicates peak abundance during April at both the stations, depicts maxima during October at the littoral region and period of nearly concurrent high abundance during September-October at the limnetic region, and registers significant positive correlation with water temperature only at the limnetic region. The summer Rotifera peaks correspond with the reports from Himachal Pradesh<sup>[8]</sup>, Bihar<sup>[52]</sup> and West Bengal<sup>[43]</sup>, and differ from winter maxima recorded Assam<sup>[12,13,53]</sup> and Manipur<sup>[14,15]</sup>. Keratella cochlearis, the dominant 'specialist' species, influences rotifer abundance at the two regions ( $r_1 = 0.843$ , p=0.0022;  $r_2 = 0.830$ , p = 0.0008). Besides, Asplanchna *priodonta* ( $r_1$ =0.899, p=0.0004;  $r_2$ =0.710, p=0.0097), *Conochilus unicornis* ( $r_1 = 0.662$ , p =0.0190;  $r_2 = 0.761$ , p =0.0040) and *Polyarthra vulgaris* ( $r_1 = 0.842$ , p =0.0022;  $r_2 = 0.749$ , p =0.0051) influence Rotifera abundance. K. cochlearis abundance depicts positive influence of specific conductivity; P. vulgaris records positive correlation with rainfall, Cl and PO<sub>4</sub> at both the regions but indicates positive correlation with pH, and an inverse correlation with rainfall at the limnetic region; and A. priodonta registers positive correlation with specific conductivity while C. unicornis registers positive influence of rainfall, Cl and PO<sub>4</sub> at the littoral region. Abiotic factors thus depict the differential spatial influence on Rotifera and its notable species.

Cladocera indicates lower abundance at the littoral > limnetic regions and register insignificant spatio-temporal variations (vide ANOVA). Our study records distinctly higher abundance of this group than earlier survey <sup>[17]</sup>, while it concurs with the reports from Meghalaya<sup>[2]</sup> and Assam<sup>[14]</sup>. The bimodal periodicity with peak in October and maxima in June at the littoral region, and broadly unimodal pattern at the limnetic region with peak during June depict spatial differences in quantitative variations of this group; the abundance pattern at the latter region affirms positive correlation with water temperature and rainfall. Besides, Cladocera abundance registers positive correlations with pH, Cl and PO<sub>4</sub> at the limnetic regions, and with NO3 at the littoral region. Bosmina *longirostris* (r<sub>1</sub>= 0.995, p< 0.0001, r<sub>2</sub>= 0.997, p < 0.0001) exclusively influences Cladocera densities, records peaks during October and June at the two regions respectively, and ANOVA registers insignificant spatio-temporal quantitative variations. B. longirostris abundance is positively influenced by water temperature, rainfall, pH, Cl and PO<sub>4</sub> at the limnetic region, and by NO<sub>3</sub> at the littoral region. Our results thus record the differential influence of individual abiotic factors on Cladocera and B. longirostris, and the limited importance at the littoral region in particular. The periods of higher cladoceran abundance differ from the winter maxima<sup>[2,12]</sup>, and higher abundance during May-July at the limnetic region concurs with reports from Meghalaya<sup>[1,2]</sup> and Uttarakhand<sup>[7]</sup>.

Copepoda depicts the relatively wider density variations at the littoral region and registers significant spatio-temporal quantitative variations (vide ANOVA). The littoral and limnetic copepod assemblages follow broadly unimodal and distinctly unimodal abundance patterns, respectively. The relatively higher Copepoda abundance from May-October and peaks during August (monsoon) at both the regions is supported by positive correlation with water temperature. Besides, abundance of this group registers positive correlation with pH and Cl at the two regions, and depicts an inverse influence of transparency at the limnetic region. The monsoon peaks and unimodal periodicity of Copepoda differ from pre-monsoon peak and autumn maxima reported from Meghalava<sup>[2,11]</sup>, Andhra Pradesh<sup>[54]</sup> and Karnataka<sup>[55]</sup>. The copepod abundance is influenced by the cyclopoid *Mesocyclops leuckarti* ( $r_1 = 0.997$ , p < 0.0001;  $r_2 = 0.985$ , p < 0.0001); this species registers positive correlation with water temperature, pH and Cl at the two regions, records positive influence of rainfall and PO<sub>4</sub> at the littoral region and depicts an inverse influence of transparency at the limnetic region. Our results thus affirm differential but concurrent spatial importance of the abiotic factors both on Copepoda and M. leuckarti. The importance of Cyclopidae and occurrence of nauplii throughout the study affirming active Copepoda reproduction are attributed to the prevalence of stable environmental conditions for these 'k-strategists' [56]. Amongst other zooplankton groups, Rhizopoda records poor abundance <sup>[1,2,11,14]</sup>, while Gastrotricha depicts quantitative insignificance.

The differential spatial influence of individual abiotic factors on zooplankton, the constituent groups and important species, and the relative importance of water temperature, rainfall, pH, Cl and PO<sub>4</sub> on limnetic zooplankton assemblages suggest habitat heterogeneity amongst the littoral and limnetic regions. The limited influence on richness concurs with the results from NEI <sup>[1,2,9,11,14,16]</sup>. Overall influence on zooplankton abundance differs from much limited role of abiotic factors [1,2,9,13,14] and lack of importance of any individual factor <sup>[17]</sup>. The canonical correspondence analysis registers broadly concurrent cumulative influence of 10 abiotic factors on the littoral (72.5%) and limnetic (78.8%) zooplankton; the CCA biplots register ~ 42% and ~30%, and ~ 51% and ~28% influence of abiotic factors along axis 1 and 2, respectively at the two regions, respectively. The CCA results record influence of alkalinity and hardness on abundance of zooplankton and Rotifera; specific conductivity on Asplanchna priodonta, transparency on Keratella cochlearis, and NO<sub>3</sub> on Cladocera and Bosmina longirostris abundance, and on zooplankton dominance and evenness at the littoral region. On the other hand, the CCA biplot indicates influence of alkalinity and harness on zooplankton abundance; water temperature influences Conochilus unicornis abundance; pH, rainfall and Cl influences abundance of Bosmina longirostris and Polyarthra vulgaris, and specific conductivity, transparency and NO<sub>3</sub> exert influence on abundance of Rotifera, *K. cochlearis* and *A. priodonta* at the limnetic region. High cumulative influence at the two regions broadly concurs with the reports from Meghalaya <sup>[1,2]</sup> and Mizoram <sup>[9]</sup> but deviates from the differential spatial cumulative influence an urban wetland of Meghalaya <sup>[11]</sup>. In spite of the spatial differences, our results affirm importance of both the individual and cumulative influence of abiotic factors on zooplankton assemblages.

Zooplankton record moderate species diversity at the limnetic > littoral regions; ANOVA registers insignificant spatial and significant temporal diversity variations. Our results depict higher diversity values during January-February (peak in January) at both the regions, and indicate H' values > 2.0 throughout this study except during March and October at the former region and during January-February, July-September and November-December at the littoral region. Our study registers lower species diversity than the reports of the selected lakes and reservoirs of Meghalaya<sup>[1,2]</sup> and the relatively lower values than the report from a reservoir of and Mizoram <sup>[9]</sup>; the differences are attributed to lower zooplankton richness in the sampled reservoir. The concurrence of high diversity values with low densities of zooplankton and important species, affirmed by significant inverse correlations with abundance of zooplankton ( $r_1$ = -0.675, p = 0.0160), Rotifera (r<sub>1</sub>= -0.678, p = 0.0154), K. cochlearis  $(r_1 = -0.738, p = 0.0061), A. priodonta (r_1 = -0.665, p = -0.665)$ 0.0168) and collective abundance of 'specialist species ( $r_1$ = -0.727, p = 0.0074) at the littoral region, is hypothesized to fine niche portioning in combination with habitat heterogeneity<sup>[57]</sup>. The diversity affirms significant inverse relationship with zooplankton dominance ( $r_1 = -0.675$ , p = 0.0160) at the littoral region. It registers inverse correlation with abundance of K. cochlearis ( $r_2 = -0.675$ , p = 0.0160), and records postive correlation with richness of zooplankton ( $r_2 = 0.883$ , p = 0.0001) and Rotifera ( $r_2 =$ 0.832, p = 0.0008) at the limnetic region. The significant positive correlation of species diversity with evenness  $(r_1 = 0.865, p = 0.0012; r_2 = 0.8911, p < 0.0001)$  at the two regions affirms concurrence of diverse zooplankton assemblages during the periods of high equitability.

Zooplanktons indicate low dominance and high evenness; both register insignificant spatial and significant temporal variations (vide ANOVA). The dominance records peak during April and maxima during December at the littoral region, while it records peak during October and maxima during March at the limnetic region. Zooplankton dominance is positively influenced by abundance of *K. cochlearis* ( $r_1$ = 0.708, p = 0.0098) at the littoral region. Evenness is inversely influenced by abundance of zooplankton ( $r_1$ = -0.739, p = 0.0060), Rotifera ( $r_1$ = -0.687, p = 0.0136), K. cochlearis ( $r_1$ = -0.817, p = 0.0011) and A. priodonta ( $r_1 = -0.716$ , p =0.0199) at the littoral region; it is positively influenced by Rotifera richness ( $r_2 = 0.698$ , p = 0.0116) and registers inverse correlation with K. cochlearis abundance  $(r_2 =$ -0.665, p = 0.0183) at the limit region. The periods of higher evenness and lower dominance correspond with low densities and equitable abundance of the 'generalist' species, while the selected 'specialist' species result in higher dominance during certain months. This trend concurs with the reports from the floodplain lakes of NEI <sup>[12-16,26]</sup> and an urban wetland of Meghalava <sup>[11]</sup>. In general, lower zooplankton dominance and higher equitability affirms that the sampled reservoir has resources for utilization by majority of species due to high amount of niche overlap<sup>[58]</sup>.

#### 5. Conclusions

The lowest specific conductivity known from any aquatic environ of the Indian sub-continent, and very soft, acidic and calcium poor waters are notable features of the subtropical Shillong reservoir. This reservoir devoid of any aquatic vegetation reveals low zooplankton richness with overall homogeneity of species composition amongst the two regions, while the community similarities and the hierarchical cluster groupings suggest heterogeneity of zooplankton composition within the two regions individually. Lower zooplankton abundance attributed to the 'de-mineralized' waters, the dominance of Rotifera, bimodal zooplankton and Rotifera temporal density variations, and the 'specialist natures of six planktonic species are notable features. The subdominant Cladocera record bimodal and broadly unimodal density variations, and Copepoda depict broadly unimodal and distinctly unimodal abundance patterns at the two regions respectively. Zooplankton record moderate species diversity, and lower dominance and higher equitability indicate temporal variations influenced by the specialist species. The spatial differences of richness, abundance, diversity indices and the influence of abiotic factors hypothesize habitat heterogeneity amongst the two regions. This study merits regional and national interest for zooplankton diversity of the subtropical environs, and the soft and de-mineralized waters in particular.

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## **ARTICLE Effects of Different Combinations of Two Spices: Clove and Nutmeg Seed Extracts on Antioxidants Levels in African Catfish (Clarias gariepinus)**

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ARTICLE INFO	ABSTRACT
Article history Received: 23 September 2021 Accepted: 8 October 2021 Published Online: 15 October 2021	The effect of different combinations of botanical spices such as clove and nutmeg in different proportion on the antioxidants activities which include lipid peroxidation (LPO), superoxide dismutase (SOD), catalase (CAT), glutathione-S-transferase (GST) and glutathione peroxide (GPX) in juveniles and adults sizes of <i>Clarias gariepinus</i> was investigated using different combinations of place (G) and putmes (G N 0.0. Control 0%).
Keywords: Spices Nutmeg Clove Anaesthetics Fish Antioxidants	of Clove and Nutmeg; C.N 1:3- 25% Clove and 75% Nutmeg; C.N 3:1- 75% Clove and 25% Nutmeg; C.N 2:2- 50% Clove and 50% Nutmeg; C.4 - 100% Clove; N4- 100%) in triplicates. The results from the study indicated that the anaesthetic caused a substantial ( $p<0.05$ ) modifications in the five antioxidants under examination. The highest deviations in the studied antioxidants were observed in the fish exposed to C4 combination of the anaesthetics and the lowest in the control. The results from this work therefore suggest that the anaesthetics can alter antioxidants levels in the fish which was more noticeable in the fish exposed to C.N 3:1- 75% Clove and 25% Nutmeg; C.N 2:2- 50% Clove and 50% Nutmeg; C.4 - 100% Clove; N4- 100%. Hence fish farmers and scientists are advised to take caution when combining these plant extracts for use in aquaculture

#### 1. Introduction

Stress is often associated with disease outbreak in cultured fish <sup>[1]</sup>. Stress in aquatic organisms such as fish can trigger substantial losses of both animals and capital in both capture and culture systems <sup>[2-4]</sup>. In reducing these losses to the barest minimum necessitate information about the nature of the stressors, stress intensity and

fitness effect <sup>[5]</sup>. In the culture medium, the welfare of cultured fish is often compromised by stress and has become an increasing concern in most of the aquaculture facilities <sup>[6-8]</sup>. Furthermore, the influence of a variety of different segment of aquaculture operations such as transportation, handling and netting, confinement and short-term crowding, inappropriate stocking densities, water quality deterioration on fish welfare and the

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consequent effects on to fish welfare must be minimized, so as to enhance productivity<sup>[9]</sup>.

One of the predominant and common types of stress in a living organism is oxidative stress. Oxidative stress can be described as a disruption of the pro-oxidantantioxidant equilibrium in favour of the former, resulting in possible impairment of cells <sup>[10]</sup>. It occurs as a result of an elevation in Reactive Oxygen Species (ROS) and destruction of antioxidant defence systems or incapacity to repair oxidative damage. The main damage induced by ROS results in alterations of cellular macromolecules such as membrane lipids (lipid peroxidation), DNA [11]. The resulting damage may alter cell task, eventually leading to cell death. Enzymatic (CAT) and non-enzymatic antioxidants such as reduced glutathione (GSH) and its precursor normally counteract damaging effects of ROS by either repairing the oxidative damage or precisely foraging oxygen radicals. Catalase is a common enzyme found in almost all the organisms which are exposed to oxygen, where it functions to catalyze the breakdown of hydrogen peroxide to oxygen and hydrogen oxide. Glutathione (GSH) is an antioxidant which helps to protect the cells from the ROS such as free radicals and peroxides. The glutathione defense enzyme systems in living cell detoxifies and eradicates the xenobiotics leading to the arrangement of products that can easily dissolve in water and their quick removal from the cell of the organism<sup>[12,13]</sup>.

Anaesthesia is a biological condition with the partial or total loss of feeling or loss of voluntary neuromotor control induced by chemical or nonchemical means [14]. Anaesthesia minimizes pain in fish and induces a calming effect followed by loss of equilibrium, mobility and consciousness <sup>[15,16]</sup>. Anaesthetics are chemical substances used by fish farmers to minimize the rate of mortality during handling and transport. This may also reduce the susceptibility of cultured fish to pathogens and infection <sup>[17]</sup>. Anaesthetics are also used in fish farming during artificial spawning, weighing, tagging, grading, blood sampling, surgery and surgical process <sup>[18]</sup>. When choosing ananaesthetics, a number of factors are put into consideration which include efficacy, cost, availability and ease of use, as well as toxicity to fish, humans and the environment <sup>[19]</sup>, and the choice may also depend on the nature of the experiment and species of fish under consideration<sup>[20,21]</sup>.

In application of anaesthetics during aquaculture operations, information about the appropriate and most favourable concentration of an anaesthetic for a range of fish species is essential because improper concentrations might lead to harmful effects such as stress <sup>[22-24]</sup>. Hence,

access to nontoxic and efficient fish sedatives is a crucial necessity of fisheries researchers, manager, and culturists <sup>[25]</sup>. Recently, nutmeg (*Myristica fragrans*) and clove bud (*Syzigium aromaticum*)h ave received favorable reviews as an alternative fish anesthetic for a number of fish species as well as for crustaceans <sup>[26,27]</sup>. Nutmeg has been utilized in different parts of the world for medicinal purposes. Nutmeg (*Myristica fragrans*) is a member of the Myristicaceae family. It is a perennial tree found ecologically in the tropics and well distributed in the north-central region of Nigeria. Extracts from its nuts contains 70-90% myristidate4-21% beta-caryophyllene, 1-21% eugenyl acetate and 10 19% tannin <sup>[28]</sup>.

Despite the several studies that have been done on some physiological changes in *C. gariepinus* exposed to clove and nutmeg <sup>[29,30]</sup>, however there is no available literature on the combined effects of clove buds and nutmeg on the these variables. This study was conducted out to assess the effect of these plant materials as anaesthetics on the selected antioxidants in the plasma of juvenile and adult sizes of *C. gariepinus*. This study will shed more light on stress management procedures in fish farming, using combination of two plants extract as anaesthetics. The aims of the present study therefore, are to evaluate the effects of anaesthesia with mixture of clove and nutmeg buds on the antioxidants in the plasma of juvenile and adult of sizes of *C. gariepinus*.

#### 2. Materials and Methods

#### Sources and acclimation of Experimental Fish

A total of 180 specimens of *Clarias gariepinus* comprising of 90 each of juveniles (mean length17.78 cm±2.88 SD mean weight 106.99g± 4.78SD), and adults (mean length 29.33 cm±3.01 SD and mean weight 654.43g±11.89SD) were obtained from African Regional Aquaculture Centre, (ARAC), Aluu, Rivers State of Nigeria. They were transported in four 50 L jerry cans to the Rivers State University Fish farm and acclimated for a period of seven days. During this time the fish were fed with a commercial feed (45.0% CP) at 3% body weight. The water in acclimation tanks was changed every two days <sup>[26]</sup>.

#### **Preparation of Clove bud and nutmeg**

Dry nutmeg seeds (Plate 1) and dried buds of clove plant (Plate 2) were purchased from Choba Market in ObioAkpor Local Government Area of Rivers State, Nigeria. Plant verification was done by means of the keys described by Agbaje<sup>[31]</sup>. The seeds were taken to the Fisheries laboratory and milled into power using a kitchen blender (Model H2, Ken wood, Japan). The pulverized seeds were afterwards sieved using 0.1 micro nylon meshes to get the fine particles of the spices.



Plate 1. Nutmeg seeds



Plate 2. Clove seeds

#### **Experimental Design and procedures**

The design of the experiment is a Randomized Complete Block Design (RCBD) having six treatments levels each with three replicates for each of the life stages. A total of 36 plastic aquaria of dimension  $(52 \times 44 \times 34 \text{ cm}^3)$  each were used for the experiments. The 36 aquaria were labelled based on the experimental units and replicates. Each aquarium was stocked with five fish. The powder was combined based on ratio of Clove : nutmeg into different proportion of C.N 0:0- Control 0% of Clove and Nutmeg; C.N 1:3- 25% Clove and 75% Nutmeg; C.N 3:1- 75% Clove and 25% Nutmeg; C.N 2:2-50% Clove and 50% Nutmeg; C.4 - 100% Clove; N4- 100% Nutmeg. This was achieved by using weighing balance. It was applied directly in three replicates into the water (10 L) in 30 L experimental plastic aquaria. The mixtures were stirred extensively to ensure homogenous mixture. They were then introduced into prepared experimental aquaria, containing 5 concentrations of powdered nutmeg and clove at the rate of ten fish per tank in triplicates. The fish was then introduced into the tank. When the fish has attained stage five anaesthesia (the point when the fish has lost sensitivity to gentle prodding with rod), blood samples were collected from the caudal vein with syringe and blood samples were preserved in heparinized bottle for analyses.

#### **Evaluation of Water Quality Parameters**

Water quality variables such as: dissolved oxygen,

nitrite, ammonia and sulphide were evaluated using LaMotte fresh water test kit (Model AQ4, Chestown, Maryland, USA). pH was determined with pH meter (Model, H 9812, Hannah Products, Portugal). The dissolved oxygen level was evaluated by the Winkler method<sup>[32]</sup>.

#### **Sample Preparation**

At the end of each experimental period, 2 ml of fresh blood sample was collected by making a caudal puncture with the help of fine needle and transferred into heparinized sample bottles. Blood samples were centrifuged immediately for 15 minutes at 5000 rpm. Plasma specimens were separated, pipetted into eppendorf tubes and stored in a refrigerator at -20°C until assayed. The results were read using a universal microplate reader on a Jenwayvisible spectrophotometer (Model 6405). The activity of antioxidants in centrifuged plasma was determined spectrophotometrically using the method of Eales<sup>[33].</sup>

#### **Statistical Analysis**

All the data were expressed as mean and standard deviation of mean. The statistical package, SPSS Version 22 was used for the data analysis. The means were separated using two-ways ANOVA and the two means were considered significant at 5% (P<0.05).

#### 3. Results

The physico-chemical indices of water in the experimental tanks of C.gariepinus exposed to combined nutmeg and clove seed powder is shown in Table 1. The results indicated significant differences (p<0.05) only among the dissolved oxygen and ammonia in the control and treated group range. The values of the other variables were not significantly different (P>0.05). Difference in the values of antioxidants in the juveniles of C.gariepinus juveniles exposed to different combination of nutmeg and clove seed powder were presented in Table 2. There was significant reductions (P<0.05) comparable to the control values in GSH, SOD and GPX, whereas the values of CAT and LPO increased. Moreover, the same trends were also recorded in adult size of C.gariepinus exposed to these plant powder (Table 3). Comparative values of GPX in juveniles and adults of C.gariepinus exposed to different combinations of nutmeg and clove seed extracts are presented in Figure 1. The highest value of 50.00 was recorded in the adult fish exposed to the control, while the lowest was in C.N 3:1. However, GPX values in the adult fish were consistently higher than that of the juveniles (Figure 1).

	Combinations of Clove and Nutmeg in Different Proportion					
Parameters	C.N 0:0	C.N 1:3	C.N 3:1	C.N 2:2	C 4	N 4
DO (mg/l)	6.33±0.30 <sup>d</sup>	5.33±0.57 °	4.00±0.00 <sup>b</sup>	4.33±0.57 <sup>b</sup>	3.66±0.57 °	5.00±0.00°
NH3 (mg/l)	0.02±0.01 <sup>a</sup>	0.05±0.00 <sup>b</sup>	0.04±0.01 <sup>b</sup>	0.03±0.00 <sup>a</sup>	0.05±0.00 <sup>a</sup>	$0.07{\pm}0.00^{a}$
Tem 0C	29.66±1.15 <sup>a</sup>	30.00±1.00 <sup>a</sup>	29.33±0.57 ª	29.33±0.57 <sup>a</sup>	29.33±0.57 <sup>a</sup>	29.33±0.57ª
рН	5.33±0.57 <sup>a</sup>	5.33±0.57 <sup>a</sup>	6.00±0.00 <sup>a</sup>	5.33±0.57 <sup>a</sup>	5.50±0.50 ª	5.33±0.57 <sup>a</sup>
Condt (S/m)	115.0±1.0 <sup>a</sup>	123.00±1.0 <sup>a</sup>	128.66±1.52 <sup>a</sup>	126.66±1.52ª	131.33±1.15 <sup>b</sup>	119.00±1.00 <sup>a</sup>
Nitrite (mg/l)	0.01±0.00 <sup>a</sup>	0.01±0.00 <sup>a</sup>	0.01±0.00 <sup>a</sup>	0.01±0.00 <sup>a</sup>	0.01±0.00 <sup>a</sup>	0.01±0.00 <sup>a</sup>
Nitrate (mg/l)	0.13±0.01 ª	0.15±0.02 <sup>a</sup>	0.13±0.02 <sup>a</sup>	0.13±0.02 <sup>a</sup>	0.12±0.01 <sup>a</sup>	0.14±0.01 <sup>a</sup>

 Table 1. Physio-Chemical Parameters of Water in the Experimental Tanks of C.gariepinus Exposed to Combined

 Nutmeg and Clove bud Powder.

Means within the same row with different super scripts are significantly different (P<0.05)

Key: C.N 0:0- (Control) 0% of Clove and Nutmeg; C.N 1:3- 25% Clove and 75% Nutmeg; C.N 3:1- 75% Clove and 25% Nutmeg; C.N 2:2- 50% Clove and 50% Nutmeg; C.4 - 100% Clove; N4- 100% Nutmeg.

 Table 2. Antioxidants Levels in the blood of C.gariepinus Juveniles Exposed to Different Combination of Nutmeg and Clove bud Powder.

Concentration	GPX	CAT	GSH	SOD	LPO
C:N 0:0	39.66±1.52 <sup>b</sup>	62.00±2.00 <sup>ª</sup>	5.33±0.57°	10.33±0.57 <sup>b</sup>	7.33±0.57 <sup>a</sup>
C:N 1:3	38.08±3.46 <sup>b</sup>	65.66±1.15 <sup>a</sup>	4.66±0.57°	9.33±0.57 <sup>a</sup>	8.66±0.57 <sup>a</sup>
C:N 2:2	30.66±1.15 <sup>b</sup>	74.33±1.15 <sup>b</sup>	2.33±0.57 <sup>b</sup>	6.00±1.00 <sup>a</sup>	12.00±1.00 <sup>b</sup>
C:N 3:1	26.66±2,08 <sup>a</sup>	76.00±1.00 <sup>b</sup>	3,33±0.57 <sup>b</sup>	6.00±0.00 <sup>a</sup>	13.00±1.00 <sup>b</sup>
C 4	28.33±1.52 <sup>ª</sup>	79.00±1.00 <sup>b</sup>	1.66±0.57 <sup>a</sup>	5.66±0.57 <sup>a</sup>	14.66±0.57 <sup>b</sup>
N 4	35.33±5.89 <sup>b</sup>	70.00±1.00 <sup>b</sup>	3.33±0.57 <sup>b</sup>	6.66±0.57 <sup>a</sup>	9.66±0.57 <sup>a</sup>

Means within the same column with different super scripts are significantly different (P<0.05)

Key: C.N 0:0- (Control) 0% of Clove and Nutmeg; C.N 1:3- 25% Clove and 75% Nutmeg; C.N 3:1- 75% Clove and 25%:2- 50% Clove Nutmeg; C.N 2and 50% Nutmeg; C.4 - 100% Clove; N4- 100% Nutmeg.

 Table 3. Antioxidants Levels in the blood of C.gariepinus Adults Exposed to Different Combination of Nutmeg and Clove budPowder.

Concentration	GPX	CAT	GSH	SOD	LPO
C:N 0:0	50.00±2.00 <sup>a</sup>	82.00±2.00 <sup>a</sup>	7.66±0.57 <sup>a</sup>	15.33±0.57 <sup>a</sup>	12.66±1.15 <sup>a</sup>
C:N 1:3	48.33±3.78 <sup>a</sup>	86.00±1.00 <sup>a</sup>	7.33±1.15 <sup>a</sup>	14.33±0.44 <sup>a</sup>	13.66±0.57 <sup>a</sup>
C:N 2:2	41.00±1.00 <sup>a</sup>	95.00±1.00 <sup>a</sup>	5.00±1.00 <sup>a</sup>	11.33±0.57 <sup>a</sup>	17.00±1.00 <sup>a</sup>
C:N 3:1	37.66±2.08 <sup>a</sup>	92.66±4.93 <sup>a</sup>	5.33±9.57 <sup>a</sup>	11.33±0.51 <sup>a</sup>	18.66±1.52 <sup>a</sup>
C 4	38.66±2.08 <sup>a</sup>	99.00±1.00 <sup>a</sup>	5.00±0.00 <sup>a</sup>	10.33±0.52 <sup>a</sup>	19.66±0.57 <sup>a</sup>
N 4	46.00±1.00 <sup>a</sup>	90.66±1.15 <sup>a</sup>	5.66±0.57 <sup>a</sup>	11.33±0.09 <sup>a</sup>	14.33±1.54 °

Means within the same row with different super scripts are significantly different (P<0.05)

Key: C.N 0:0- (Control) 0% of Clove and Nutmeg; C.N 1:3- 25% Clove and 75% Nutmeg; C.N 3:1- 75% Clove and 25% Nutmeg; C.N 2:2- 50% Clove and 50% Nutmeg; C.4 - 100% Clove; N4- 100% Nutmeg.

The activity of Glutathione perioxide in both juveniles and adult declined below their control values under the exposure to the anaesthetics (Figure 2). The highest value of CAT (99.0 IU/L) was recorded in C4 (100 % Clove), while the lowest (62.0 IU/L) was recorded in juvenile fish in the control (Figure 2). Despite this, the values of CAT were higher in the adult fish when put side by side to the juveniles in all the exposure combinations. Furthermore, the similar inclinations were also recorded in the values of GSH (Figure 3), with the values in adult fish higher than that of the juveniles in all combinations of clove and nutmeg extracts. The values of SOD and LPO in both sizes of *C.gariepinus* exposed to these plant extracts are presented in Figures 4 and 5 respectively. However, these values were higher in adult fish when compared to the juveniles.



Figure 1. Effect of different combinations of Nutmeg seed and Clovebuds powder on Glutathione peroxide (GPX) activities in the sera of juvenile and adult *C. gariepinus* 



Figure 2. Effect of different combination of nutmeg and clove bud powder of Catalase (CAT) activities in the blood of juvenile and adult *C. gariepinus* 



Figure 3. Effect of different combination of Nutmeg and Clove bud powder of Glutathione GSH activities in the plasma of juvenile and adult of *C. gariepinus* 



Figure 4. Effects of different combination of nutmeg and clove bud powder on SOD activities in the plasma of juvenile and adult of *C. gariepinus* 



Figure 5. Effects of different combination of Nutmeg and Clove bud powder on Lipid peroxidation LPO activities in the plasma of juvenile and adult of *C. gariepinus* 

#### 4. Discussion

Oxidative stress is a condition when stable position ROS concentration is transiently or chronically enhanced, distressing cellular metabolism and its regulation and damaging cellular constituents <sup>[34]</sup>. It can also be described as a condition when antioxidant defences are overwhelmed by pro-oxidant forces <sup>[35]</sup>. The commencement of oxidative manifestations leads to the response of antioxidants activation in articulation of genes encoding antioxidant enzymes. Nevertheless, there are substantial disparities in the knowledge on response to oxidative stress, specifically in aquatic animals. Antioxidant enzymes are included in stress evaluation because of their indelibility under conditions of mild oxidative stress and their potential role in adaptation to aquaculture -induced stress. It is expected that they may be more responsive at revealing preliminary stress induced insult on the cells <sup>[36]</sup>. Laboratory studies established that the dimension of variations in the manifestation of a large number of specific genes or performances of certain enzymes of antioxidant defence can be investigated in an early warning system of toxicant exposure<sup>[34]</sup>. Apparently, the early caution can be used at what time temporal effect of stress is expected. Typically, the range of oxidative stress indices in fish includes the activities of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPX), lipid peroxidase (LPO) and glutathione s transferase (GST). The evaluation of these has most frequently been utilized in eco-toxicology programmes for fish<sup>[37]</sup>.

In this work, increased values of CAT and LPO activities in combination with the decrease in the values of SOD, GSH and GPX were recorded in the plasma of C.gariepinus sampled in both juveniles and adult sizes of C.gariepinus exposed to different combinations of nutmeg and clove seed extracts. A similar result was observed in C. carpio exposed to higher concentrations of clove oil <sup>[38]</sup>. Moreover, the concerted elevation of LPO and CAT activities was equally indicated in the plasma of starlet fish (Acipenser ruthenus) exposed to 2-phenoxy- ethanol as anaesthetics <sup>[39]</sup>. In the study of three populations of brown trout (S. trutta) treated with elevated levels of anaesthetic MS222, all the exposed fish recorded higher activities of CAT in their plasma compared to unexposed trout <sup>[40]</sup> (Hansen et al., 2006). Conversely, imbalanced antioxidant activities were observed in the various oxidative stress biomarkers in the Indian freshwater fish,

Wallagoattu exposed to anaesthetics metomidate [41].

The increase recorded in the values of catalase activity or its steadiness alongside with reduction in SOD and GST activities have been reported by some authors <sup>[39,40]</sup>. In spite of Lowry et al. <sup>[42]</sup> observed that in plasma, hepatic and adrenal tissues of white sucker (C. commersoni) exposed to some anaesthetics from some farms in Agricultural region in Québec (Canada) had their catalase activities higher than those fish from the control. On the other hand, the same trend was observed in this study. Nevertheless, Falfushynska and Stolvar<sup>[43]</sup>. ascribed the soaring catalase activity in fish exposed to a quantity of extracts, to little production of oxygen, which has been reported to boost the making of catalase as cell stability in the case of surplus infusion of pollutants <sup>[44]</sup>. On the contrary Catalase decline and activation can be considered as a last resort of antioxidant defence in teleost fish. The catalase role in the antioxidant defence of fish was reported by Porte [45], based on the information on its activation by hydrogen peroxide at high concentrations. They suggested that catalase on the whole plays a comparatively inconsequential role in hydrogen peroxide catabolism at low rates of peroxide generation, but it becomes crucial when the rate of hydrogen peroxide production is enhanced, for example, at oxidative stress.

GSH is a foremost cytosolic low molecular weight sulfhydyl compound that functions as cellular reducing and protective reagent against a wide range of contaminant through SH-group. It directly acts a scavenger of oxyradical and also as an antioxidant enzyme base. Apparently, GSH is essential in protecting against deleterious effects of the cell exposed to ROS by reacting with them to form glutathione disulphide (GSSG). This antioxidant defence effect occurs spontaneously through GSH or by GST. It acts as cofactor for glutathione transferase, which make it easy for the elimination of some chemicals and erstwhile reactive molecules from the cells [46]. Thus a change in GSH levels may be a vital indicator of detoxification capacity of an organism. During present investigation, significant decrease in GSH level observed in the plasma of both sizes of C.gariepinus at different exposures could be due to its utilization to confront the current oxidative stress under the influence of ROS produced from anaesthetics exposure. Reduced GSH and its metabolizing enzyme make available the principal resistance in opposition to ROS induced cellular destruction <sup>[47]</sup>. This reduction may be because of increased utilization of GSH, which can be transformed to oxidized glutathione and potentially fragile GSH regeneration. Furthermore, these authors affirmed that GSH depletion point to its exhaustive phase II biotransformation; by this means boost the risk of oxidative stress due to reduced cell protection activity <sup>[48]</sup>. Similar decrease in the plasma of *Cyprinus carpio* have also been reported on exposure to clove at higher concentrations <sup>[49]</sup>. Present observations are in agreement with the findings of Avilez *et al.* <sup>[15]</sup> who studied effect of propiconazole on *Oncorhynchus mykiss*, these researchers reported a decrease in GSH level in the plasma of the fish and pointed it to be a primary protective response of the cell against oxidative stress induced by pollutants.

GPX is a group of multifunctional isoenzymes, which play an important role in detoxification of toxic electrophiles by catalyzing the conjugation of a wide variety of electrophilic substrates to GSH and thus protects the cell from oxidative stress. It is considered as first line of defence against oxidative stress injury, decomposing superoxide radicals and hydrogen peroxide before interacting to form the reactive hydroxyl, which has a number of adverse biological effects when present in high amounts <sup>[50, 51]</sup>. The reduced GPX activity is concomitant to the decrease in GSH level in the plasma liver and gills. From the above discussion, it could be inferred that GPX utilizes GSH for the xenobiotic detoxification.

#### 5. Conclusions and Recommendations

The parameters measured provided useful information for evaluating the toxicological effects of different combinations of nutmeg and clove buds in *C.gariepinus*. From the results of this study, it was revealed that the combination of C.N at ratio 1:3 which consists of 25% Clove and 75% Nutmeg is suitable for use in aquaculture, because of its minimal alterations in the antioxidant activities in *C.gariepinus* when compared to other concentrations. From this study it is obvious that this combination can be effectively applied to anaesthesized fish during aquaculture operations.

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## ARTICLE Consequences of Direct Transfer to Fresh Water on the Blood Variables of Sarotherodon melanotheron (Rüppell, 1852)

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ARTICLE INFO	ABSTRACT
Article history Received: 23 September 2021 Accepted: 8 October 2021 Published Online: 15 October 2021	Alterations in blood variables of <i>Sarotherodon melanotheron</i> transmitted immediately from brackish water (salinity 13.71%) to fresh water (salinity 0.12%) were examined to evaluate the consequence of abrupt alterations in the environment on fish blood parameters. The results acquired revealed that significant reduction ( $p < 0.05$ ) in mean values of Haemoglobin (Hb) from 7.37+1.22 to 4.30+0.82dl <sup>-1</sup> . Packed Cell Volume (PCV) 23.48+3.22
Keywords: Haematology Sarotherodon melanotheron Salinity Fresh water Brackish water	to 15.08±2.97%; Red Blood Cell (RBC) 5.99±0.72 to 3.31±0.77 Cells x10 <sup>6</sup> ; Mean Corpuscular Haemoglobin Concentration (MCHC) 31.68±3.66 to 28.48±2.03 pg; and Platelets (PLT) 195.44±16.77 to 138.99±9.05 $10^3$ uL <sup>-1</sup> . However, notable elevation was recorded in the values of Erythrocyte Sedimentation Rate (ESR) from 4.89±0.16 to 10.58±1.48 mm/ hr; White Blood Cell (WBC) 27.03±2.96 to 33.09±3.72 x 10 <sup>9</sup> L <sup>-1</sup> ; Mean Corpuscular Volume (MCV) 39.33±3.07 to 45.88±7.03 fL Neutrophils (NEUT) 41.71±3.08 to 48.30±7.99%; Lymphocytes (LYMP) 54.60±3.99 to 46.36±9.87% and Monocytes (MON) 3.80±1.04 to 5.97±1.99%. These variations in the blood parameters were more perceptible in adult than juvenile fish. Results from this study consequently imply that direct transfer of fish to a region of lower salinity may have damaging effect on the physiology of S. <i>melanotheron</i> as observed in this study.

#### 1. Introduction

The specie, *Sarotherodon melanotheron* is considered widely distributed in the coastal areas of Niger Delta region of Nigeria, <sup>[1]</sup>. Adaptation to changes in environmental factors has been identified as one of the major area of research necessary for *S.melanotheron* culture development <sup>[2]</sup>. In the wild, *S. melanotheron* are estuarine, found mostly in the coastal parts of the country. There is the need therefore to assess the possibility of culturing this specie in fresh water. Lemarie *et al.* <sup>[3]</sup> recounted that the ability of tilapia

to inhabit diverse environment with irregular salinities depends on the species, mean individual weight, methods of transfer, feeding techniques for pre-acclimation, the physiological status of the fish and more commonly the consequence of environmental features.

Consistent changes in composition of aquatic environment have been observed to alter the behaviour, and physiology of fish <sup>[4]</sup>. In aquaculture, the performance of cultured fish species is controlled by not only genetic potential and technological manipulation but also by its immediate environmental conditions <sup>[5]</sup>. Sudden change in the

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environmental condition of fish, reportedly cause serious stress, which will eventually disrupt the haematological parameters of the fish. The disrupted blood components involved in maintaining or expanding the capacity of water, ion transport, and homeostasis are now limited in performing their functions, thus leading to immuno depression, low productivity disease outbreaks and ultimately mortality <sup>[6]</sup>. Hence, proper knowledge of haematological response in fish to changes in environment will help in improving production of fish and control of stress in aquaculture. This work therefore examines the effect of direct transfer from brackish to fresh water on the blood indices of *S.melanotheron* a popular fish for culture among local farmers in the Niger Delta regions of Nigeria.

#### 2. Materials and Methods

#### **Experimental Fish and Procedure**

One hundred and twenty S.melanotheron comprising of 60 juvenile sizes (mean length 9.00 cm±2.92SD; weight  $37.61g\pm 6.02SD$ ) and 60 adult sizes (mean length 14.88  $cm\pm 5.99SD$ ; mean weight 120.87 g $\pm 12.82SD$ ) were collected from brackish water in Buguma creek at low tide. They were immediately conveyed to the outdoor hatchery where the initial blood samples were drawn from the fish and taken to laboratory for analysis. They were later kept at 20 each in three rectangular tanks  $(0.36 \text{ m}^3)$ filled to half capacity with fresh water for interlude of seven days for each experimental fish sizes. The fish was fed twice daily with crumblized pelleted feed (35% c.p) at 2% body weight. The mortality during the experimental period was ascertained by counting the number of the dead fish in each experimental tank for each size from the first day to the seventh day.

#### **Evaluation of Physico-Chemical Parameters**

Physico-chemical parameters namely temperature pH, ammonia nitrogen, nitrate, dissolved oxygen, sulfide and salinity in the creek (brackish water) and in the experimental tanks (fresh water) were monitored. Temperature was taken with mercury in glass thermometer; pH was taken with pH meter while Ammonia, nitrate, sulfide and dissolved oxygen were determined with Horiba U-7 water checker. And salinity was measured with hand held refractometer (Model HRN-2N Atago Products, Japan).

#### **Blood Sampling and Analysis**

Blood samples were collected from a total of 40 fish that is 20 each before and after transfer, comprising of 10

fish each for both juvenile and adult sizes. Blood samples were obtained with heparinized plastic syringe fitted with 21 gauge hypodermic needle and preserved in disodium salt of Ethylene Diamine Tetraacetic Acid (EDTA) bottles for analysis. The blood samples collected at the hatchery were labeled initial and those collected after seven days of transfer were labeled final samples. The values of Packed Cell Volume (PCV) were determined using microhaematocrit method of Snieszko<sup>[7]</sup>. Haemoglobin (Hb) was done by cvanomethaemoglibin method<sup>[8]</sup>. The Red Blood Cell (RBC) were enumerated in an improved Neubaeur haemocytometer using Handricks diluting fluid, while the total White Blood Cell Counts (WBC) were similarly enumerated in an improved Neubaeur haemocytometer using shaw's diluting fluid <sup>[9]</sup>. The Erythrocyte Sedimentation Rate (ESR) was done by Wintrobe method. Also, Platelet (PLT) was done with Rees and Ecker Method<sup>[10]</sup>, while the Red Blood Cell indices; Mean Corpuscular Haemoglobin Concentration (MCHC), Mean Corpuscular Haemoglobin (MCH) and Mean Corpscular Volume were calculated from the equation given by Anderson and Klontz<sup>[11]</sup>. The differential counts were done on blood film stained with may Grumwald-Giensa stain as described in Hrubec et al., [12].

#### **Data Analysis**

All data obtained were subjected to Analysis of variance (ANOVA) at 0.05% probability and differences among means were separated with the significant difference using SAS software <sup>[13]</sup>.

#### 3. Results

The physico-chemical parameters of water in the creek (before transfer) and the experimental tanks (after transfer) was significant (p<0.05) only at salinity (Table 1). After transfer the highest mortality  $(50.00\pm8.03)$ was recorded in day 1 for adult fish while the lowest (3.00±1.01) was observed in juvenile fish in day 7 (Table 2). The initial and final haematological parameters of juvenile and adult size of S.melanotheron transfer to fresh water environment were shown in Table 3 and 4. the results indicated a consistent reduction in the values of Hb, PCV, RBC, MCHC, MCH and PLT, while the values of ESR, WBC, MCV, Neutrophils, lymphocytes, and Monocytes increased significantly (p<0.05) which was more pronounced in adult fish than the juvenile (Table 3 and 4). The pooled data for the variations in the blood variable of the transferred fish regardless of the size was presented in Table 5, the lowest value was observed in RBC, while PLT had the highest value.

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Parameters	Before Transfer (Brackish water)	After Transfer (Fresh Water)		
pН	6.69±0.88 <sup>a</sup>	6.89±0.41ª		
Temperature ( <sup>0</sup> c)	28.97±1.80 <sup>a</sup>	28.77±1.74 <sup>a</sup>		
Ammonia (mgL <sup>-1</sup> )	$0.41{\pm}0.01^{a}$	$0.61{\pm}0.34^{a}$		
Nitrate (mgL <sup>-1</sup> )	0.0023±0.02ª	$0.0042{\pm}0.01^{a}$		
Dissolve oxygen (mgL <sup>-1</sup> )	4.45±0.89 <sup>a</sup>	$4.01{\pm}0.88^{a}$		
Sulfide (mgL <sup>-1</sup> )	$0.04{\pm}0.01^{a}$	$0.02{\pm}0.01^{a}$		
Salinity (‰)	14.22±2.02 <sup>a</sup>	0.02±0.01 <sup>b</sup>		

 
 Table 1. Physico-Chemical Parameters of Water before and after Trial

\*Means with the same superscript within the row under before and after trial are not significantly different (p>0.05)

## Table 2. Mortality (Mean $\pm$ SD) Observed in S.melanotheron Transfer to Fresh Water for a Period of<br/>Seven Days

Dava	Fish	Sizes
Days	Juvenile	Adult
1	26.00±5.99 <sup>a</sup>	50.00±8.03 <sup>b</sup>
2	$18.00 \pm 4.88^{a}$	$34.00{\pm}7.90^{b}$
3	$14.00{\pm}2.07^{a}$	$28.00 \pm 4.77^{b}$
4	$10.00{\pm}1.82^{a}$	23.00±3.71 <sup>b</sup>
5	$8.00{\pm}2.02^{a}$	19.00±3.53 <sup>b</sup>
6	5.00±1.03 <sup>a</sup>	14.00±4.99 <sup>b</sup>
7	3.00±1.01 <sup>a</sup>	$10.00 \pm 3.08^{b}$

\*Means with the same superscript within the row under before and after trial are not significantly different (p>0.05)

 Table 3. Haematological (Mean±SD) in Juvenile size of S.

 melanotheron before and after transfer

	Before Transfer	After Transfer
<b>Blood Parameters</b>	(Brackish water)	(Fresh water)
Hb (gld1)	6.77±0.92 <sup>a</sup>	4.03±0.92 <sup>b</sup>
PCV (%V)	20.02±3.77 <sup>a</sup>	15.09±3.99 <sup>b</sup>
ESR (mm/hr)	2.90±0.19 <sup>a</sup>	$8^{\pm}09{\pm}1.19^{b}$
RBC (x10 <sup>6</sup> /mL)	4.99±0.66 <sup>a</sup>	$3.59{\pm}0.92^{b}$
WBC (x10 <sup>9</sup> /L)	24.99±7.90 <sup>a</sup>	29.20±4.88 <sup>b</sup>
MCHC (%)	33.81±3.88 <sup>a</sup>	26.70±2.72 <sup>b</sup>
MCH (g/dl)	13.57±2.02 <sup>a</sup>	11.23±2.90 <sup>b</sup>
MCV/(fl)	40.12±4.77 <sup>a</sup>	$42.03 \pm 4.88^{b}$
PLT/(10 <sup>8</sup> /mL)	191.79±9.99 <sup>a</sup>	148.88±9.98 <sup>b</sup>
Neutrophils (%)	42.79±3.99ª	47.88±3.21 <sup>b</sup>
Lymphocytes (%)	52.85±2.92ª	46.38±13.12 <sup>b</sup>
Monocytes (%)	4.56±1.06 <sup>a</sup>	6.99±0.82 <sup>a</sup>

Mean within the row with different superscript are significant (P < 0.05)

Key: Hb – Haemoglobin; PCV – Packed Cell Volume; ESR – Erythrocyte Sedimentation Rate; RBC – Red Blood Cell; WBC – White Blood Cell; MCHC – Mean Corpuscular Haemoglobin Concentration; Mean Corpuscular Haemoglibin PLT – Platelets. Table 4. Haematological in Adult size of S. melanotheronbefore and after transfer

Blood Parameters	Before Transfer	After Transfer
	(Brackish water) Mean ± SD	(Fresh water) Mean ± SD
Hb (gld1)	7.96±0.94 <sup>a</sup>	4.56±0.99 <sup>b</sup>
PCV (%V)	26.94±1.09 <sup>a</sup>	15.07±6.66 <sup>b</sup>
ESR (mm/hr)	$6.88 \pm 0.88^{a}$	13.07±2.89 <sup>b</sup>
RBC (x10 <sup>6</sup> /mL)	6.99±0.83ª	$3.03{\pm}0.03^{b}$
WBC (x10 <sup>9</sup> /L)	29.02±3.77 <sup>a</sup>	36.99±3.88 <sup>b</sup>
MCHC (%)	29.54±3.07 <sup>a</sup>	30.26±3.99 <sup>b</sup>
MCH (pg)	$11.39{\pm}1.77^{a}$	15.04±3.77 <sup>b</sup>
MCV/(fl)	38.54±5.04ª	49.74±5.88 <sup>b</sup>
PLT(10 <sup>8</sup> /mL)	199.88±9.99ª	$128.88 \pm 6.00^{b}$
Neutrophils (%)	40.62±3.41ª	$48.73 \pm 8.07^{b}$
Lymphocytes (%)	56.35±7.41ª	46.33±5.62 <sup>b</sup>
Monocytes (%)	3.03±0.77 <sup>a</sup>	$4.94{\pm}0.77^{b}$

Mean within the row with different superscript are significant (P < 0.05)

Key: Hb – Haemoglobin; PCV – Packed Cell Volume; ESR – Erythrocyte Sedimentation Rate; RBC – Red Blood Cell; WBC – White Blood Cell; MCHC – Mean Corpuscular Haemoglobin Concentration; Mean Corpuscular Haemoglibin PLT – Platelets.

**Table 5.** Mean Values of Haematological Parameters of S.melanotheron Transfer Directly to Fresh water

<b>Blood Parameters</b>	Before Transfer	After Transfer
	(Brackish water) Mean ± SD	(Fresh water) Mean ± SD
Hb (gld1)	7.37±1.22 <sup>a</sup>	4.30±0.82 <sup>b</sup>
PCV (%V)	23.48±3.22ª	15.08±2.97 <sup>b</sup>
ESR (mm/hr)	4.89±0.16 <sup>a</sup>	$10.58{\pm}1.48^{b}$
RBC (x10 <sup>6</sup> /mL)	5.99±0.72ª	$3.31{\pm}0.77^{b}$
WBC (x10 <sup>9</sup> /L)	27.03±2.96 <sup>b</sup>	33.09±3.72 <sup>b</sup>
MCHC (pg)	31.68±3.66 <sup>b</sup>	28.48±2.03 <sup>b</sup>
MCH (g/dl)	12.48±3.11ª	13.14±4.71ª
MCV(fl)	39.33±3.07 <sup>b</sup>	45.88±7.03ª
PLT(10 <sup>8</sup> /mL)	195.44±16.77 <sup>a</sup>	138.99±9.05 <sup>b</sup>
Neutrophils (%)	41.71±3.08 <sup>b</sup>	48.30±7.99ª
Lymphocytes (%)	54.60±3.99 <sup>b</sup>	46.36±9.87 <sup>b</sup>
Monocytes (%)	$3.80{\pm}1.04^{b}$	5.97±1.99 <sup>a</sup>

Mean within the row with different superscript are significant (P < 0.05)

Key: Hb – Haemoglobin; PCV – Packed Cell Volume; ESR – Erythrocyte Sedimentation Rate; RBC – Red Blood Cell; WBC – White Blood Cell; MCHC – Mean Corpuscular Haemoglobin Concentration; Mean Corpuscular Haemoglibin PLT – Platelets.

#### 4. Discussion

Understanding of the haematological characteristics in aquatic animal is an essential instrument in conjunction with the environmental factors that can deduce the health of fish population. Fish reacts to external stimuli in various ways, depending on the stressor involved, as observed by Martins et al., <sup>[14]</sup> and Gomes et al., <sup>[15]</sup>. According to Akinrotimi et al., <sup>[16]</sup>, the haematological reaction of fish to stress is determined by the disposition of the stress itself, that is definite stress elicit a definite responses. According to Luskova <sup>[17]</sup> and Gbore et al., <sup>[18]</sup> who reported that the importance of the haematological conditions of a fish, as a biological indication of stressful situation was established and a variety of haematological parameters were shown as very sensitive to intoxication with trace metals, micro contaminants, diseases, and changes in the environmental components.

The current trial revealed various degrees of haematological responses of S.melanotheron exposed to direct transfer from brackish to fresh water. The rapid change in the environment stimulates a significant (P<0.03) decrease in the following blood parameters: haemoglobin, packed cell volume, red blood cell, mean corpuscular haemoglobin concentration, mean corpuscular haemoglobin and platelets. The enumeration of old disintegrated erythrocyte was significantly higher in adult than the juvenile fish. Patterns of changes in these parameters found by other authors depended on the salinity level, duration of exposure, mode of transfer and fish species. The decrease in blood parameters as observed in this study agrees with the finding of Gabriel et al., [19] in T.guineeensis where comparable decrease in blood parameters were reported. The results observed in this work may be due to the radical effect of stressor (change in environment) which leads to haemodilution, as a result of damaged erythropoiesis (production of erythrocytes) as a result of interrupted osmoregulation activities in the fish [20]. Moreover, Zhiteneva et al. [21] reported inhibition of erythrocyte production, splitting of the cytoplasm of erythrocytes in tilapia exposed to impulsive adjustment in environment.

Direct transfer of *S.melanotheron* into a new environment also reduces an increase in erythrocyte sedimentation rate, white blood cell, mean corpuscular volume, neutrophils, lymphocytes and monocytes. The erythrocyte sedimentation rate which evaluated the rate at which red cell settle out of their plasma, increased significantly, is an indicative of anaemia, due to fragility of the erythrocytes consequence of stress induced interference in the development and discharge of erythrocytes from haematopoeitic tissue <sup>[22,23]</sup>.

The present experiment established considerable

elevation in the amount of white blood cells (WBC), this is in the same opinion with the findings of Sardella *et al.*, <sup>[24]</sup> who reported a notable increase in WBC in the Mozambique tilapia after exposure to new environment. Also, number of WBC increased tremendously in goldfish (*Carassius auratus*) after exposure of the fish to various categories of stressors <sup>[25]</sup>. An increase in the quantity of WBC may be as a result of liberation of cells amassed in the spleen, when fish is exposed to stressors <sup>[26]</sup>. The differential counts namely monocytes, neutrophils and lymphocytes increased to a large extent as observed in this study, this support the findings of Ginling *et al.*, <sup>[27]</sup> in green back flounder (*Rhombosulea* tapirina) exposed to fresh water. The increased observed may be due to release of more of the white cells to combat the stressor <sup>[28]</sup>.

#### **5.** Conclusions

The present study confirmed that haematological parameters are very sensitive indicators of fish organism response to changes in the environmental factors. The study revealed that an abrupt and unexpected modification in the fish environment can distort fish physiological conditions and affects its performance in the culture medium. Hence, it is essential and important to culture fish in an appropriate environment for its adaptive and functional physiological mechanism. When there is the need to culture *S.melanotheron* in fresh water it should undergo gradual acclimation rather than direct transfer which may lead to decline in production capability of the fish and in some results in mortality.

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