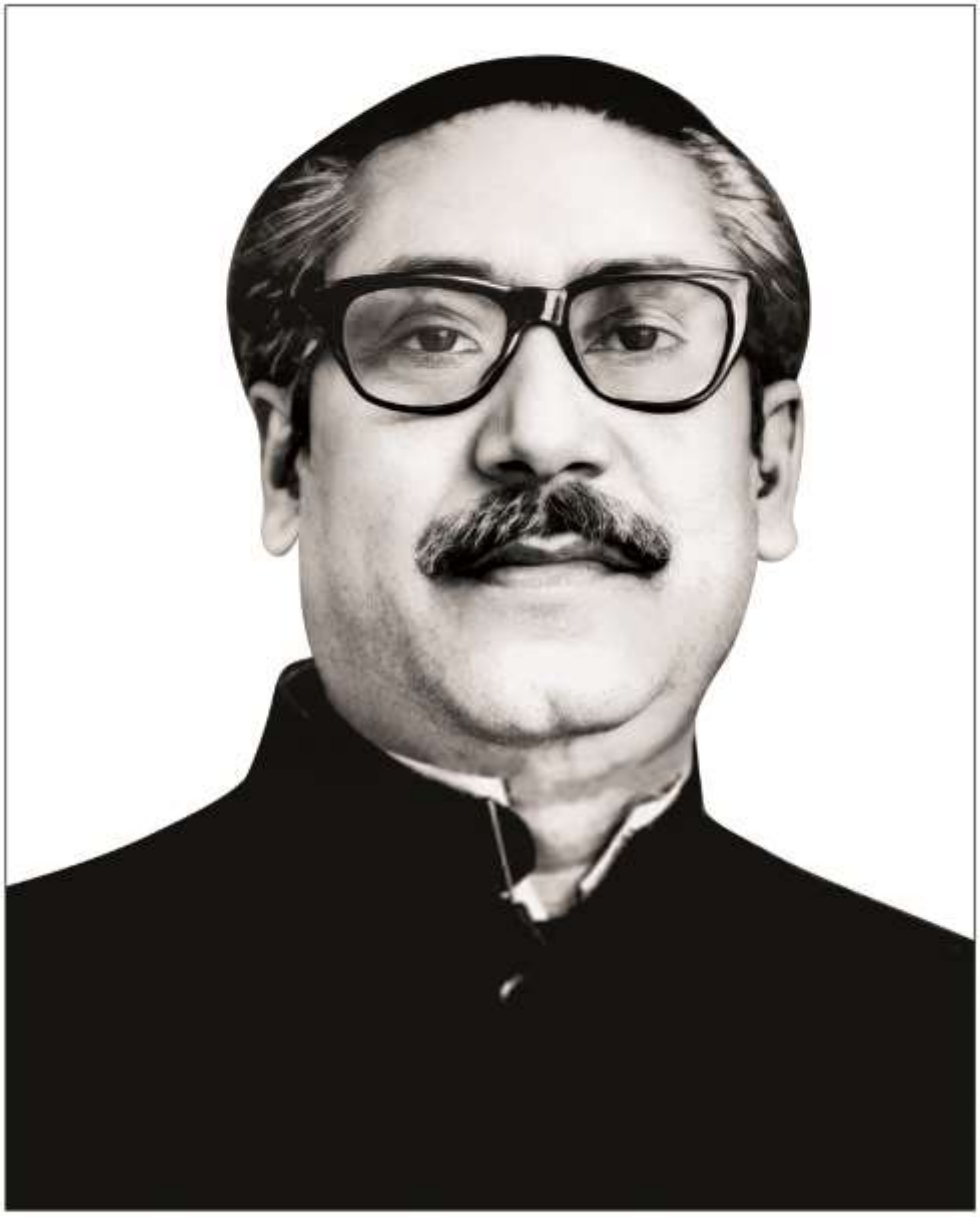


Father of the Nation Bangabandhu Sheikh Mujibur Rahman

Sheikh Hasina
Hon'ble Prime Minister of the People's Republic of Bangladesh

S M Rezaul Karim MP
Minister
Ministry of Fisheries & Livestock
Govt. of the People's Republic of Bangladesh
Bangladesh Secretariat, Dhaka

Message

I am very happy to learn that Bangladesh Fisheries Research Institute (BFRI) is going to publish Annual Report 2021-22. It is very important and praiseworthy initiative.

Fisheries sector has a lot of contribution to food, nutrition, and socio-economic development of Bangladesh. As a nutrient rich ideal food, fish plays an important role in improving public health and building healthy nation. Under the dynamic leadership of our Honorable Prime Minister Sheikh Hasina, the present government is making all out efforts for conservation, development and scientific management of these important resources for sustainable production. For her dynamic steps even in the corona pandemic situation, the production and supply of fish was uninterrupted. In Covid-19 situation three countries of the world achieved a good progress in fish production. Bangladesh is one of them.

Modern aquaculture technologies developed by the Bangladesh Fisheries Research Institute (BFRI) is playing an important role in the fisheries sector of the country. BFRI has already developed breeding and culture technologies of 37 endangered species and established Live Gene Bank for the conservation of the fisheries resources. On the other hand, the Government is implementing various programmes for the development of the fisheries sector which include management of hilsa fish, open water stocking, establishment of sanctuaries and beel nurseries, habitat restoration and transfer of new technologies to the fish farmers. BFRI is working a lot in this regard.

Under the diplomatic leadership of our Honorable Prime Minister Sheikh Hasina sovereign right over a vast area of the sea was established, which is almost equivalent to the land boundary of Bangladesh. That's why a new horizon of blue economy was unveiled. Proper exploration and utilization of both conventional and non-conventional fisheries items have the potential to become a major source of the blue economy. The Government has taken various steps for seafood product development from conventional and non-conventional items including seaweeds, crustaceans and molluscs for domestic and international market through scientific research. Fishing ban for 65 days from May 20 to July 23 each year is in force to control over exploitation, maintain marine fisheries biodiversity and propagation. BFRI is contributing to explore sea centered resources through their research work.

I would urge upon the scientists, development and extension workers and all concerned working in the fisheries sector to work together to achieve the goals as outlined in 8th Five Year Plan, SDG, Vision 2041, Delta Plan 2100 and all other plans of the Government in realizing the dream of the Father of the Nation Bangabandhu Sheikh Mujibur Rahman to build Sonar Bangla, a prosperous Bangladesh.

I expect all the research works and innovative activities of BFRI will be depicted in annual report. I also expect this publication will be a knowledge reference for the future scientist in this sector. I wish all the success of the activities of BFRI.

Joy Bangla, Joy Bangabandhu,
May Bangladesh Live Forever.

(S M Rezaul Karim MP)

Dr. Nahid Rashid
Secretary
Ministry of Fisheries & Livestock
Govt. of the People's Republic of Bangladesh
Bangladesh Secretariat, Dhaka

Message

The present Government is totally committed to harness the full potential of the fisheries sector. With this view, the Government has undertaken various development projects for a balanced development of the inland capture and culture fisheries, and coastal and marine fisheries. The important programmes that are being implemented under these projects include cluster-based shrimp forming, eco-friendly aquaculture, community-based fisheries management, conservation of natural fisheries, restoration of degraded fisheries habitat, establishment of fish sanctuaries and closed fishing season, alternate employment for fishers, establishment of Live Gene Bank, development of breeding techniques for endangered fish, utilization of non-conventional fisheries resources and value addition and quality control. As a result, production has significantly increased and thus, fish which once became very scanty is now available in plenty.

Fisheries sector has also made a special position in world fish production, being one of the top ten fish producing countries. In global production, Bangladesh ranks 3rd in inland capture fisheries and 5th in inland aquaculture production. These two sectors have made much advancement due to scientific research. Now the government has given special attention on the development of the marine fisheries resources which have great potential to enhance Blue Economy.
The present per capita daily fish intake in the country is 62.58 g which exceeded the targeted 60 g. According to the FRSS, 2022 the fisheries sector contributes 3.57% of the county's total GDP and 26.50% of the agricultural GDP. In 2008-09, the country produced 27.01 lakh MT fish, which has increased to 46.52 lakh MT in 2020-21. More than 12% of the total population of the country, directly or indirectly are engaged in fisheries and ancillary activities.

Bangladesh Fisheries Research Institute (BFRI) have done a commendable job in fisheries research and development activities. It has successfully developed breeding and culture technologies for many endangered fishes which are now being commercially cultured by the fish farmers and entrepreneurs. The current research focus of the institute is the development of technologies for utilization of the non-conventional marine fisheries resources. In the meantime, the institute has made a good progress in this aspect. In addition to development of breeding and culture technologies for some important non-conventional items, it has successfully produced a number of seaweed- based food products which have a great demand in export market. We need to further refine and popularize these value-added products among the consumers.

I am very happy to learn that Bangladesh Fisheries Research Institute (BFRI) is going to publish Annual Report 2021-22 focusing mainly the research progress and achievement of the institute. I believe that this document will be helpful to researchers and planners for formulating future plans for the development of fisheries sector.

I wish continued progress of BFRI in its all research endeavor.

(Dr. Nahid Rashid)

Dr. Yahia Mahmud
Director General
Bangladesh Fisheries Research Institute

Message

Fisheries sector plays an important role in nutritional food security, livelihood development, poverty reduction, export earnings and economic development of Bangladesh. However, there is a great scope to enhance the contribution of the sector through proper research and development. Keeping this in view, Bangladesh Fisheries Research Institute (BFRI) has been conducting research reflecting the national policy and demands. BFRI has so far developed 75 improved aquaculture and management technologies many of which have been disseminated in the field. As a result, fish production has increased from 2.70 million MT to 4.621 million MT during the last 12 years.

The mandate of BFRI is to carry out and co-ordinate fisheries and aquaculture research in the country in consonance with the national policy and plan and Sustainable Development Goals (SDG) of the United Nations. To achieve this goal, we have also undertaken the collaborative research with relevant universities and organizations for faster development of appropriate technologies through utilization of available expertise in the country. The Institute follows bottom-up approach in planning research programmes which are finalized through the annual workshop participated by different stakeholders such as academicians, scientists, extension workers, policy makers, farmers and entrepreneurs.

The research programmes and other administrative activities performed by the Institute during 2021-22 for the development of the sector have been presented in these Annual Reports. A total of 48 research project during 2021-22 were implemented in different regional stations and sub-stations of the institute during the reporting period. It succeeded to develop seed production and culture technologies of endangered fish species, Balachata (*Somileptes gongota*), Mud Crab (*Scylla olivacea*) and identified a new breeding ground of Hilsa in Baleshwar River of Barisal district during the reporting period. Other activities included in the report are training, publication and financial matter of the Institute.

While aquaculture has been progressing very well due to development of various technologies, some of the new and emerging issues have cropped up in the process, which need to be seriously addressed to maintain the current growth of the aquaculture industry. On the other hand, marine sector is a thrust area in the current context, where new research intervention is needed on priority basis. In this context, along with the conventional resources, BFRI has given more focus on the development of non-conventional marine fisheries items like oyster, mussels, crabs and seaweeds.

We hope, this Annual Report with a focus on the major research conducted last year will be useful to researchers and planners of different national and international organizations in formulating project proposals and policy guidelines for fisheries development in Bangladesh.

(Dr. Yahia Mahmud)

ANNUAL REPORT
2021-2022

Bangladesh Fisheries Research Institute
Mymensingh
www.fri.gov.bd

ANNUAL REPORT
2021-2022

Editors
Dr. Yahia Mahmud
Dr. Md. Anisur Rahman
Dr. Md. Zulfikar Ali
Dr. Mohosena Begum Tanu
Dr. Anuradha Bhadra

Co-Editors
Mohammad Ferdous Siddique
Md. Hashibur Rahman
Md. Ayenuddin Haque

Published by
Director General
Bangladesh Fisheries Research Institute
Mymensingh

Designed by
S. M. Sariful Islam

Annual Report 2021-2022. Bangladesh Fisheries Research Institute, Mymensingh. 352 p.

CONTENTS

Bangladesh Fisheries Research Institute: An Overview

The fish and fisheries are integral part of the culture and heritage of Bangladesh. The sector plays a significant role in nutrition, employment generation and foreign exchange earnings. Keeping in view of the immense potentials of the sector in providing better nutrition and job opportunities, particularly to the poorest of the poor, and the urgency for optimum scientific utilization of the aquatic heritage, the President of the People's Republic of Bangladesh was pleased to promulgate an Ordinance entitled "The Fisheries Research Institute Ordinance 1984" on 11 July 1984. In pursuance of this Ordinance, the Fisheries Research Institute (FRI) was established in July 1984. In 1997, the FRI has been renamed as Bangladesh Fisheries Research Institute (BFRI) through the amendment of the 1984 Ordinance.

Though the Institute was established in 1984, it actually started functioning in 1986 with the recruitment of required manpower and creation of initial research facilities. Since then, the institute has been playing a key role in assisting the nation to achieve the goal of fisheries development as set out in successive development plans.

Vision of the Institute

Development of need-based technology leading to increasing fisheries production of the country.

Mission of the Institute

To conduct research for the development of need-based technology on aquaculture and fisheries resource management of the country.

Mandate of the Institute

- To carry out basic and adaptive research for development and optimum utilization of all living aquatic resources and coordinate fisheries research activities in Bangladesh;
- To conduct experiment and standardize techniques for maximizing productions and better management of living aquatic resources;
- To identify new production opportunities and develop them to usable levels;
- To develop skilled research manpower through training;
- To transfer developed technologies to users through training of extension workers, planners, fish farmers and other stakeholders;
- To advise the Government in all matters relating to research and management of living aquatic resources.

Management of the Institute

The Institute (BFRI) is an autonomous research organization and linked up administratively with the Ministry of Fisheries and Livestock, Government of the Peoples' Republic of Bangladesh. The general direction, administration and supervision of the affairs of the institute is vested in the

Board of Governors consisting as follows:

Board of Governors

Chairman	: Hon'ble Minister, Ministry of Fisheries and Livestock
Vice-Chairman	: Secretary, Ministry of Fisheries and Livestock
Members	: Executive Chairman, Bangladesh Agricultural Research Council
	: Vice-chancellor, Bangladesh Agricultural University, Mymensingh
	: Member (Agriculture), Planning Commission
	: Director General, Department of Fisheries
	: Two Members of the Parliament to be appointed by the Govt.
	: Two persons to be appointed by the Govt. among the persons having interest in fisheries development
	: Two persons to be appointed by the Govt. engaged in research in BFRI
Member-Secretary	: Director General, BFRI

Board of Governors may exercise all powers and doing all acts and things that may be performed or done by the Institute. The Board may appoint such committees, as it may consider necessary to assist it in the performance of its functions. As the Chief Executive of the Institute, the Director General takes appropriate steps in implementing its programs in the light of the policies and directives formulated by the Board of Governors.

BFRI Organogram

The Headquarters of the Institute is located at Mymensingh. The Institute has five research stations and five sub-stations based on different aquatic ecosystems. The organogram of the institute is shown in next page.

Stations and Sub-stations

Headquarters, Mymensingh

The Headquarters of the Institute is located at the south-west corner of the Bangladesh Agricultural University, Mymensingh, which is about 120 km north of the capital city, Dhaka. The Headquarter functions through its various divisions in respect of administrative development, coordination and operation of its research programs.

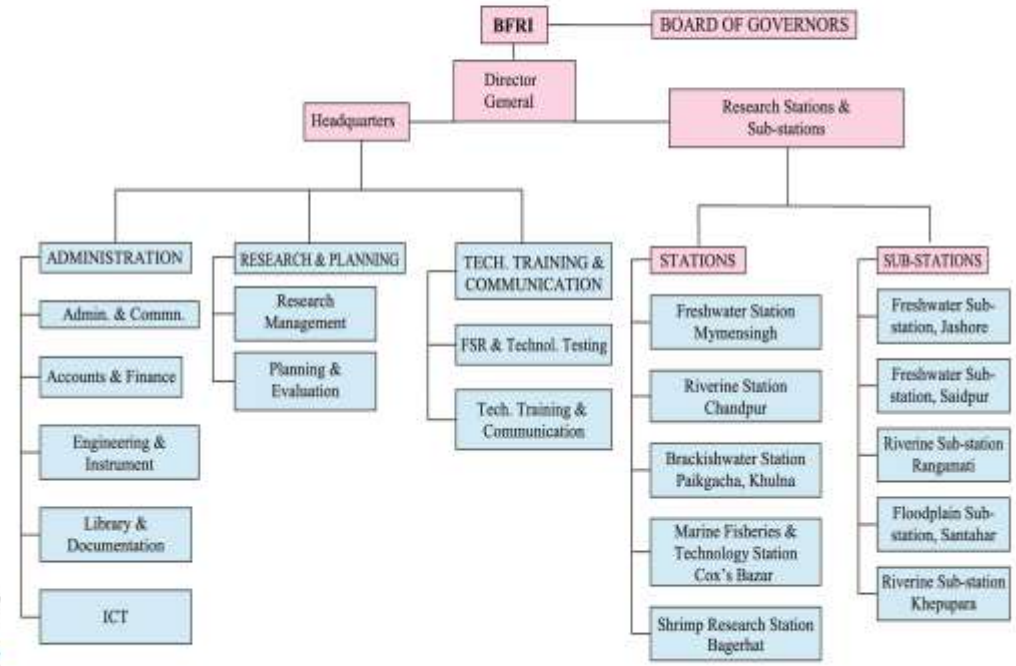
The divisions are:

- Research and Management
- Planning and Evaluation
- Technology Testing, Training and Communication
- Administration and Common Service
- Engineering and Instrument
- Library, Documentation and Public Relations
- ICT
- Accounts and Finance

ORGANOGRAME

Bangladesh Fisheries Research Institute

Mymensingh



Freshwater Station (FS), Mymensingh

The largest station of the Institute, with an area of 40 ha is located at Mymensingh attaching to the BFRI Headquarters. The station has well established and equipped with sophisticated carp and prawn hatcheries. The station has as many as 118 drainable ponds consisting of 20 mini ponds; 52 nursery ponds (0.1 ha each), 47 rearing ponds (0.25 ha each) and 16 grow-out/brood stock ponds (1.6-2.6 ha each). Other physical facilities include a feed store, office buildings, residential quarters, a 35-bed dormitory, a community center and a 5-bed guesthouse. The station is actively involved in conducting research on hatchery management, fish genetics and reproduction, carp polyculture, integrated fish farming, fish feed and nutrition, pearl culture, fish disease, health management and socio-economic aspects. The various research activities of the station are implemented by the following divisions:

- Reproductive Physiology and Genetics
- Aquaculture and Farming System
- Nutrition, Food and Feed Technology
- Fish Disease Diagnosis and Health Management
- Soil, Water and Productivity Management
- Fisheries Socio-economics

Mr. S M Rezaul Karim MP, Honourable Minister for Fisheries and Livestock inaugurated the Live Gene Bank of BFRI.

Three sub-stations are in operation with the Freshwater station.

Floodplain Sub-Station, Santahar: To support the floodplain fisheries development program taken up by the Government, studies on the ecology, limnology and gear selectivity of floodplains are being undertaken at the Santahar Sub-station. The sub-station has succeed in breeding and culture of certain endangered fish species such as *Aspidoparia jaya*, *Neotropius atherinoides* etc.

Freshwater Sub-station, Jashore: To support the farmers and hatchery operators of greater Jashore region, the Freshwater Sub-station has been conducting research on breeding and culture of BFRI super Tilapia, carp disease diagnostic services and also farming system research and development.

Freshwater Sub-station, Saidpur: To support the fisheries development program in northern region of Bangladesh, a freshwater sub-station is established in Saidpur Upzilla under Nilphamari. The prime objective of the sub-station is to conduct need-based research to suit with the ecosystem of northern Bangladesh and to transfer technology to the farmers through effective training and demonstration. The sub-station succeeds in breeding and culture of certain endangered fish species such as *Barilius* spp., *Mystus bleekeri*, *Labeo dero*, *Labeo angra* etc.

Riverine Station (RS), Chandpur

The station is situated in the riverine port city of Chandpur, with an area of 17.2 ha and has 36 non-drainable ponds ranging in size from 0.12 to 0.37 ha each and with a total of 8.6 ha water area. In addition, the station has one carp, catfish and prawn hatchery, two deep tube-wells, specialized laboratories, library, office buildings, residential quarters and an 8-bed guest house. One research vessel, one mechanized wooden boat equipped with research facilities, and three speed boats are available for undertaking riverine survey and studies relating research and management to hilsa and other riverine fisheries resources. The Riverine Station consists of 6 research divisions, which are as follows:

- Stock Assessment and Resource Dynamics
- Fisheries Resource Management and Conservation
- Culture-based Fisheries Management
- Reproductive Biology of Riverine Species
- Environment and Aquatic Pollution

Two Sub-stations are attached with the Riverine Station, and these are:

Riverine Sub-station, Rangamati: To devise sustainable management and development strategies for the Kaptai lake fishery, Riverine Sub-station (RSS) undertakes various adaptive research programs. Priorities are given on continuous monitoring of biological productivity, stock assessment, natural spawning, and population dynamics of various commercially important fishes and major carps, in particular. Recently, RSS has been introducing pen and cage aquaculture programs in the creeks and lagoons of Kaptai lake to culture fingerlings of major carp and thus to support artificial stocking of the lakes by Bangladesh Fisheries Development Corporation (BFDC), Kaptai lake project. Extension works are being carried out through adaptation of pen and cage aquaculture, installation of pens and cages in the creeks/coves in Kaptai lake on participatory basis.

Riverine Sub-station, Khepupara, Patuakhali: The fish landing and wholesale center of BFDC at Khepupara Upazilla has been handed over to BFRI to develop as a Sub-station and carry out research mainly on hilsa fishery. The old infrastructure has now been renovated by BFRI. In addition to this, technical advice to the fish farmers is being provided and improved fish seeds are distributed to the local farmers time to time.

Brackishwater Station (BS), Paikgacha, Khulna

The station was established in 1987 with a view to undertake research and development activities on various aspects of coastal aquaculture and fisheries management. The station is located at Paikgacha Upazilla under Khulna and has an area of 30.56 ha. The station has got 53 drainable experimental bracksihwater ponds of different sizes ranging from 0.05 to 1.0 ha, an experimental hatchery for the production of prawn and commercially important brackishwater finfish seeds and a number of laboratories. The station has 5 research divisions, such as:

- Nutrition and Feed Technology
- Disease Diagnostic and Health Management
- Brackishwater Aquaculture
- Estuarine Ecology and Environment
- Soil, Water and Productivity Management

This station is involved in conducting research on increasing productivity of coastal ghers, environment friendly shrimp culture development, crab seed production and fattening, seed production and culture of commercial finfishes, diseases management, aquatic environment monitoring etc. The research work undertaken so far by this station includes socio-economic studies on shrimp farming, survey and assessment of shrimp fry resources and its breeding ground, production potential of gher fishery (with improved management practices), polyculture of shrimp and mullet, breeding, culture and fattening of mud crab (*Scylla olivacea*), breeding and nursing of *Macrobrachium rosenbergii*, improved method of shrimp farming, breeding and culture of brackishwater catfish and green back mullet etc.

Marine Fisheries and Technology Station (MFTS), Cox's Bazar

This station, with an area of 4 ha, was established at Cox's Bazar in 1991. The station is being equipped with a crab breeding hatchery, live feed laboratory, outdoor complex with 39 cisterns (200 m^2 each), residential buildings for officers and staff accommodation, service building and an 8-bed guest house. There is a new 7-storied laboratory cum office building now under construction.

The mandate of the station includes research on marine ecology, seaweeds culture, environmental studies, stock assessment and population dynamics of commercially important species, diseases diagnosis and control, development of processing and preservation technologies, socio-economic studies of marine and coastal fishers and quality control of marine products.

Shrimp Research Station (SRS), Bagerhat

The station was established on 2010 at Sadar Upazilla under Bagerhat with an area of 8.0 ha. The mandate of the station is to conduct research on enhancing shrimp production, shrimp health management, shrimp feed and nutrition, post harvest handling and quality control of shrimp and shrimp products. The station consists of a 2-storied Office-cum-Laboratory building, 3-storied Staff dormitory, and 4-storied Training dormitory of the station. Moreover, a pond complex composing 9 experimental ponds of different sizes are being used for experimental purposes. The laboratories of the station are:

- Shrimp Health Management
- Quality Control
- Shrimp Feed and Nutrition
- Water and Soil Quality Management

Manpower

The manpower status of the Institute is highlighted in the following table:

Head	Approved posts			Filled up posts		Vacant posts	
	Officer	Staff	Total	Total	Staff	Officer	Staff
Revenue	268	257	525	112	186	155	71

Development of Technologies

Regular research activities of the institute lead to generate various aquaculture and management technologies for better management of the resources and increase the fish production. Till 2022, the Institute has evolved more than 75 aquaculture and fisheries management technologies. Among them, 11 technologies have been developed during 2021-22 period and these are as follows:

- ✓ Induced breeding and seed production of endangered fish sp. loach, balachata *(Somileptes gongota)*
- ✓ Breeding and seed production technology of mud crab *(Scylla olivacea)*
- ✓ Induced breeding technology of boirali *(Barilius barila)*
- ✓ Induced breeding technology of angus *(Labeo angra)*
- ✓ Induced breeding technology of kholisha *(Colisa fasciatus)*
- ✓ Induced breeding technology of jatpunti *(Puntius saphore)*
- ✓ Pearl culture in freshwater mussel
- ✓ Induced breeding technology of kursha *(Labeo dero)*
- ✓ Induced breeding technology of loittatengra *(Mystus bleekeri)*
- ✓ Induced breeding and seed production technology of dhela *(Osteobrama cotio)*
- ✓ Determination of standing biomass sustainable yield (MSY) of hilsa *(Tenualosa ilisha)*

Technology transfer: Subsequent to development of technologies or management practices, the generated research results were transferred through various mechanisms. Different government agencies including Department of Fisheries, NGOs, farmers and entrepreneurs were offered training on research-evolved technologies. After successful maturation of technologies, printing materials like manuals, booklets, leaflets, posters etc. were published and distributed among the users.

On-Farm trials: Field trials of the on-station research findings were conducted for adaptation of technologies in on-farm conditions through government and non-government extension agencies, private entrepreneurs and NGOs.

Farmer's Advisory Services: The Institute through its different stations and sub-stations provided advisory services to the farmers on improved fish farming technologies, water quality monitoring, feed quality, diseases control etc. Scientists of the institute also provided service on national crises related to fisheries and environmental issues as and when deemed necessary.

Development Projects

Four development projects have been implemented by the Institute during the 2021 to 2022:

Project Title	Cost (Lakh Tk.)	Project Period	Objectives of the Project
Strengthening of Hilsa Research in Riverine Station, Chandpur	3474.00	January 2017- June 2022	• To establish office cum hilsa laboratory building and other infrastructures for strengthening hilsa research in the Riverine Station. • To carry out demand driven research for development of appropriate technologies for increasing production and conservation of hilsa fisheries resources. • To provide technology-based training to different stakeholders on production and conservation of hilsa fisheries.
Strengthening Marine Fisheries Research and Infrastructure Development	5425.72	July 2017- December 2022	• To strengthen marine fisheries research capability in Bangladesh. • To develop strategy for the management of fisheries resources of the Bay of Bengal of Bangladesh. • To develop technology for the breeding and culture of commercially important marine species. • To develop post-harvest technology for proper utilization of marine fisheries resources of Bangladesh.
Seaweed culture and seaweed product development in Bangladesh coast	1686.00	January 2018-June 2022	• To survey and identify the economically important seaweed from Bangladesh coast • To select proper culture area and sustainable development of different seaweed culture technology • To ensure the commercial utilization of seaweed derived from Bangladesh coast.
Conservation, Propagation and Culture of Mussels and Snails in Bangladesh	1130.00	July 2017- June 2021	• Development of population base line of mussels and snails in Bangladesh. • Development of seed production and culture technology of important mussels and snails. • Conservation of natural stock of mussels and snails. • Awareness development for conservation of mussels and snails, and hands-on training for extension workers, farmer and entrepreneurs.

Training Programs

A series of well-structured training programs are organized by the Institute every year to disseminate the research evolved technologies to the end users. Moreover, effective transfer and dissemination of the technologies and management procedures such as training of extension workers both of Government and NGOs, teachers, students and Journalists are also organized by Institute. The training programs organized on

different aspects are as follows:

- Improved fish culture and management
- Seed production and culture techniques of endangered fish species
- Pearl culture techniques in freshwater ponds
- Shrimp nursery, culture and management
- Crab fattening techniques
- Pen and cage culture techniques
- Fisheries and aquaculture research management
- Mud eel culture technique
- Seaweed culture and product development
- Effect of sanctuary on Hilsa production
- Culture technique of Mussels and Snails in Bangladesh

The Institute also conducts training on research methodology, financial management, office management, e-filing, e-GP and other research-oriented programs for researchers of the Institute to shine up their capability.

Training programs conducted: For boosting-up fish production and to ensure better utilization of aquatic resources, BFRI organizes series of training programs every year for farmers, entrepreneurs, unemployed youth, rural women and university students, extension workers both of Government and NGOs, teachers, journalists and LGED fisheries facilitators. The main objective of offering such type of need and opportunity-based training is to transfer and disseminate technologies among various stakeholders and end users. During July 2021-June 2022 a total of 124 training batches were completed and 2,845 individuals were trained up by the Institute.

Institutional manpower development: For strengthening the capabilities of scientists, administrative and management personnel, the institute organizes different in-country and overseas short-term and long-term training programs, study tour and experience-sharing visits. During 2021-2022, a total of 13 scientist achieved overseas short-term and long-term training in 4 programs, besides, 14 different in-country training programs have been organized for the scientists and officers. 2 scientists have been awarded PhD from abroad

Workshop/Seminar organized: The Institute organized 8 National workshops and seminars in different disciplines to identify the problems and sharing and exchanging knowledge generated through research in this year.The institute and its stations and sub-stations organize regional and national workshops every year to review the research projects and to present the research progress of the institute.

Public Relation and Publications

Bangladesh Fisheries Research Institute Library and Documentation Centre (FRILDOC) act as a repository of literature and technical information and provides latest information on scientific research and experimental development in all branches of fish and fisheries. The most of the FRILDOC collection backup on the subjects: aquaculture, brackish water aquaculture, mariculture, marine science, biology, ecology, environmental science, agriculture, life sciences, sea weeds, plankton, food processing, feeds, zoology, botany, geography, economics, marketing, geology, socioeconomics, rural development etc.

Library and Documentation

The library has 9108 technical and general books 186 titles of scientific periodicals 5120 miscellaneous publications. The library also has several types of reference books, academic dissertations, government and others departmental publications.

The FRILDOC is operating in fully automated environment. The various activities of the centre have been computerized using Library Management Information System (LMIS) software.

The FRILDOC provides the following documentation services:

Head	Approved posts
• Document Delivery Service • Current Awareness Service i) Current Content Service ii) Monthly Accession list iii) Monthly News paper Articles	• Reference service • Bibliographical service • Abstracting service • SDI (Selective Dissemination Information) Service • Internet Service • Photocopy Service • ASFA (Aquatic Sciences and Fisheries Abstract) DVD Service • TEEAL (The Essential Electronic Agricultural Library) Service • Digital Library Service (BFRI in Aquatic Commons digital repository (http://aquaticcommons.org/view/issuing_agency/Bangladesh_Fisheries_Research_Institute.html) • Hinari, AGORA, OARE, ARDI and GOALI (The Research4 Life programme) Service

During the reporting period of July 2021 to June 2022, a number of books, Journals, periodicals etc. procured for the library. The library has also received a noticeable number of books journals, periodicals, proceedings, research reports, annual report, newsletters and magazines on complimentary and exchange basis. The library-maintained exchange programme with more than 75 leading national and International organizations. The category wise list is shown below:

Items	2021-2022
Books	185
Journals	17
Reports/Proceeding of seminars and workshops/papers	11
Newsletters/Bulletins/Reprints/Off prints	43
ASFA (Aquatic Sciences and Fisheries Abstract) DVD	up to 2017
TEEAL (The Essential Electronic Agricultural Library)	up to 2013

The library maintained free mailing of institutional publications to various research organizations, Universities, NGOs, entrepreneurs and farmers to keep the aware with the latest development in fisheries research.

Working Linkage

The overall research, training and management activities of the institute were carried out in close cooperation and linkages with various national and international organizations/agencies. The institute also maintained close contact with public extension organizations, different NGOs working in the country, for dissemination of technologies and obtaining feed-back from them. BFRI collaborated with national universities and maintained close liaison for fisheries research and development (R and D). Among the national collaborators, definitely the main focus implies to the Department of Fisheries (DOF) followed by NARS Institutions and joint research and development programs with different NOGs.

Finance and Accounts

The sources of funds of the institute comprise grants from the government, and grants from different donor agencies. Government grant from the revenue budget is usually provided to meet only salaries and allowances of staff small protein of operational costs. The cost of development, maintenance and research is also borne by the government from its development budget provided in the form of development project.

Receipts and Expenditure

The institute received an amount of 4274.50 lakh during the year 2021-22 from the government revenue budget and the expenditure incurred of the financial year 2021-22 was 3954.42 lakh.

RESEARCH PROGRESS

2021-2022

Improved Germplasm Production of Carps, White Pangas and Pure-line Breeding of Kalibaus (*Labeo calbasu*)

Researchers

Dr. Mohammad Ashaf-ud-Doulah, SSO
A.K.M. Saiful Islam, SSO
Md. Rabiul Awal, SO
Abul Bashar, SO
A.N.M. Rezvi Kaysar Bhuiyan, SO

Objectives

- To upgrade and produce quality seeds of Indian Major Carps, Subarno Rui, Catfish and distribute to the fish farmer, entrepreneurs and hatchery owners
- To develop live gene bank with quality brood stocks through implementation of effective breeding plan
- To produce and evaluate the growth performance of selected pure breeds with non-selected breeds of Kalibaus (Generation to generation)

Achievements

Detailed work carried out during reporting period (July 2021 to June 2022)

The following research work have been carried out under the project during July 2021 to June 2022.

Experiment 1.Production of F_1 base population of pure-line Kalibaus (Labeo calbasu) through genetic selection

Production of F_1 base population of Kalibaus (pure and cross breed line) during 2021.

Two wild populations of Kalibaus *(L. calbasu)* were collected in 2020 from river Meghna and *Haor* sources and reared in separate earthen ponds in Freshwater Station's pond complex. The present status of the pure Kalibaus of river Meghna and Haor stocks are shown in Table 1.

Table 1. Present status of wild breeds Kalibaus of the river Meghna and Netrokona *Haor.*

Sources of stock	Length range (cm)	Weight range (g)	Number (pairs)	Remarks
Haor	25 - 35	300-400	50	Broods collected from wild sources were used for planned breeding program during 2021.
Meghna (group 1)	32 - 55	800-2000	110	
Meghna (group 2)	9-20	25-50	510	Used for brood stock development

In 2021, founder F_1 generation (both pure and cross breed) was produced through mating the broods following the standard breeding protocol in BFRI hatchery complex. Two pure line crossbred lines *viz*., Meghna (Female) X *Haor* (Male), Meghna (Male) X *Haor* (Female) and two pure-lines *viz*., Meghna (Female) X Meghna (Male), *Haor* (Female) X *Haor* (Male), respectively were produced.

Development of broods to produce F_1 generation of pure and crossbreed line Kalibaus

After pond preparation (drying, repairing, liming, and watering), fry of F1 founder Kalibaus were stocked in BFRI pond complex in October 2021. For brood stock development, fingerlings were stocked at the rate of 10000 fingerlings/ha. During all phases of the growing period, the fish were fed 25-28% protein rich feeds. At the age of 0.5 years or more, randomly selected 20% fish (sex ratio female X male 1.1) were kept in brood ponds until being used for planned breeding for producing next generations *viz*., F_2, F_3, F_4 and to be continued. The overall breeding protocols of Kalibaus are shown in Figure 1.

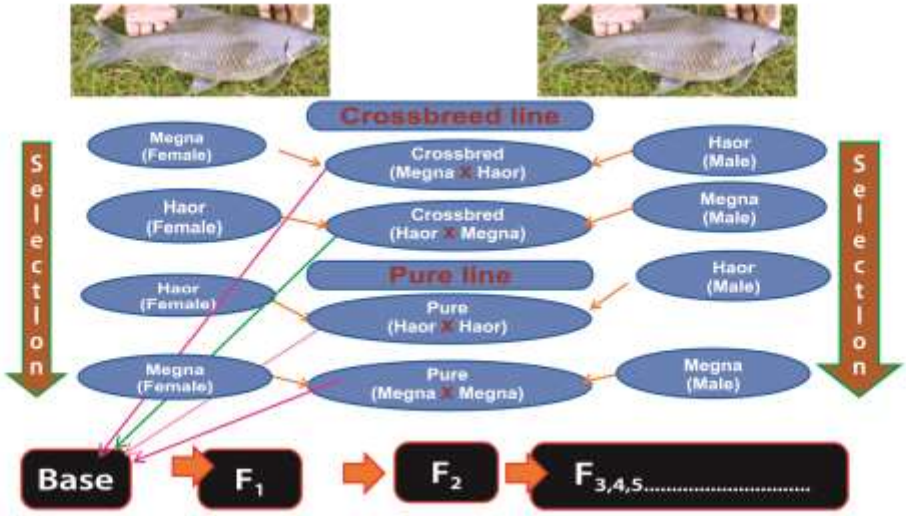


Figure 1. Schematic flow-chart of breeding protocol of Kalibaus

Experiment 2. Growth evaluation of pure line stocks of Kalibaus (L. calbasu)

For evaluation of growth performance of F_1 generation, growth trial was conducted using fingerlings (4-6 cm) of Kalibaus at rearing ponds since September 2021 in Freshwater Station Pond complex. The experimental design is given below.

Study Area	: On-station earthen ponds
Ponds	: 20 decimal
Breeds (Kalibaus)	: Crossbreed with pure-line Kalibaus
Stocking density	: 60 fingerlings/deci. (monoculture system)
Feeding	: 5% bw/d (25% protein)
Rearing Period	: 6 months

The stocking density was maintained 60 fingerlings/deci. and the fish were fed commercially available nursery feed (25-28% protein). To assess the growth performance, sampling was done on monthly basis. At least 30 fish from each pond were sampled to measure body weight in gram and total length in centimeter with a digital balance and wooden measuring scale, respectively.

Figure 2. Sampling of Kalibaus during growth trial

a. Growth performance

The experiment was conducted at the pond complex of Freshwater Station (FS) of BFRI, Mymensingh for a period of 6 months from November 2021 to April 2022 to observe the growth performance and yield performance of two crossbred lines *viz*., Meghna (Female) X *Haor* (Male), Meghna (Male) X *Haor* (Female) in the captive condition. The fish were stocked as per experimental design. After 150 days of culture, mean sampling weight was found 180.4±7.0 g with mean weight gain was 172.25±7.0 g. Daily average weight gain was 0.96 g. The SGR of this stock was 1.95 % per day. Health condition (g/cm) of two crossbreed lines with pure line stocks of Kalibaus (*L. calbasu*) was a 3.39±0.56. (Table 02).

Table 2. Growth evaluation of two crossbreed lines with pure line stocks of Kalibaus (*L. calbasu*) reared in ponds for 180 days culture period.

Growth parameters	Pure line crossbred Kalibaus
Mean initial weight (g)	6.15±0.42
Mean initial length (cm)	4.20±0.20
Mean sampling weight (g)	180.4±7.0
Mean sampling length (cm)	28.3±5.6
Mean weight gain (g)	172.25±7.0
Average daily weight gain (g/day)	0.96±0.1
Specific growth rate (%/day)	1.95±0.23
Health condition (g/cm)	3.39±0.56

b. Water quality parameters

Mean water quality parameters of the ponds are presented in (Table 3). As evidence from the Table, different water quality parameters were almost same in all the ponds and very much congenial for fish culture. The range of air temperature recorded was 19.0 to 31.0°C and range of water temperature was 15 to 28°C. pH of water was alkaline throughout the culture period and fluctuate from 7.3 to 8.2. Dissolved oxygen varied from 4.0 to 6.0 mg/l. Total alkalinity of the ponds ranged from

150 to 185 mg/l indicating high potential of water for primary production. Ammonia content of both ponds were below the toxic level and varied from 0.02 to 0.01 mg/l.

Table 3. Mean value of water quality parameters of ponds during 150 days of culture period.

Water quality parameters	Pure line crossbreed Kalibaus
Air temperature (°C)	22.58±4.04
Water temperature (°C)	19.91±3.84
Water pH	7.80±0.26
DO (mg/l)	5.6±0.38
Ammonia (mg/l)	0.02±0.01
Alkalinity (mg/l)	159.8±10.59
Hardness (mg/l)	110±7.43

Continuation of the quality mass seeds production of Catla, Rohu, Mrigal, BFRI Rajpunti, Pure-Line Silver/Bighead carps

- Source of broods.river Halda, Jamuna and BFRI improved stocks
- Establishment of Live-gene bank with improved quality broods
- Continuation of breeding followed standard protocols

Brood development of BFRI germplasm

a) Pond preparation

After complete drying of the ponds, weeds were cleared, pond dykes were repaired (if needed) and each pond was treated with lime at 250 kg/ha, cow dung at 3000 kg/ha, urea at 25 kg/ha, and TSP at 25 kg/ha. After preparation, ponds were filled with ground water up to 5 ft depth.

b) Stocking of broods

After pond preparation, broods of BFRI Rohu, Subarno Rui, Catla, Mrigal, Silver carp, and Bighead carp, were stocked in brood ponds maintaining stocking density at 1300 kg/ha. About1500 BFRI-GISB (individual weight ranges 200-500 g) were stocked in brood ponds and about 200 white Pangas (Vietnamese) of 2000-3000 g individual body weight were stocked.

c) Feeding

Fish was fed with feed composed of fishmeal (9%), Mustard oil cake (20%), Rice bran (70%), and Vitamin (1%) at a rate of 3% of body weight per day.

d) Fertilization

Cowdung and inorganic fertilizers were provided at 2000 kg/ha/month and at 50 kg/ha/month, respectively. Both fertilizers were used in every alternate week.

Table 4. Present status of wild breeds of river Halda and Jamuna in brood ponds.

Wild sources of breeds	Halda	Jamuna
	Individual weight range (g)	Individual weight range (g)
Catla	3500-8200	3100-7200
Rohu	2000-5500	2400-6300
Mrigal	2100-5400	2200-4200
Improve breeder	**Improved stock of BFRI**	
Pure line Kalibasu	800-1500 (g)	
Pure line Silver carp	2000-3000 (g)	
Pure line Bighead carp	2000-3000 (g)	
BFRI Rjpunti	200-1000 (g)	

These broods will be used for mass seed production in the year 2022 followed by the established breeding protocols.

Production anddistribution of BFRI improved germplasm

Following the established techniques of breeding, BFRI produced high quality carp seeds will be distributed to fish farmers, hatchery owners, and nursery owners across the country. Quantity of mass seed production target of carp species during the year 2021-22 is shown in Table 4.

Table 5. Production Target of improved germplasm of carps (2019-20).

Head	Production target (2021-22)		Breeding period
	Spawn (kg)	Fry/Fingerling	
Suborno Rui	20	10,000	April-June
BFRI Rohu	130	50,000	April-July
Mrigal	70	20,000	April-July
Silver carp	20	10,000	March-July
Bighead carp	20	10,000	March-July
BFRI-GISB	60	50,000	March-July
Catla	50	10,000	April-June
White Pangus	25	50,000	April-June
Kalibaus	15	5000	April-June
Total	**430**	**2,15,000**	

The declared production target of the carp hatchery of Freshwater Station under the project exceeded its margin for 2021-222. Overall production is shown in Table 6.

Table 6. Production achievement in 2021-22.

Species	Production target achieved (as of June 23, 2022)		Breeding period
	Spawn (kg)	Fry/fingerlings (Nos.)	
BFRI-Suborno Rui	69.050	50,000	April-July
BFRI-Rui	155.625	2,00,000	April-July
Mrigal	108.050	1,50,000	April-July
Silver carp	4.450	5,000	March-July
Bighead carp	31.125	20,000	March-July
BFRI-Rajpunti	120.125	30,000	March-July
Catla	58.975	5,000	April-June
White Pangus			April-June
Kalibaus	2.750	500	April-June
Total	**551.150**	**4,60,500**	

Distribution of spawn and fingerlings at field level

BFRI-produced spawn and fingerlings are being delivered nationwide to farmers, hatchery owners, and entrepreneurs to spread high-quality spawn and fingerlings at the farmer level. Most fingerlings were provided at no cost to government hatcheries, while spawn was provided to hatcheries and prominent farmers at the nominal rate

Table 7. Distribution of fry and spawn to field level.

Species	Description and size (cm)	Number/ amount	Distributed to
BFRI-Suborno Rui	Brood 30-40 cm	7 pair	Govt. fish seed production farm, Ishwargonj, Mymensingh
	Brood 30-40 cm	5 pairs	Farmers, Ishwargonj, Mymensingh
	Fry 2-3 cm	12000	DoF Hatcheries
		15000	DoF-NATP farmers
		1500	Freshwater Sub-station, BFRI, Jessore
		2000	river ine station, BFRI, Chandpur
		1200	IUBAT
	Spawn (4 days old)	2.5 kg	Fish farmers and hatchery owners
		2 kg	BFDC
		0.5 kg	IUBAT
Pure-line Kalibaush	Fry 3-5 cm	2500	Fish Seed Multiplication Farm, Parbotipur, Dinajpur
		2500	Central Fish Hatchery, Koatchandpur, Jenaidah
		2500	Fish Training and Extension Centre, Faridpur

White Pangas	Fry 3-4 cm	20000	DoF-NATP farmers
BFRI-Rajpunti	Fry 8-10 cm	2500	riverine station, BFRI, Chandpur

Proposed work during next period

i. Continuation of development of broods of founder F1 base population through genetic selection

ii. Growth evaluation of two crossbreed lines with pure line stocks of Kalibaus (*L. calbasu*)

iii. Rearing of the broods of Catla, Rohu, Mrigal, BFRI Rajpunti, pure-line Silver/Bighead carps and continuation of the quality mass seed production

Stock improvement of major carps (Rohu and Catla) and DNA-barcoding of important freshwater fishes in Bangladesh

Researchers

Dr. Jonaira Rashid, SSO

Md. Amdadul Haque, SO

Objectives

- To improve Rui and Catla stocks using DNA technology
- To analyze genetic variability of Rui stocks using DNA markers
- To identify freshwater fish at the species level based on DNA barcoding data

Achievements

Detailed work carried out during reporting period (Component A).

Experiment 1. Stock improvement of Rohu through DNA based protocol

Growth performances of F_4 Rohu during communal rearing has been shown in Table 1. The growth performances of F_4 Rohu in terms of mean length and mean weight gain were 47.13±2.85 cm and 1524.07±36.28 g, respectively. Data on body weight, body length and sex were recorded 12 months of rearing.

Table 1. Growth performances of BFRI F_4 generation of Rohu in communal grow-out pond.

Parameters	Initial status (July 2021)	Status (December 2021)	Present status (June 2022)
Length (cm)	37.34±1.68	42.57±2.18	47.13±2.85
Weight (g)	810.92±34.39	1163.16±38.25	1524.07±36.28

Experiment 2. Stock improvement of Catla through DNA based protocol

For stock improvement, collected Catla fish from the river Halda were stocked in a pond having an area of 50 decimals. The average length and weight of stocked fish were 46.75±5.12 cm and 1385.23±230.37 g, respectively. The stocked fish were administered commercially available carp feed at 5-3% body weight once daily. The results of the present study on growth performances in terms of mean length (54.82±4.17 cm) and mean weight gain (1907.49±243.13 g) of Catla has been furnished in Table 2.

Table 2. Growth performances of Catla (The river Halda origin) in grow out pond.

Parameters	Initial status (July 2021)	Status (December 2021)	Present status (June 2022)
Length (cm)	46.75 ±5.12	49.15±4.83	54.82±4.17
Weight (g)	1385.23 ±230.37	1642.07 ±245.41	1907.49 ±243.13

Another wild stock of Catla fish was collected from the river Jamuna that were reared in the well-prepared pond having an area of 50 decimals using lime and fertilizer. Fish are being fed with commercially available carp feed at 5-3% body weight daily. The results of growth performances in terms of mean length (43.15±3.56 cm) and mean weight gain(1015.53±46.18 g) of Catla (The river Jamuna origin) has been presented in Table 3.

Table 3. Growth performances of Catla (The river Jamuna origin) in grow-out pond.

Parameter	Initial status (July 2021)	Status (December 2021)	Present status (June 2022)
Length (cm)	36.80±3.14	39.27±3.48	43.15±3.56
Weight (g)	498.68±35.14	782.31±34.53	1015.53±46.18

Figure 1. BFRI Suborno Rui

Experiment 3. Genetic evaluation of wild and BFRI Rohu (F_4) stocks

Genomic DNAs were extracted from fin tissues using commercially available kit (PureLink™ Genomic DNA Mini Kit, Invitrogen by Thermo Fisher Scientific, USA) following manufacturer's instructions. PCR reactions were performed on each DNA sample in a 20 µl reaction mix containing 1.5 µl of 20 pmol primer, 7.5 µl of DreamTaq PCR Master Mix (2X) (Thermo Fisher Scientific) and 50 ng of genomic DNA and a suitable amount of sterile deionized water. DNA amplification was performed in a thermal cycler (GeneAtlas Type G). The reaction mix was preheated at 94°C for 3 min followed by 40 cycles of 30 sec denaturation at 94°C, 1 min annealing at 34°C and extension at 72°C for 2 min. After the last cycle, a final step of 7 min at 72°C was added to allow complete extension of all amplified fragments. The amplified product from each sample was separated electrophoretically on 1% agarose gel (UltraPureTM, Invitrogen by Thermo Fisher Scientific, USA) containing SYBRTMSafe DNA Gel Stain (Invitrogen by Thermo Fisher Scientific, USA) in 1X TAE buffer at 120 V for 30 minutes. A molecular weight marker DNA (1 kb ladder) was electrophoresed alongside the RAPD reactions. DNA bands were observed on UV-transilluminator and photographed with a Gel Documentation system.

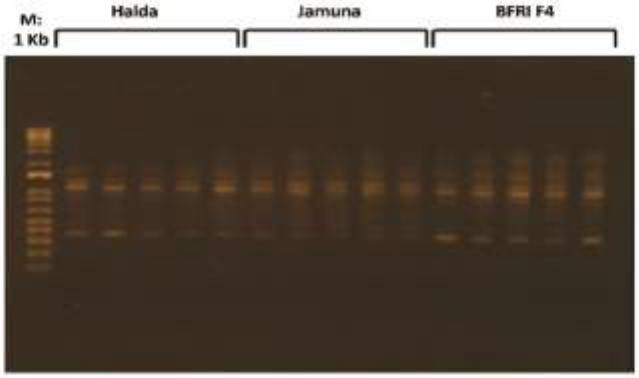


Figure 2. Genetic data analysis through RAPD marker.

Table 4. Number and proportion of polymorphic loci for the studied stocks.

Stocks	No. of polymorphic loci	Percentage of polymorphic loci	Gene diversity (h*)
Wild (Halda)	18	85.28	0.293±0.205
BFRI Rohu (F_4)	15	71.43	0.271±0.198
Wild (Jamuna)	14	66.67	0.257±0.209

Component B.

Experiment. 1 Identification and characterization of selected freshwater fish based on DNA barcoding data

A total of 82 freshwater fish samples were collected from different regions of Bangladesh for DNA barcoding. COI genes were successfully amplified from Deshi Pangas, Thai Pangas, White Pangas and different species of Gutum, Hiralu, Kholisha, Chapila and different species of Hisha. The PCR products

were purified using PureLink™ PCR purification kit for sequencing and data analysis. A total of 48 samples were sequenced and among them 21 species confirmed. In addition, DNA extractions and PCR amplifications were completed for 17 samples.

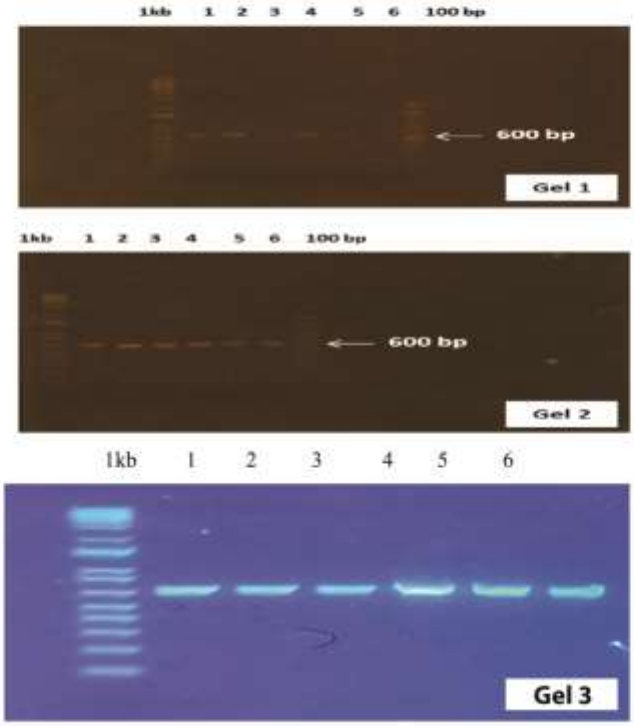


Figure 3. PCR amplifications of 600 bp amplified product of mt-COI gene of selected fish species; Gel 1. 1, 2. Deshi Pangas; 3, 4. Thai Pangas; 5, 6. White Pangas; Gel 2. 1-6; Gutum, Gel3. 1-Dhela, 2. Telchita, 3.Chela, 4. Botia, 5. Mohashol, 6.Chapila. Molecular weight marker (1Kb and 100 bp DNA ladder).

Table 4. Identification and characterization of selected freshwater fish based on DNA barcoding data

Gene bank		Conservation status	Family	Accession no.
Scientific name	Identity (%)			
Pangasius pangasius	100	EN	Pangasiidae	KC572135.1
Lepidocephalicthys annandalei	100	VU	Cobitidae	MZ606660.1
Lepidocephalichthys guntea	99	LC	Cobitidae	AP011338.1
Gudusia chapra	99	VU	Clupeidae	AP011603.1
Hilsa kelee	99	LC	Clupeidae	FJ158558.1
Coilia dussumieri	98	LC	Engraulidae	MK572136.1
Acanthocobitis botia	99	NT	Balitoridae	KT762380.1
Paracanthocobitis mackenziei	99.84	DD	Balitoridae	MK572439.1

Gene bank		Conservation status	Family	Accession no.
Scientific name	Identity (%)			
Canthophrys gongota	100	LC	Cobitidae	KX455897.1
Botia dario	99	EN	Cobitidae	KP974803.1
Tor barakae	99.56	DD	Cyprinidae	KJ936789.1
Glyptothorax telchitta	99	VU	Sisoridae	MT670296.1
Salmostoma bacalia	100	EN	Cyprinidae	MH087030.1
Puntius chola	100	LC	Cyprinidae	KJ936779.1
Salmophasia bacaila	99	LC	Cyprinidae	AP011223.1
Nemapteryx caelata	98	LC	Ariidae	KU894610.1
Cyprinus rubrofuscus	99	LC	Cyprinidae	MW969691.1
Esomus danricus	99	LC	Cyprinidae	KJ936756.1
Mystus gulio	99	NT	Bagridae	KX455898.1
Rhinomugil corsula	99	LC	Mugilidae	KT364790.1
Osteobrama cotio	99	NT	Cyprin	KT762359.1

EN = endangered, **VU** = vulnerable, **LC** = least concern, **NT** = not threatened, **DD** = data deficient)

Phylogenetic tree construction

A phylogenetic tree, also called a phylogeny, is a diagram that shows the evolutionary branches from which various species, creatures, or genes have descended from one another. We have constructed a phylogenetic tree to examine the evolutionary links between the sequences of the five species studied in this work using the software programs MEGA version 7 (Kumar et al., 2016) and ClustalW version 2.1 (Jeanmougin et al., 1998). In this phylogenetic tree, *P. mackenziei* and *A. botia* are very closely related and very recently they form separate branch.

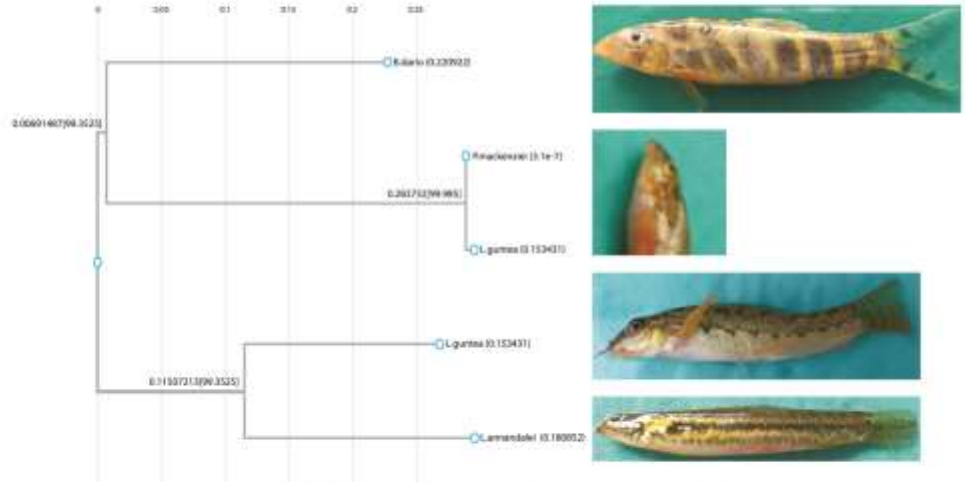


Figure 4. ClustalW phylogenetic tree of five fish species based on COIgene multiple sequence alignment

Improvement of Freshwater Pearl Culture Technology in Bangladesh

Researchers
Md. Nazmul Hossen, SO
Mohammad Ferdous Siddique, SSO
Sonia Sku, SSO

Objectives

- Improving existing techniques for producing different types of freshwater pearls
- Estimating economics of pearl culture

Achievements

The following research works were carried out under the project during reporting period (2021-2022)

Experiment 1. Pearl culture in confined (Aqurium, cistern and plastic drum) systems.

Preparation of confined systems (aquarium, cistern, and drum)

Aquarium, cistern and drum were used as confined systems which were disinfected by $KMnO_4$ and washed very well. After cleaning the system were filled up by the tap and pond water with proper aeration.

Collection of mussels

Mussels were collected from different freshwater regions and stocked in pond for three months before operation.

Calcium image preparation

Mussel shell powder marble, powder, grout powder, sun stone powder and glue were mixed to prepare a dough.According to desire design image were made by dough on dice. To set the images properly inside the mussel, the images were kept on dead shell of mussel for 3-4 hours while dough was soft. The prepared image was soaked in water overnight to eliminate the stench of glue.

Operation method

Mussel was opened to 8 mm and cleaned with tap water. Pocket was made between mantle tissue and mussel shell and single image was inserted into the pocket in each mussel. After insertion the inner air and water were removed, and valves of mussel were closed.

Culture method

After operation, the mussel was cultured in aquarium, cistern, and drum using the net bag hanging technique at a stocking density of 15 mussels/m^2. Water depth was kept 0.3-0.5 m. Different water quality parameters *viz*. temperature, pH, ammonia, DO and Ca^{2+} were monitored fortnightly. During culture period in confined condition plankton was provided for the cultured mussel as food. The production of plankton was done by using several systems that applied organic and inorganic fertilizers on a biweekly basis at the rates of 5 kg of organic manure, 0.125 kg of TSP., and 0.1 kg of urea per decimal.

Table 1. Design of the experiment.

Treatment	Types of treatment	Stocking density	Mussel species
T_1	Aquarium	15 mussels/m^2	*Lamellidens marginalis*
T_2	Cistern		
T_3	Plastic Drum		

In this experiment, a total of 600 mussels were operated with acrylic image to observe the image pearl production performance. During the time of culture, three distinct systems were set up in three distinct environments. an aquarium was placed in the laboratory, a cistern was placed in the hatchery beneath a shed, and a plastic drum was placed in direct sunlight. Regular monitoring was done to observe the survival rate in different condition. The mortality was found 33% in Aquarium, 25% in Cistern and 12% mortality occurred in plastic drum. Pearl quality was observed after final harvest.

Table 2. Image pearl production from different condition.

Treatment	Types of treatment	No of operated mussel	Mortality rate of operated mussel (%)	Comments
T_1	Aquarium	200	33%	Experiment ongoing
T_2	Cistern	200	25%	
T_3	Plastic Drum	200	12%	

Water quality parameterswere monitored fortnightly, and data were recorded. According to the recorded data, water quality parameters were found in suitable ranges for pearl culture. Parameters are given in Table 3.

Table 3. Water quality parameters.

Parameter / Treatment	Temp (℃)	DO (mg/l)	pH	NH_3 (mg/l)	Alkalinity (mg/l)
Aquarium	24.35±0.12	6.26±0.26	7.32±0.37	0.04±0.08	180.00±26.46
Cistern	26.53±0.65	5.84±0.35	7.16±0.36	0.02±0.04	190.00±10.00
Plastic drum	27.55±0.25	5.14±0.35	7.05±0.36	0.04±0.001	180.00±13.20

Experiment 2. Uses of inorganic and organic medicine to reduce operated mussel mortality

Pond preparation

Ponds were selected for stocking and rearing the collected mussels. Pond was prepared by following standard procedure. Ponds were totally drained out and dried. After drying, lime and salt were applied at the rate of 1 kg and 0.25 kg per decimal to remove the insect and earthworm. After 6-7 days of liming, pond was filled with water. After pond preparation mussels were collected from different freshwater habitats of Bangladesh. Collected mussels were reared in stocking ponds with foods and fertilizers. After three months of rearing the reared mussels were brought to laboratory before two hours of operation and then mussel was operated by following operation procedure. In this experiment *Lamellidens marginalis* was used for image/design pearl production. These species were collected from different freshwater regions.

Treatment

Just after operation, mussels were treated with turmeric powder as an organic medicine and antibiotic from flucloxacillin group in aquarium providing natural plankton. Medicine was applied at the rate of 3.5ml/250L water twice a day and water was exchanged on daily basis. After 7 days of treatment in aquarium the operated mussel transferred to the culture pond.

Table 4. Design of the experiment.

Treatment	Medicine	Replication	Number of operated mussels	Operated mussel species
T_1	Organic (Turmeric Powder or paste)	R_1	100	*Lamellidens marginalis*
		R_2	100	
		R_3	100	
T_2	Inorganic (Antibiotic-flucsoxacilin)	R_1	100	
		R_2	100	
		R_3	100	

Culture method

Operated mussels were cultured in pond under net bag hanging method, at a stocking density of 80/decimal. Net bag hanging method is a method where net bag is hanged from a rope into 30-35cm depth with float. The rope stretched across the pond in the surface of water. The distance between adjacent two bags were 25-30 cm and two hanging rope were 1.5 m. For stimulating and maintaining the growth of natural plankton, organic and inorganic fertilizers were applied fortnightly to the pond at the rate of 5kg organic manure, 0.125 kg TSP and 0.1 kg urea per decimal, respectively. Lime powder was applied at a rate of 0.5 kg per decimal. Water depth was kept in 1.5 meter. Water temperature, pH, NH_3, DO parameters was monitored fortnightly.

In aquarium, during 07 days of treatment there no mortality were observed. During the culture period, 30% mortality occurred in the pond treated with turmeric powder and 32.12% mortality occurred in Antibiotic treatment. Both organic and inorganic treatment showed similar results. Quality of pearl and final survival data will be collected and analyzed after final harvest.

Table 5. Survival rate observation after medicine treatment.

Treatment	Medicine	Number of operated mussels	Mortality rate of operated mussel	Comment
T_1	Organic (Turmeric Powder)	300	30.00%	Experiment is going on
T_2	Inorganic (Antibiotic)	300	32.12%	

Water temperature, pH, ammonia, dissolved oxygen and alkalinity were monitored fortnightly. In culture pond temperature varied from 21 to 30.40 ^{0}C while pH ranged from 7.1 to 8.1. In pond water dissolved oxygen differed from 5.6 to 5.9 mg/l while ammonia varied from 0.00 to 0.02 mg/l. Alkalinity of water ranged from 115 to 135 mg/l.

Experiment 3. On farm trial of image pearl culture at different location

Image pearl production technology was given in 3 different regions of Mymensingh for field trial. Field monitoring and evaluation is going on. At the end of the experiment produced pearl will be collected. After collection complete data will be recorded. Total yield will be given to the farmers. The details of demo farms are given below.

a. Muktagacha

- Among 400 operated mussels 10 % mortality was observed
- Recorded data of water quality parameters, Temperature 14-27 ^{0}C, DO 5.22-5.40 mg/l, pH 7.3-7.5, ammonia 0.02-0.033 and alkalinity 123-146 mg/l
- Pearl quality will be observed after final harvest

b. Valuka

- Among 400 operated mussels 12 % mortality was happened
- Recorded data of water quality parameters, temperature 18-26 ^{0}C, DO 4.77-5.90 mg/l, pH 7.14-7.77, ammonia 0.04 and alkalinity 130-190 mg/l
- Pearl quality will be observed after final harvest

c. Phulpur

- Among 400 operated mussels 15 % mortality was happened
- Recorded data of water quality parameters, Temperature 19-26.77 ^{0}C, DO 4.66-5.86 mg/l, pH 7.16-7.40, ammonia 0.02-0.04 and alkalinity 170-190 mg/l
- Pearl quality will be observed after final harvest

Improvement of Breeding and Culture Technique of Cuchia, *Monopterus Cuchia*

Researchers

Nur-A-Raushon, SSO
Saymuna Tarin Lupa, SO
Abul Bashar, SO

Objective

- To develop artificial breeding technique of Cuchia, *M. cuchia* using hormone
- To improve fry rearing technique of *M. cuchia* using different types of feed
- To disseminate control breeding and baby eel rearing technology of Cuchia, *M. cuchia*

Achievements

Experiment 1. Improvement of fry rearing technique of M. cuchia using different types of feed

An experiment was conducted to improve rearing technique of *M. cuchia* at the cistern (size. 2.76 m^2) complex of Freshwater Station, BFRI, Mymensingh during July to August. Nine cisterns were selected,

dried, and cleaned with lime at 250 kg/ha and then water was supplied from a deep tube well and filled up to the depth of 1 m. Baby eel or Cuchia fry were stocked at 50/m^2 in all Treatments. In Treatment 1, Cuchia fry were fed only commercial eel feed. In Treatment 2, fry was fed commercial eel feed (90%) comprising with earthworm (10%) where earthworms were used at 1 day interval. In Treatment 3, Cuchia fry were fed commercial eel feed (90%) and earthworm (10%) where earthworm was used at 2 days interval. Vermi or earthworms are being produced in the vermi compost unit. Earthworm were used as feed ingredients and applied 10 % of total feed utilization. The initial weight of baby eel in all Treatment was 4.0-5.0 g. After 30 days of rearing the final weight were 10.0-11.0 g in Treatment-1,14.0-15.5 g in Treatment- 2,12.0-13.5 g in Treatment- 3 where survival was 88.67%, 92.33% and 91.33%, respectively.

Table1. Effects of different types of feed on growth and survival of fry of *M. cuchia.*

Treatments	Initial Weight (g)	Final weight (g)	Survival (%)
T-1 (commercial eel)	4.0 -5.0	10.0 -11.0	88.67
T-2 {Com. eel feed (90%) +earthworm (10%)} 1 day interval		14.0 -15.5	92.33
T-2 {Com. eel feed (90%) +earthworm (10%)} 2-day interval		12.0 -13.50	91.33

Figure 1. Initial weight of Cuchia fry.

Figure 2. Final weight of Cuchia fry.

Experiment 2. Development of artificial breeding technique of Cuchia, M. cuchia using different types of hormones

The experiment was conducted in the cistern ecology during last week of April to Middle of June. So that, cistern was prepared with soil. During breeding season, broods were collected from natural sources. For the experiment, matured male and female broods were acclimatized for 3-4 days in cemented cisterns. The best broods of almost same size were selected based on visual examination of the secondary sexual characteristics i.e., abdomen and genital opening. Then the broods were treated with hormone in deep muscle at the dorsal side of the fish at different doses of PG, HCG and Ovatide. After hormone administration, the fish were stocked in cisterns for breeding. Detailed hormone doses and its response were given in following.

Trial-1.

Sex	Dose (PG mg/kg)	Response
Male	2.00	No response
Female	4.00	
Male	3.00	
Female	6.00	
Male	3.00	
Female	9.00	

Trial-2.

Sex	1st Dose (PG mg/kg)	Interval	2nd Dose (PG mg/kg)	Response
Male	0.00	6 hrs	2.00	No response
Female	6.00		8.0	
Male	0.00		2.00	
Female	4.5		7.5	

Trial- 3.

Sex	1st Dose (PG mg/kg)	Interval	2nd Dose (Ovatide ml/kg)	Response
Male	0.00	6 hrs	3.00	Most broods absorb their ova and sperm
Female	2.00		4.0	
Male	0.00	12 hrs	3.00	
Female	6.00		8.00	

Trial-4.

Sex	1st Dose (PG)	Interval	2nd Dose (HCG)	Interval	3rd Dose HCG/ PG	Response
Male	0.00	6 hrs	0.00	4 hrs	2 mg/kg	Broods showed mortality up to 50 to 75%
Female	4.00 mg/kg		250 IU/kg		500 IU/kg	
Male	0.00		0.00		4 mg/kg	
Female	6.00 mg/kg		750 IU/kg		1000 IU/kg	

Trial-5.

Sex	1st Dose (PG mg/kg)	Interval	2nd Dose (PG mg/kg)	Response
Male	0.00	6 hrs	12	Broods showed mortality up to 80%
Female	4		24	
Male	0.00	12 hrs	12	
Female	4		24	

Figure 3. Hormone application with different doses.

Expt. 3. Adoption on control breeding technique of *M. cuchia* in farmer's pond

Breeding technique of *M. cuchia* was demonstrated in farmer's pond at Tarakanda Upazila. Two ponds (size. 3 decimal) were selected for this validation trial. During pond preparation, 1.0-1.5 ft bottom soil was removed from both ponds and then filter net was placed on the bottom.

Figure 4. Farmer's pond preparation.

After setting the filter net, removed soil was further used on the net. Compost was used at one corner of the selected ponds. After supplying water, aquatic plants were provided to the ponds. In this demonstration program farmer's ponds were treated as Treatment 1 and 2. Brood Cuchia (150-250g) were stocked in last week of February/2022 at the stocking density of 30/decimal.

Figure 5. Stocking of broods in farmer's pond.

After stocking, supplementary, and live feed was applied at 2-3% of estimated body weight. Baby eels were produced in both ponds. The number of baby eel was 311 in pond 1 and 272 in pond 2. The detail result is shown in following.

Table 2. Production performances of baby eel in demonstration ponds.

Treatments	Production of baby eel (nos)	Feed
Pond -1	311	Fish paste and live spawn
Pond -2	272	

Figure 6. Sampling for baby eel production

Figure 7. Baby eel produced in farmer's pond

Sampling was done at monthly interval. The physico-chemical parameters of two demonstration ponds such as water temperature, pH, dissolved oxygen and total hardness are presented in Table 3.

Table 3. Different water quality parameters of demonstration ponds.

Parameters	Treatments	
	Pond -1	Pond -2
Temperature (^{0}C)	28.2 -34.2	28.0 -33.6
pH	6.77 -7.56	6.66 -7.62
DO (mg/l)	4.98 -7.88	4.8 -8.1
Total Ammonia (mg/l)	0.00 -0.01	0.00 -0.01

Experiment 4. Adoption on baby eel rearing technique of M. cuchia in farmer's cistern

This experiment was carried out in Tarakanda Upazila to demonstrate the rearing strategy of *M. cuchia* in a farmer's cistern (size. 2.5 m^2). This validation study wasconducted in two cisterns. Cisterns were cleansed with lime at a rate of 250 kg/ha before being fed with water and filled to a depth of 1 meter. Baby eels or Cuchia fry were stocked at a stocking density of 400-500/m^2. Cuchia fry were given live spawn as feed, accounting for 10% of total feed consumption.

Figure 8. Stocking of baby eel.

Figure 9. Harvesting baby eel from two cisterns.

The initial weight of baby eel was 0.24 g and final weight were 2.05 g and 1.98 g according to treatments. Survival rate was 65 and 63% in cistern 1 and 2. Details results are shown in the following Table.

Table 4. Growth performance of Baby eel in farmer's cistern.

Treatment	Initial weight (g)	Final weight (g)	Survival (%)
Cistern -1	0.24±0.03	2.05±0.21	65
Cistern -2		1.98±0.11	63

Improving Feed Formulation and Quality from Conventional and Non-Conventional Feed Ingredients Supplementation with Amino Acids for Commercially Important Fish Farming

Researcher
Dr. Md. Zulfikar Ali, CSO
Dr. Durin Akhter Jahan, PSO
Nur-a-Raushon, SO
Md. Hashibur Rahman, SO
Saymuna Tarin Lupa, SO

Objectives

- To optimize dietary protein to energy ratio (P/E ratio) for *Pangasianodon hypophthalmus*
- To evaluate the effect of supplementation limiting amino acids in the formulated diets for commercially important fish species
- To develop microalgae culture technique for using as fish feed ingredients
- To develop feed formulation and quality from conventional and non-conventional feed ingredients in this fish farming
- To recommend the potential limiting amino acids as feed additives in the formulated diets

Achievements

A series of feeding trials were conducted to develop quality feeds with supplementation of synthetic amino acids in plant protein-based diets for *P. hypophthalmus*. Two feeding trials were tested to investigate the optimum dietary protein to energy ratio (P/E ratio (trial-1) and supplementation of synthetic amino acids in plant protein-based diets (trial-2) of *P. hypophthalmus*. Experimentswere conducted in an indoor rearing system of Freshwater Station, BFRI, consisting of a series of cylindrical fiber glass tanks (70 L each) for 8 weeks. The follow up feeding trial in pond conditions were conducted to develop and optimize feeds with supplementation of synthetic amino acids in plant protein-based diets of *P. hypophthalmus* for 4 months (trial-4) is also progressing. Details of technical progress of the feeding trials are described below.

Expt. 1. Optimizing dietary protein to energy ratio (P/E ratio) in *P. hypophthalmus*

The feeding trial was carried out in a static indoor rearing system at Freshwater Station, BFRI, consisting a series of cylindrical fiber glass tanks (70 L each) for 8 weeks. Thirty Same aged and uniform size fingerlings of P. hypophthalmus were randomly distributed to 70 L fiberglass tank.Artificial aeration was used to maintain an adequate level of dissolved oxygen in each test tank for the study. The fish were individually weighed at the start of the experiment and then weekly. Fish sampling was done weekly to adjust the daily feed ration for the following week. At the beginning of the experiment, 15 fish were randomly sacrificed and kept for analysis of whole-body composition.Six experimental diets were formulated to contain two levels of protein (30 and 35%), each with three levels of lipid (5, 10 and 15%), to produce a range of protein to energy ratios. Fish meal and mustard oil cake were used as protein source. As

a lipid sources, soybean oil was used. Starch and wheat flour were used as sources of carbohydrate. Alpha cellulose was used as filler and 2% carboxymethyl cellulose was used as a binder. Vitamin and mineral premix were added 0.20%. Formulated diets and calculated proximate compositions are shown in Table 1. The bite-sized (1.0-2.0 mm) pellet feeds was made with the help of hand pellet machine. The fish were offered the test diets two times daily at the rate of 10-5% of their body weight and then divided into three equal meals at 9.30, 13.00 and 17.00 h. Feeding rate was adjusted based on weekly sampling weights of fish.

Standard methods were followed for the analysis of proximate composition of the dietary ingredients, experimental diets, and fish samples according to AOAC (2003).

Table 1. Formulation and proximate composition of the experimental diets (% dry matter basis) for *P. hypophthalmus*

	Dietary Treatments					
Diet no. (Protein/Lipid), (%)	**1 (30/5)**	**2 (30/10)**	**3 (30/15)**	**4 (35/5)**	**5 (35/10)**	**6 (35/15)**
Ingredients.						
Fish meal	22.00	22.00	22.00	25.00	25.00	25.00
Soybean meal	20.00	20.00	20.00	25.80	25.80	25.80
Mustard oil cake	18.00	18.00	18.00	20.00	20.00	20.00
Rice bran (auto)	20.00	21.00	21.00	20.00	20.00	20.00
Starch	14.80	11.80	6.80	7.00	5.80	1.00
Soybean oil	0.00	2.00	7.00	0.00	1.20	6.00
Alpha cellulose	3.00	3.00	3.00	0.00	0.00	0.00
Binder	2.00	2.00	2.00	2.00	2.00	2.00
Vit. and minerals premix	0.20	0.20	0.20	0.20	0.20	0.20
Proximate composition						
Crude protein	30.01	30.05	30.05	35.03	35.04	35.03
Crude lipid	5.50	10.02	15.05	5.80	10.15	15.00
Ash	6.80	6.97	6.97	7.74	7.87	7.87
NFE	39.49	37.04	32.04	34.29	33.09	28.29
GE (kJ/g)	17.00	17.43	18.54	17.70	17.97	19.04
P / E ratio	17.62	17.24	16.21	19.79	19.31	18.40

NFE = Nitrogen free extractives, calculated as 100 – (% protein + % Lipid + % Ash + % Fiber)

GE = Gross energy content

P / GE ratio = Protein to energy ratio in mg protein/ kJ-1 GE

Table 2. Mean growth performance and feed utilization of *P. hypophthalmus* fed various P/E ratio for 8 weeks.

Parameters	Dietary Treatments					
	1 (30/5)	2 (30/10)	3 (30/15)	4 (35/5)	5 (35/10)	6 (35/15)
Initial wt. (g)	1.45±0.05	1.45±0.05	1.45±0.05	1.45±0.05	1.45±0.05	1.45±0.05
Final wt. (g)	11.26±0.04	11.55±0.06	11.43±0.05	11.70±0.05	11.82±0.01	11.75±0.25
Weight gain	9.81±1.54	10.10±2.36	9.98±1.95	10.25±2.95	10.37±1.71	10.3±2.46
% Weight gain	676.55±2.54	696.55±1.63	688.28±3.05	706.90±2.15	715.17±1.67	710.34±1.84
Survival rate (%)	96.00±2.00	94.33±1.53	94.00±2.00	98.67±2.31	97.33±1.15	97.00±1.73
Food conversion ratio (FCR)	1.68	1.70	1.76	1.84	1.85	1.86
Protein efficiency ratio (PER)	1.98	1.95	1.96	1.55	1.53	1.54
Specific growth rate (SGR) (%/day)	3.42	3.46	3.44	3.48	3.50	3.49

Growth performances in terms of final body weight, % weight gain, specific growth rate (SGR, %/day) and protein utilization of fish fed the experimental diets were influenced by the levels of protein and energy as lipid (Table 2). Growth rates increased in response to higher dietary protein, but the highest dietary energy level in higher protein diet resulted in reduced weight gain (Table 2). Based on growth performance and protein utilization, it may be stated that the diet 2, containing 30% protein and 17.5 kJ/g gross energy performed best. This diet presumably contained the most appropriate P/E ratio 17.24 (16.62 mg protein/ kJ of GE) in *P. hypophthalmus*. However, the optimum dietary protein to energy ratio (P/E ratio) found for 17.24 mg protein/kJ of GE, for a diet containing crude protein 30%, crude lipid 10% and gross energy 17.50 kJ/g.

Expt.-2. Effects of supplementation of synthetic amino acids in plant protein based formulated diets in *P. hypophthalmus*

The follow up feeding trial was conducted with a series of cylindrical fiber glass tanks (70 L each) for 8 weeks at Freshwater Station, BFRI based on results from previous study (P/E ratio) in laboratory conditions. The same aged and uniform size fingerlings of *P. hypophthalmus* were randomly distributed into (groups of 30 fish) fiberglass tank and two replicate tanks were used for each test diet. Artificial aeration was used to maintain an adequate level of dissolved oxygen in each test tank. The fish were individually weighed at the start and end of the experiment by weekly. Sampling weight of fish was done to adjust the daily feed ration for the following week. Water quality parameter such as temperature, pH, dissolved oxygen and total ammonia were monitored through weekly sampling. At the beginning of the experiment, 15 fish were randomly sacrificed and kept for analysis of initial whole-body composition. At

the end of the feeding trial all fish were weighed, and survival rate were determined. Five fish were taken out from each tank for determination of whole-body carcass composition.

Five experimental diets (iso-nitrogenous and iso-energetic) were formulated to contain 30% crude protein and around 17.50 kJ/g of gross energy. Feeds were prepared using locally available fish feed ingredients such as fish meal, soybean meal, mustard oil cake and rice bran in different combinations. Fish meal, soybean meal and mustard oil cake were used as protein sources. Rice bran and starch was used as sources of carbohydrate. Alpha-cellulose was used as filler and carboxymethyl cellulose was used 2% as a binder. Vitamin and mineral premix were added 0.20%. The limiting amino acids. (i) Lysine and (ii) Methionine were added in the diets following the requirement of fish species. A control diet (diet-1, fish meal based) was prepared without adding amino acid, diets 2-3 were prepared fully and partially replace animal protein with plant protein without amino acids supplementation and diets 4-5 were prepared partially and fully replaced animal protein with plant protein and adding limiting amino acids of lysine and methionine as per requirement levels for pangus (Table 3). The bite-sized (1.0-2.0 mm) pellet feeds were made with the help of hand pellet machine. The pelleted feeds were sun-dried or dried an oven at 40° C for two days. Each dietary treatment was conducted in duplicate tanks. The fish were offered the experimental and control diets, 2 times daily at the rate of 10-5% of their body weight and feeding rate was adjusted based on weekly sampling (fish weighing) of fish.

Standard methods were followed for the analysis of proximate composition of the dietary ingredients, experimental diets, and fish samples according to AOAC (2003). Formulation and proximate composition of the experimental diets (% dry wt.) are shown in the following Table 3.

Table 3. Formulation and proximate composition of the experimental diets (%/drywt.) for *P. hypophthalmus.*

Diet no.	Dietary Treatments				
	Diet 1 (Control)	Diet 2	Diet 3	Diet 4	Diet 5
Ingredients					
Fish meal	22.00	11.00	0.00	11.00	0.00
Soybean meal	20.80	31.80	47.20	31.00	47.00
Mustard oil cake	20.00	20.00	20.00	20.00	20.00
Rice bran	11.00	29.00	24.00	30.00	25.00
Starch	16.50	5.00	5.35	3.45	2.50
Soybean oil	2.50	1.00	1.25	1.20	1.00
Alpha cellulose	5.00	0.00	0.00	0.00	0.00
Binder	2.00	2.00	2.00	2.00	2.00
Vitamin and Minerals	0.20	0.20	0.20	0.20	0.20
Lysine	0.00	0.00	0.00	0.80	1.60
Methionine	0.00	0.00	0.00	0.35	0.70
Proximate composition					
Crude protein	30.02	30.05	30.05	30.06	30.06
Crude lipid	10.00	10.07	10.10	10.15	10.00
Ash	6.34	7.48	6.92	7.46	7.00
Fiber	8.57	5.67	5.92	5.68	6.00
NFE	37.10	37.05	37.50	35.68	35.15
GE (kJ/g)	17.41	17.48	17.56	17.31	17.30

NFE = Nitrogen free extractives, calculated as 100 – (% protein + % Lipid + % Ash + % Fiber)
GE = Gross energy content

Table 4. Mean growth performance and feed utilization of *P. hypophthalmus* fed experimental diets for 56 days.

Components	Diet numbers				
	1 Control	2	3	4	5
Initial weight (g)	2.67±0.33	2.60±0.33	2.79±0.33	2.44±0.33	2.72±0.33
Final weight (g)	12.52±0.04	12.21±0.05	11.85±0.05	12.12±0.01	12.19±0.06
Weight gain (g)	9.85±0.36	9.61±0.54	9.06±0.38	9.68±0.71	9.47±0.31
% Weight gain	368.91±0.56	369.62±0.29	324.73±0.48	396.72±0.35	348.16±0.32
Survival rate (%)	96.00± 2.00	94.33±1.15	94.00±2.00	98.67±2.31	93.33±1.53
Food conversion ratio (FCR)	1.98	1.95	1.99	1.79	1.96
Protein efficiency ratio(PER)	1.69	1.71	1.67	1.86	1.70
Specific growth rate (SGR) (%/day)	2.58	2.58	2.41	2.67	2.50

Growth response parameters are shown in Table 4. The growth rate in terms of mean final body weight, percent weight gain of experimental fish fed diet 1 was higher than the other diets but there were no statistical different with diet 4. Fish fed diet 4 also showed good FCR value and better SGR with the control diet (Table 4).

Expt. 3. Development of microalgae culture technique for using as fish feed Ingredients

The present study was undertaken to investigate the growth performance of *S. platensis* in various concentrations of papaya skin powder medium. The experiment was conducted for 90 days from January to March 2022 in the laboratory of Nutrition Division of Bangladesh Fisheries Research Institute, Freshwater Station, Mymensingh.The two types of media *viz*. papaya skin powder medium (PSPM) and kosaric medium (KM) were used for the culture of S. platensis. For PSPM, 3 Treatments each of 3 replications and for KM 1 Treatment with 3 replications were used. Culture period for both the media 12 days was maintained. Papaya skin was collected and dried in an oven at 50°C for overnight. For complete drying, these papaya skins were dried under sun for another seven days. Blender was used to make powder of the dried papaya skin. For getting very fine particle of papaya skin, it was sieved through a

siever. Then it was kept in a 5-liter capacity reagent bottle and added 4 liter distilled water. Aeration was provided for 22 days for preparing papaya skin powder medium (PSPM). About 600 ml distilled water was added into the conical flask having capacity 1.0 L volume and then prepared papaya skin powder medium was added 15, 20 and 25% each with three replications and then sterilized at 120°C for 15 minutes with moist heat autoclave. After cooling of the prepared media *S. platensis* were inoculated and mixed well gently. The KM was prepared by adding required amount of different chemical ingredients with distilled water. After mixing, sterilization and cooling *S. platensis* were inoculated in the prepared media and then mixed well gently. These cultured bottles were continuously aerated using electric aerator. Samplings were taken at every alternative day from each bottle to observe *S. platensis* cell density, different water quality parameters of culture media. The pure stock of *S. platensis* was maintained in the laboratory in kosaric medium (KM) and growth was monitored then checked under microscope to check the purity. *S. platensis* sample from each Treatment was taken and filtered with an electric filtration unit using filter papers and shifted to the oven at 105°C for 24 hours and then transferred to the desiccator for cooling. Proximate composition of *S. platensis* was done and shown in Table 5.

Table 5. Proximate composition of *S. platensis* (% dry matter basis).

Treatments	Proximate composition			
	Moisture (%)	Protein (%)	Lipid (%)	Ash (%)
T-1.(15% PSPM)	13.09	42.0	7.00	14.84
T-2.(20% PSPM)	13.35	43.0	7.15	15.05
T-3.(25% PSPM)	13.92	47.0	7.36	15.49
T-4. (KM)	14.01	50.0	7.50	15.95

The physico-chemical parameters *i.e.* temperature were ranged 24.00-31.8°C, pH. 9.3-9.53, dissolved oxygen (DO). 4.27-6.58 mg/l, measuring the voltage between a pH sensitive glass electrode (MVPH). 128.70-149.47, total dissolved solid (TDS). 1026.00-3875.00, electric conductivity (EC). 1501.00-2576.00, hectopascal pressure unit (hpa%). 1012.00-1476.00 and salinity. 0.60-4.32 were recorded. Details results are shown in Table 6.

Table 6. Water quality parameters of different Treatments during experimental period.

Observed parameters	Treatments			
	T-1 (15% PSPM)	T-2 (20% PSPM)	T-3 (25% PSPM)	T-4 (KM)
pH	9.30±0.05	9.44±0.02	9.42±0.01	9.42±0.13
Temperature	24.00±0.08	29.39±0.37	28.47±0.40	28.16±0.84
DO	4.27±0.10	5.86±0.22	6.47±0.35	6.58±2.03
MVPH	128.70±4.62	143.18±11.59	149.47±0.43	147.13 ±5.75
TDS	1070.67 ±72.60	1101.00 ±90.80	1026.00 ±103.11	3875.00±1554.52
EC	1501.17±146.37	2218±208.88	2011.33±193.41	2576.12±515.89
hpa%	1012.00±0.50	1014.33±1.04	1018.17±1.26	1017.33±2.08
Salinity	0.60 ±0.08	0.88 ±0.32	0.64 ±0.20	4.32±1.86

The growth of cells was varied in KM and different concentrations of PSPM were also found. The growth rate of *S. platensis* was higher in KM than various concentrations of PSPM. Among the 3 concentrations of PSPM, the growth rate was higher in the concentration of 25% PSPM. Higher growth of cells was found due to the favorable water quality parameter and suitable amount of nutrients. In different treatments growth of *S. platensis* were observed and showed in Table 7.

Table 7. Cell weight of S. platensis in different concentrations of PSPM and KM

Treatments	Initial weight (g/l)	Final weight (g/l)
T-1. (15% PSPM)	0.041±0.008	0.498^{c}±0.02
T-2. (20% PSPM)		0.569^{b}±0.01
T-3. (25% PSPM)		0.680^{a}±0.01
T-4. (KM)		0.714^{a}±0.01

Expt. 4. Development of quality feeds with supplementation of synthetic amino acids in plantprotein based formulated diets in *P. hypophthalmus*

The feeding trial was carried out in the pond complex of Freshwater Station, Bangladesh Fisheries Research Institute (BFRI), Mymensingh for a period of 8 weeks. The experiment is being carried out. Three experimental diets were formulated to contain 30% crude protein. Fish meal, mustard oil cake, soybean meal, wheat flour, vitamin and mineral premix were used as diet ingredients. Limiting amino acids. Lysine and Methionine was added in the diets and a control diet (diet-1; fish meal based) was prepared without adding amino acid. Diet 2 was prepared partially replace animal protein with plant protein and Diet 3 was prepared fully plant protein based with adding limiting amino acids lysine and methionine. Pellet feeds (1.0-2.0 mm) made with hand pellet machine. The fish was offered the test diets two times daily at the rate of 10-3%. During experimental period water quality parameter and whole-body composition was observed. Diets formulation and growth performance are shown the following Table 8 and 9, respectively. Growth response was observed monthly for the feed adjustment.

Table 8. Formulation and proximate composition of the diets (% dry weight basis) for *P. hypophthalmus.*

Diet no .	Diet number		
	1	2	3
Ingredients .			
Fish meal	19.00	12.00	0.00
Soybean meal	22.80	30.00	40.00
Mustard oil cake	18.00	20.00	27.00
Rice bran	32.00	28.60	22.40
Wheat flour	6.00	6.00	6.00
Binder	2.00	2.00	2.00
Vitamin and mineral premix	0.20	0.20	0.20
Lysine	0.00	0.80	1.60
Methionine	0.00	0.40	0.80
Proximate composition			
Crude protein	30.08	30.04	30.15
Crude lipid	9.75	9.70	9.50
Ash	8.43	8.05	7.88
Fiber	5.84	5.96	5.56
NFE	37.00	36.50	36.06
GE (kJ/g)	17.04	17.14	17.20

Table 9. Mean growth performance and feed utilization of *P. hypophthalmus* fed experimental diets in ponds for 8 weeks.

Components	Diets number		
	1	2	3
Initial weight (g)	74.95±14.34	73.19±13.21	75.38±10.76
Final weight (g)	283.33±15.34	281.00±10.76	274.67±13.21
Weight gain (g)	208.38±1.00	207.81±2.55	199.29±2.41

From the results of this feeding trial, it is logical to conclude that partially replaced animal protein with plant protein with adding limiting amino acids lysine and methionine can be used as a fish feed additive in *P. hypophthalmus* culture, to enhance better feed efficiency and growth performance.

Culture of Indigenous Small Fish in Biofloc Aquaculture System (Comp. A)

Researchers
Md. Ashikur Rahman, SO
Shayla Sultana Mely, SO

Objectives

- To optimize the stocking density of Shing, *Heteropneusteus fossilis*; Pabda, *Ompok pabda*; Gulsha, *Mystus cavasius* in Biofloc system
- To evaluate the growth and production of Prawn, M. rosenbergii in Biofloc system
- To evaluate the growth and production of Magur, Clarias batrachus in Biofloc system (2021-2022)
- To analyse the economic benefits of Biofloc system

Achievement

Growth and production of Magur, *Clarias batrachus* in Biofloc system (2021-2022)

Tank facilities and experimental design

The experiment was carried out in plastic indoor tanks with volume of 8000 L where water capacity is 6000 L for the period of 6 months culture period. The design of the experiment has been presented in Table 1.

Table 1. Design of Experiment.

Treatments	Shing (Individual/ 6000 L)
T_1	1200
T_2	1500
T_3	1800

Fish stocking and tank management

Mixed sex of Magur was stocked according to design of the experiment as presented in the above Table. After stocking of fish, commercial feed (30% crude protein,) was applied twice a day up to satiation level. For maintaining C/N ratio, probiotics and Molasses were added to tank.

Assessment of water quality parameters

The temperature in the tanks during the growth test were monitored daily by a digital thermometer. Other water quality parameters including pH, dissolved oxygen (DO), total ammonia nitrogen (TAN), were measured twice at two days' intervals using the spectrophotometer HACH DR1900. Total Dissolved solid (TDS) were measured by HANNA digital meter. Floc density was measured by Emhoff-cone.

Table 2. water quality parameter of different treatment.

Treatments	Temp (^{0}C)	DO (mg/l)	pH	TAN (mg/l)	TDS (mg/l)	Flock density (ml/l)
T_1	25.7±1.4	5.2±1.1	7.40±0.4	0.15±0.01	340±32.5	22±1.2
T_2	25.9±1.1	5.3±1.2	7.00±0.5	0.18±0.05	315±22.7	21±3.1
T_3	25.3±2.1	5.6±1.5	7.54±0.3	0.26±0.06	370±41.9	23±1.1

The water temperature varied between 23 and 27.5°C during the experiment and there was no significant difference among the treatments. The dissolved oxygen ranged between 4-6 mg/lin all treatments whereas the value of pH was 7-8. The other parameter such as TAN, TDS were found suitable for fish culture. Throughout the experiment flock density ranged between 20-24 ml/l.

Harvesting of fish

After Seven months of rearing, fish were harvested from the tank by draining out of water. After harvesting, comprehensive data were recorded (Table 3). Then growth, survival and production were estimated. Cost benefit analysis was done to know the economic viability of biofloc system. Moreover, which species is more suitable for biofloc system that information was also generated. The growth parameters such as initial weight, final weight, survival rate (%), FCR, feed requirement and electricity requirement were observed. The results are presented in Tables 3.

Table 3. Production performance of Magur (*Clarias batrachus*) under different treatment.

Parameter	T_1	T_2	T_3
Stocking density	1200	1500	1800
Initial weight (g)	5.15	5.15	5.16
Final weight (g)	150.0	125.0	115.00
No. of Fish harvested	1032	1245	1440
Survivability	86.50	83.50	80
FCR	1.2	1.20	1.15
Feed required (kg)	185	187	190
Electricity requirement	43.20 KW/Month	43.20 KW/Month	43.20 KW/Month

The FCR value was the highest in T_1 and T_2 (1.2) and the lowest in T_3 (1.15). Analytical results showed, the FCR values were directly related to the stocking density. The survival rate was estimated after harvesting the experimental fish at the end of the study. For current experiment, the values of survival rate were 86.50, 83.50 and 80 % in T_1, T_2 and T_3 respectively. Same amount of electricity is consumed for all treatments.

Economic analysis

Economic analysis was performed to estimate the benefit cost ratio from different treatments of the Magur in Biofloc system (Table 4). Several variable costs like stocking cost, chemical cost, labour and electricity cost were estimated during the period of farming. There were some fixed costs such as land rent, tank preparation as well. Cost and return analysis were performed on both variable and total cost basis. To achieve the objectives of the study a simple tabular analysis was done (Table 4). The benefit cost ratios were estimated as 1.28, 1.14 and 1.04 for T_1, T_2 and T_3, respectively.

Table 4. Economic analysis of the Magur at Biofloc system system.

Inputs	T_1		T_2		T_3	
	Qty.	Cost (Tk.)	Qty.	Cost (Tk.)	Qty.	Cost (Tk
Fingerling (Nos.)	1200 @ 1.5	1800	1500@ 1.5	2250	1800@ 1.	2700
Feed (kg)	185 @ 75/-	12950	187@ 75/-	14025	190@ 75/	14250
Molasses (kg)	90 @ 35/-	3150	95 @ 35/-	3325	95 @ 35/	3325
Probiotics (kg)	5	5000	5.5	5500	6	6000
Electricity	302.4 KW @ 11/-	3327	302.4 KW @ 11/-	3327	302.4 KV @ 11/-	3327
Others cost (labour, harvesting, depreciation etc.)		5000		5000		5000
Total cost		31227		33427		34602
Sell price of fish	154 kg @ 260	40040	155.6 @ 245	38122	165 @ 22	36300
Net benefit/6000 liter	8813		4695		1698	
BCR	1.28		1.14		1.04	

Optimization of the stocking density of Shing, *Heteropneusteus fossilis* in biofloc System

Growth performance

Table 1 shows the sampling weight of Shing under different Treatments after six months of rearing. Based on sampling weight, Treatment 1 showed the best results in terms of growth. Where, Treatment 3 attained the lowest growth in biofloc system. The average sampling weight of Treatments-1, 2 and 3 were 38±1.2. 33±2.1 and 31±2.5 g, respectively. Feed conversion ratio indicating the effectiveness of feed management and economic performance in aquaculture. In this experiment the best FCR comes from Treatment 1 and Treatment 3.

Table 5. Production performance of Shing under different treatment.

Parameter	T_1	T_2	T_3
Stocking density	4500	6000	7500
Initial Wt. (g)	3.0 ±0.4	3.3 ±0.5	3.2 ±0.48
Harvesting Wt. (g)	38±1.2 (158 kg)	33±2.1 (165 kg)	31±2.5 (169 kg)
No. of Fish harvested	4100	4910	5450
Survivability	91.11	81.83	72.66
FCR	1.01	1.03	1.01
Feed required (kg)	160	170	172
Electricity requirement	43.20 KW/Month	43.20 KW/Month	43.20 KW/Month

Several variable costs like stocking cost, chemical cost, labour and electricity cost were estimated during the period of farming. There were some fixed costs such as land rent, tank preparation as well. Cost and return analysis were performed on both variable and total cost. To achieve the objectives of the study a simple tabular analysis was done (Table 6). The benefit cost ratios were estimated as 1.35, 1.22 and 1.10 for T_1, T_2 and T_3, respectively.

Table 6. BCR analysis for Shing under different Treatments.

Inputs	T_1		T_2		T_3	
	Qty.	Cost (Tk.)	Qty.	Cost (Tk.)	Qty.	Cost (Tk.)
Fingerling (Nos.)	4500	6750	6000	9000	7500	11200
Feed (kg)	160kg kg@62	9920	110 kg	10230	172 kg	10664
Molasses	70kg@35	2100	75kg	2625	80 kg	2800
Probiotics	4.0 kg	4800	4.5 kg	5400	5.1kg	6120
Electricity	173KW@11	1093	173KW@11	1093	173KW@11	1093
Others cost (labour, harvesting, depreciation etc.)	-	4000	-	4000		4000
Total cost	-	28663	-	32348	-	35877
Sell price of fish	158kg @ 245 tk/kg	38710	165 kg @ 240 tk/kg	39600	169 kg @ 235 tk/kg	39715
Net benefit/6000 liter	9,257		7,252		3,838	
BCR	1.35		1.22		1.10	

Water Quality Parameter

The results of water quality parameters such as water temperature, disso lved oxygen (DO), pH, total ammonia nitrogen (TAN) and total dissolved solids (TDS) are shown in table7. The values indicate that all the observed water quality parameters were congenial for fish growth.

Table 7. Water quality parameters of different tanks in Biofloc System.

Parameters	T_1		T_2		T_3	
	Tank 1	Tank 2	Tank 3	Tank 4	Tank 5	Tank 6
W. Temp (^{0}C)	25.2±1.2	29.1±3.0	28.7±1.5	29.6±0.3	30.8±0.6	29.6±0.4
DO (mg/l)	5±1.2	5.5±0.5	5.12±0.3	5.30±0.5	4.5±0.6	4.33±.1
pH	7.55±0.3	7.67±0.2	7.91±0.6	7.71±0.4	7.73±0.2	7.70±0.0
TAN (mg/l)	0.12±0.1	0.11±0.1	0.14±0.1	0.01±0.01	0.15±0.01	0.10±0.09
TDS (mg/l)	255±11.7	274±11.2	258±27.8	266±11.8	291±32.0	290±21.1

Identification of etiological agents responsible for fish diseases using PCR techniques and mitigation measures

Researchers
Dr. Md. Shirajum Monir, SSO
Dr. Jonaira Rashid, SSO
Md. Ashikur Rahman, SO
Maria Al Mazed, SO
Farjana Jannat Akhi, SO

Objectives

- To isolate and identify the causal agents responsible for fish diseases with special references to Pabda and Gulsha
- To detect the etiological agents based on PCR techniques
- To evaluate the antibiotic resistivity of isolated pathogens
- To find out the protective measures against diseases

Achievements

1. Clinical and post-mortem findings

The mortality recorded in the infected Gulsha farms is up to 60 to 90% acute, and numerous fish died within two weeks. Around 95% infected fish showed gross lesions that were associated with motile septicemia, including necrosis and haemorrhages of fins and tail (Figure 1a and b).Congestion and enlargement with haemorrhage was also observed in liver, kidney and spleen of the diseased Gulsha (Figure 1c and d).

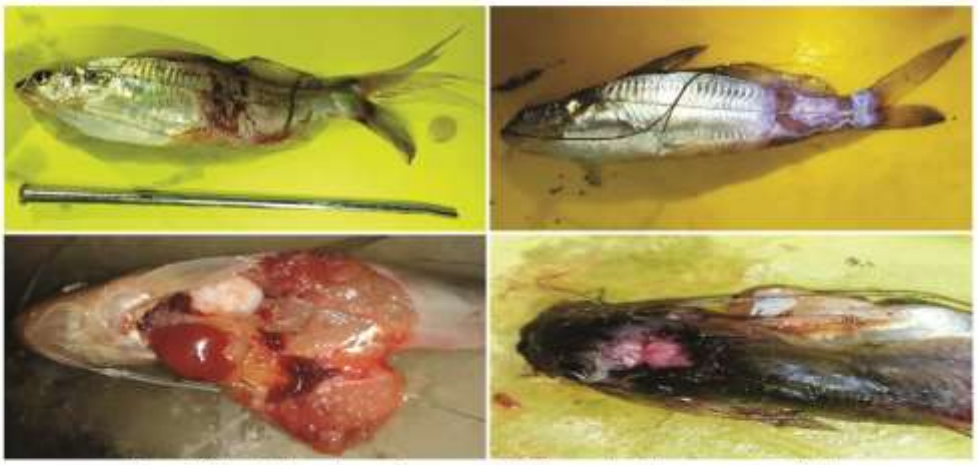


Figure 1. Gross lesions observed in necropsied with suspected *Aeromonas* spp. infection. Infected Gulsha with body and fin erosions

2. Biochemical and molecular tests for the isolated bacterial from infected Gulsha

Bacteria of the Aeromonas spp. were identified from the lesion (skin and fin), liver, kidney and brain of the diseased Gulsha(Table 1). The isolated Aeromonas spp. (strains) showed motile and oxidase (2a and b), O/F, VP, catalase, indole, H_2S production, nitrate reduction test were positive whereas MR and urease production test were negative. While the other isolated strains showed oxidative Gram-negative rod and indole, H2S production, nitrate reduction, urease production test negative but O/129 test was sensitive.

Table 1. Biochemical characteristics of the isolated bacteria from infected Gulsha.

Test name	Isolated bacteria											
	SK 01-22	*Sk 02-22*	*Sk 03-22*	*Li 04-22*	*Li 05-22*	*Kd 06-22*	*Kd 07-22*	*Kd 08-22*	*Br 09-22*	*Br 10-22*	*Sp 11-22*	*Br 14-22*
Gram staining	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Motility	+	+	+	+	-	+	+	+	-	+	-	+
Oxidase test	+	+	+	-	+	+	+	-	+	+	+	+
O-F test	+	+	+	+	O	+	+	+	+	O	+	+
MR test	-	-	-	+	-	-	-	-	+	-	-	-
VP test	+	+	-	+	+	+	+	+	-	+	+	+
Catalase test	+	+	+	+	+	-	+	+	+	-	+	+
Indole test	+	+	+	+	+	+	+	+	+	+	+	+
H_2S production	+	+	+	+	+	+	+	+	+	+	+	+
Nitrate reduction	+	+	-	+	+	+	+	d	+	+	d	+
Urease production	-	-	+	-	-	d	-	-	-	-	-	-
TSI test	A/A	A/A	K/A	A/A	K/N	A/A	A/A	A/A	A/K	A/A	A/d	A/A
Production of acid from												
Glucose	+	+	-	+	+	+	-	+	+	+	d	+
Galactose	+	+	d	d	+	-	d	d	+	+	+	d
0/129 test (10 µg and 150 µg)	R	R	R	R	R	R	R	R	R	R	S	S

Note. d= variable reaction, O= oxidative, A= acid, K= alkaline, R= resistant, S= sensitive

Molecular detection was conducted by PCR to identify isolates where positive bands were found at 1500 bp, and the presence of *Aeromonas* spp. were identified by BLAST analyses of the 16S rRNA sequences of the PCR products.

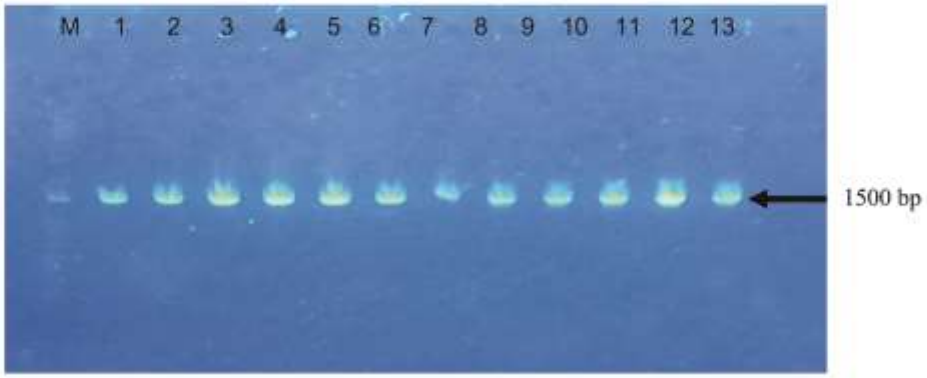


Figure 2. Agarose gel electrophoresis of PCR amplification generated by isolated bacteria at ⊠1500 bp. Lanes. (M) 1 kbp DNA marker; (1-12) positive samples from field; (13) negative control.

3. Bacteria isolated from different organs of diseased Gulsha

A total of 65 bacterial strains were isolated from 90 samples of diseased Gulsha. The species composition and sources of these strains are presented in Table 1. The isolation frequencies of these 65 strains upon anatomical parts of infected Gulsha were lesion (skin and fin) 20 (30.77%), liver 16 (24.61), kidney 14 (21.53%) and brain 15 (23.07%).

Table 2. Bacteria isolated from different organs of infected Gulsha.

Isolated bacterial strains	Distribution (Number and %) of different bacterial stains (n=50) according to site of isolation				Total
	Anatomical parts of infected Gulsha				
	Infected area (skin and fin)	Liver	Kidney	Brain	
SK 04-22	8 (12.31)	4 (6.15)	2 (3.07)	4(6.15)	18 (27.69)
SP 06-22	4(6.15)	1 (1.54)	3 (4.62)	2 (3.07)	10 (15.38)
SD 09-22	2(3.08)	3 (4.62)	2 (3.07)	3 (4.62)	10 (15.38)
RP 05-22	3(4.62)	2 (3.07)	3 (4.62)	1 (1.54)	9 (13.84)
SW 07-22	1(1.54)	2 (3.07)	3 (4.62)	2 (3.07)	8 (12.30)
CP 07-22	1(1.54)	2 (3.07)	0	2 (3.07)	5 (7.69)
AK 06-22	1(1.54)	2 (3.07)	1 (1.54)	1 (1.53)	5 (7.69)
Total	**20 (30.77)**	**16 (24.61)**	**14 (21.53)**	**15 (23.07)**	**65 (100)**

3. Susceptibility to antimicrobial agents in-vitro condition

Ten different antibiotic dices namely, cefradine, chlortetracycline, ciprofloxacin, doxycycline, erythromycin, gentamicin, levofloxacin, penicillin, oxytetracycline and streptomycin were tested against 15 different isolated bacterial strains. It was observed that all the isolated bacteria were mostly sensitive to cefradine and ciprofloxacin (Table 3). Cefradine was highly effective against Sp 13-22 and Br 14-22 bacterial strains, while erythromycin andgentamicin showed moderate effect against all the isolated bacteria. Penicillin did not show any effect against all bacteria.

Table 3. Antibiotics sensitivity test on isolated bacteria from infected Gulsha

Isolated bacterial strains	Antibiotic sensitivity									
	Oxy	Chl	Cef	Cip	Lev	Step	Dox	Gen	Ery	Pen
SK 01-22	+	+	+++	+++	+	+	+	++	++	-
Sk 02-22	+	+	+	+++	++	-	++	++	++	-
Sk 03-22	-	-	++	-	++	-	++	++	-	-
Li 04-22	+	+	++	++	++	-	++	++	+	-
Li 05-22	+	+	++	+++	++	+	++	+	-	-
Kd 06-22	-	++	++	+++	-	-	-	+	+++	-
Kd 07-22	++	+	+	+	++	-	-	-	++	-
Kd 08-22	+	+	+++	+	-	-	-	++	-	-
Br 09-22	+	+	+++	+++	+	-	+	++	+	-
Br 10-22	-	+	-	+	-	+	-	+	+++	-
Sp 11-22	++	-	+	+	++	-	++	+	++	-
Sp 13-22	-	+	+++	+++	++	-	++	++	++	-
Br 14-22	-	+	+++	+++	++	-	-	++	+	-
Sk 15-22	+	+	+	+++	++	-	++	++	++	-
Sk 16-22	+	+	+	-	-	-	++	-	+++	-

Note. no inhibition, +. inhibitory zone between 5-12mm, ++. inhibitory zone between 13- 20mm. +++. inhibitory zone between 21-30mm above

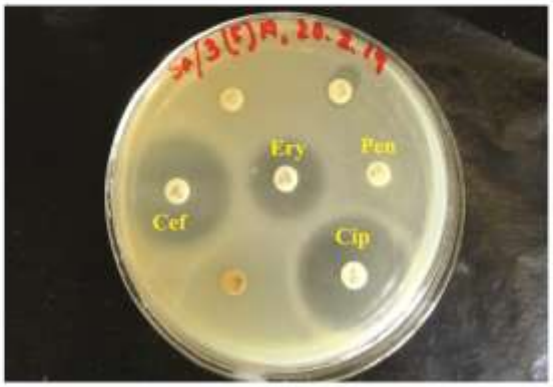


Figure 2. Figure 3.Antibiotic sensitivity and resistant pattern of bacteria isolated from infected Gulsha.

4. Virulent test of the isolated bacteria

To ascertain which isolates to select for vaccine development, the virulence of a selection of the isolates bacterial strains were assessed following the intraperitoneal (ip.) infection challenges. Forty out of sixty-five isolates with a history of causing severe mortality in farmed fish (higher than 80%) were selected as virulent isolates and five isolates eliciting low mortality (less than 20%) was selected as an avirulent isolates.

Ecological assessment of inland open water fisheries population with bio-physicochemical properties to frame EBFM approach (Comp-A)

Researchers
Dr. Ehsanul Karim, SSO
Md. Nahiduzzaman, SO
Nazia Naheen Nisheeth, SO

Objectives

- To estimate population ecology and diet composition of available inland (haorand beel) open water fish
- To assess bio-physicochemical properties of inland waterbodies with the seasonal variation
- To assess stock of available major fish groups based on catch and CPUE data
- To assist for framing or formulating ecosystem based management approach for inland open waters with emphasizing to increase productivity and conservation of fisheries resources

Achievements

Selected study areas. Dingapota haor, Mohoganj, Netrokona, Charigram haor, Charigram, Kishoreganj, Tanguar haor, Sunamgonj

Dingapota Haor Charigram Haor Tanguar Haor

Figure 1. Map of the study areas.

Study 1.Sampling of Bio-Physicochemical properties of inland open watersto collect hydrological data

Collection of hydrological data

Monthly monitoring of water quality parameters (temperature, water depth, transparency, dissolved oxygen, pH, alkalinity and ammonia-nitrogen) has been conducted. In each sampling water temperature (°C), pH, DO, TDS was recorded by using a high equipped Multi-Parameter Water Quality Meter. Water depth was measured with a measuring tape. Water transparency (cm) was measured by a Secchi disk. Alkalinity (mg/l) and ammonia-nitrogen (mg/l) were determined with the help of a spectrophotometer DR-1900 at water quality and pond dynamics laboratory.

Assessment of plankton community dynamics

Ten liters of water samples was collected from five different places of each plot and passed through plankton net (mesh size 20 µm). The concentrated samples were transferred to a measuring cylinder and made up to 50 ml with distilled water. Samples were preserved in small plastic bottles with 10% buffered formalin. Plankton numbers were estimated using a Sedgwick-Rafter counting cell (S-R cell) under a binocular microscope (Optika, B-190/B-290) following APHA, 1992. Identification of plankton to genus level was carried out using the keys from Ward and Whipple (1959), Prescott (1962) and Bellinger (1992). The quantitative estimation was done following the same procedure mentioned in Azim et al. (2001) and the estimation will be made by using the following formula.

$N = (P \times C \times 100)/L$

In which,
N= Number of plankton cells or units per liter of original water
P = Total number of plankton counted in 10 fields
C= Volume of final concentrated sample (ml)
L= Volume of original water (l)

Statistical analysis

Collected data were analyzed into a database system with using the programs. Microsoft Excel (MS Excel,) PAST 4 (Paleontological Statistics), Computer software SPSS (version 20) was used.

Results

Water quality parameters of the studied haors during study period

Water quality parameters observed and recorded during the study were summarized in Table 1. Water quality parameters measured for the three haors and different seasons. Water temperature varied from 29.82±0.56°C (Monsoon) to 27.65±1.63°C (pre-monsoon), depth from 6.49±1.43 m (Monsoon) to 1.86±1.01 m (pre-monsoon), water transparency was 22.76±1.88 cm (Monsoon) to 37.40±5.05 cm (pre-monsoon), DO was 6.38±0.96 mg/l (monsoon) to 5.21±0.79mg/l (pre-monsoon) and water pH was 7.56±0.86 (post-monsoon) 6.70±0.65 (pre-monsoon). During monsoon air temperature remain higher than other season. On the other hand, due to heavy rainfall and mixing of runoff water, transparency was very low in monsoon compared with other season and attributedto lower DO level. However, by the location, the highest and lowest temperature was observed (26.83±3.86 and 26.57±4.01 at Tanguar and Dingapota haors). Constant increase and decrease in water depth were observed during the study period with a higher water depth was recorded at Tanguar during and lower at Dingapota. Dissolved oxygen, pH and alkalinity were also found the highest at Tanguar and lower at Dingapota.

Table 1. Water quality parameters of studied seasons.

Seasons	Temp. (°C)	Transparency (cm)	Depth (m)	pH	DO (mg/l)	NO_3-N (mg/l)	PO_4-P (mg/l)	Total alkalinity (mg/l)	TDS (mg/l)
Monsoon	29.82 ±0.56	22.76± 1.88	6.49 ±1.43	7.80 ±0.97	6.38 ±0.96	0.17 ±0.04	1.24 ±0.14	106.75 ±4.83	105.57 ±7.59
Post-monsoon	22.58 ±3.94	34.76± 4.71	2.56 ±0.85	7.56 ±0.86	7.52 ±1.29	0.30 ±0.13	1.90 ±0.64	119.91 ±5.85	125.27 ±8.47
Pre-monsoon	27.65 ±1.63	37.40± 5.05	1.86 ±1.01	6.70 ±0.65	5.21 ±0.79	0.39 ±0.08	1.61 ±0.56	133.41 ±8.14	141.71 ±6.09

Table 2. Water quality parameters of studied areas.

Locations	Temp. (°C)	Transparency (cm)	Depth (m)	pH	DO (mg/l)	NO_3-N (mg/l)	PO_4-P (mg/l)	Total alkalinity (mg/l)	TDS (mg/l)
Netrokona (Mohanganj)	26.57 ±4.01	34.09± 7.44	3.18 ± 2.24	6.79 ±0.80	5.64 ±1.04	0.22 ±0.11	1.26 ±0.11	114.35 ±10.97	117.88 ± 15.49
Kishoreganj (Charigram)	26.65 ±3.91	32.37±7.61	3.68 ±2.35	7.37 ±1.01	6.06 ±1.05	0.28 ±0.12	1.62 ±0.54	121.23 ±12.00	124.87 ±17.26
Tanguar	26.83 ±3.86	28.47±6.63	4.04 ±2.34	7.90 ±0.71	7.42 ±1.41	0.35 ±0.13	1.86 ±0.69	124.49 ±12.80	129.80 ±14.74

Plankton analysis

A total of 32 phytoplankton species were identified under 4 classes. A total of 14 genera of Chlorophyceae, 10 genera of Bacillariaophyceae, 5 genera of Cyanophyceae and 3 genera of Euglenophyceae were listed from the study area (Table 3).

A total of 3 groups of zooplankton were identified i.e. Rotifera, Cladocera and Copepoda in studied haors. A total of 5 genera of Rotifera, 4 genera of Copepoda and 4 genera of Cladocera were identified during the study period in the studied haors (Table 2). Analyzed plankton groups with their abundance value in different sampling seasons at the three studied haors are shown in Table 3. Chlorophyceae was the dominant among the four phytoplankton groups and its higher abundance was recorded during monsoon in all the studied locations. Phytoplankton groups were arranged in order of Chlorophyceae > Bacillariophyceae> Cyanophyceae > Euglenophyceae in all the studied locations. Total cell density of phytoplankton was the highest in monsoon and the lowest in pre-monsoon in all the studied locations (Figure 2). Among the zooplankton, Rotifera was the most dominant followed by Cladocera and Copepoda. The abundance of Rotifera was the highest during post-monsoon and Copepoda was the lowest during the study period. Total cell density of zooplankton was the highest in post-monsoon and the lowest in monsoon in all the studied locations (Figure 3).

Table 3. Check list of plankton of the studied areas during study period.

Plankton	Group	Genera	Dingapota	Charigram	Tanguar
Phytoplankton	Chlorophyceae	*Actina strum*	+	+	+
		Ankistrodesmus	+	+	–
		Chlorella	+	+	+
		Closterium	+	–	+
		Coleochaete	–	–	+
		Cosmarium	+	+	+
		Melosira	+	+	+
		Microspora	+	+	+
		Pediastrum	–	+	+
		Spirogyra	+	+	+
		Spirulina	+	+	+
		Scenedesmus	+	+	–
		Ulothrix	+	+	+
		Volvox	+	+	+
	Bacillariaophyceae	*Asterionella*	+	+	+
		Bacillaria	+	+	+
		Chaetoceros	–	–	+
		Cyclotella	+	+	+
		Fragilaria	+	+	+
		Gyrosigma	+	+	+
		Navicula	+	+	+
		Nitzschia	+	+	+
		Rhizosolenia	+	+	+
		Synedra	+	+	+
	Cyanophyceae	*Anabaena*	+	+	+
		Apanizomenon	+	+	+
		Chroo coccus	–	+	+
		Microcystis	+	+	+
		Oscillatoria	+	+	+
	Euglenophyceae	*Euglena*	+	+	+
		Phacus	+	+	+
		Trachelomonas	–	+	+
Zooplankton	Rotifera	*Asplanchna*	+	+	+
		Brachionus	+	+	+
		Filinia	+	–	+
		Keratella	+	+	+
		Polyarthra	–	+	+
	Cladocer a	*Bosmina*	–	+	+
		Daphnia	+	+	+
		Diaphanosoma	+	+	+
		Moina	+	+	+
	Copepoda	*Cyclops*	+	+	+
		Diaptomus	+	+	+
		Macrocyclops	+	–	+
		Mesocyclops	–	+	–

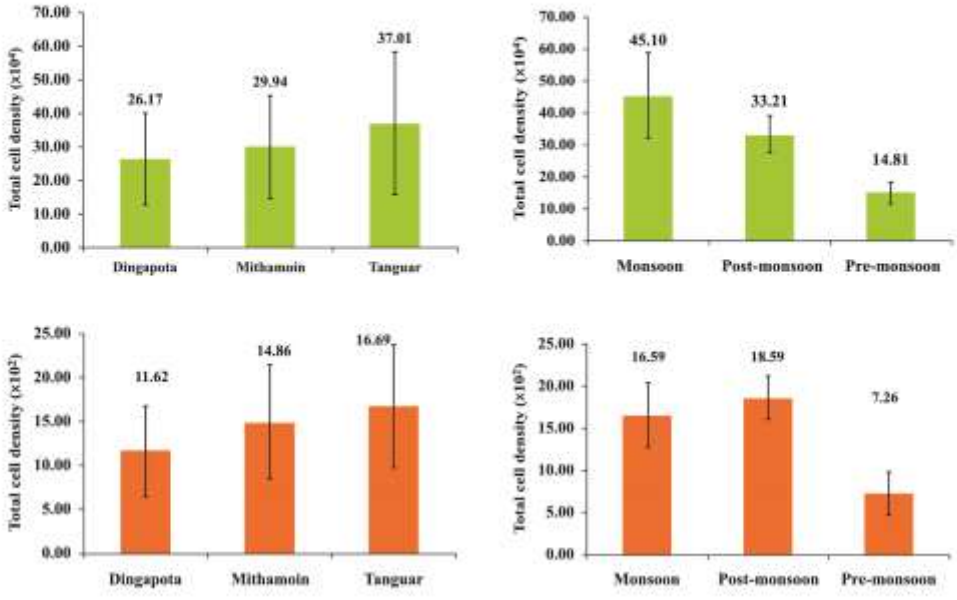


Figure 2. Total cell density of Phytoplankton and Zooplankton of Studied *haors*

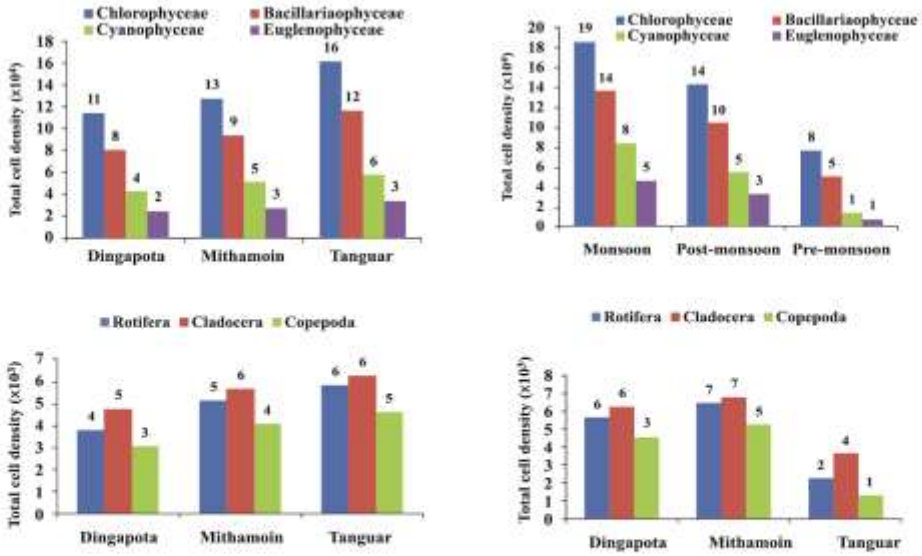


Figure 3. Group wise total cell density of Phytoplankton and Zooplankton of Studied haors.

Study 2.Field data collection for estimating population ecology of commercially significant beelfish

Fish catch composition and species diversity

Fish were collected using a seine net (10 m × 1.5 m; 0.5 mm mesh). Three, 30-m seine hauls were done at nearer areas of the selected site of the studied haors on each sampling date. Each haul was from deep water (1–1.5 m) to the bank of the haors. The fish were then sorted and identified according to their morphometric and meristics characters following Talwar and Jhingran (1991), Rahman (2005) and Rahman et al. (2009). After identification, fish were systematically classified according to Nelson (2006).

Statistical analysis

Collected data were analyzed into a database system with using the programs. Microsoft Excel (MS Excel,) PAST 4 (Paleontological Statistics), Computer software SPSS (version 20) was used.

Results

Fish catch composition and species diversity

A total of 109 fish species was recorded from 21 families and 8 orders (Table 4). *Hyporhamphus limbatus*, *Devario devario*, *Botia lohachata*, *Notopterus notopterus*, *Badis badis*, *Otolithoides pama*, *Rita rita*, *Eutropiichthys vacha* and *Silonia silondia* were absent in the sample of Dingapota. *Hyporhamphus limbatus*, *Botia lohachata*, *Badis badis*, *Otolithoides pama* and *Silonia silondia* were absent in the sample of Charigram, Cypriniformes was the largest group in all the studied locations. Herbivores was the most dominant trophic groups followed by omnivores and the least dominant trophic was detrivores in all the studied locations.

Shannon-wiener, Evenness and Margalef richness index of fish at studied haors during the study period were shown in (Table 5). Shannon index was the highest at Tanguar and the lowest at Dingapota. Evenness index was also the highest at Tanguar and the lowest at Dingapota. Furthermore, Margalef index of richness was the highest highest at Tanguar and the lowest at Dingapota.

Table 4. Check list of fish of the studied areas during study period.

Order	Family	Local Name	English Name	Scientific Name	Ding	Mitha	Tang
Beloniformes	Belonidae	Kakila	Freshwater Garfish	*Xenentodon cancila* (Hamilton, 1822)	+	+	+
		Ek Thuita	Congaturi Halfbeak	*Hyporhamphus limbatus* (Valenciennes, 1846)	−	+	+
Channiformes	Channidae	Taki, Lata	Spotted Snakehead	*Channa punctatus* (Bloch, 1793)	+	+	+
		Shol	Stripped Snakehead	*Channa striatus* (Bloch, 1793)	+	+	+
		Cheng	Walking Snakehead	*Channa orientalis* (Bloch andSchneider, 1801)	+	+	+
		Gajar	Giant Snakehead	*Channa marulius* (Hamilton, 1822)	+	+	+
Clupeiformes	Clupeidae	Kachki	Ganges river-Sprat	*Corica soborna* (Hamilton, 1822)	+	+	+
		Chapila	Indian river Shad	*Gudusia chapra* (Hamilton, 1822)	+	+	+
Cypriniformes	Cyprinidae	Silver carp	Silver carp	*Hypophthalmichthys molitrix* (Valenciennes, 1844)	+	+	−
		Big head carp	Big head carp	*Aristichthyes nobilis* (Hamilton, 1822)	−	−	+
		Grass carp	Grass Carp	*Ctenopharyngodon idella* (Valenciennes, 1844)	−	+	−
		Catla	Catla	*Gibelion catla* (Hamilton, 1822)	+	+	+
		Rui	Rahu	*Labeo rohita* (Hamilton, 1822)	+	+	+
		Bata	Bata Labeo	*Labeo bata* (Hamilton, 1822)	+	+	+
		Kalibaus	Kalbasu	*Labeo calbasu* (Hamilton, 1822)	+	+	+
		Gonia	Kuria Labeo	*Labeo gonius* (Hamilton, 1822)	+	+	+
		Mrigel	Mrigal	*Cirrhinus cirrhosus* (Bloch, 1795)	+	+	+
		Carpio	Common Carp	*Cyprinus carpio* (Linnaeus, 1758)	+	+	+
		Mola	Mola Carplet	*Amblypharyngodon mola* (Hamilton, 1822)	+	+	+
		Chela	Chela	*Chela laubuca* (Hamilton, 1822)	+	+	+
		Laacho	Reba	*Cirrhinus reba* (Hamilton, 1822)	+	+	+
		Baspata	Danio	*Devario devario* (Hamilton, 1822)	−	+	+
		Dhela	Cotio	*Osteobrama cotio* (Hamilton, 1822)	+	+	+
		Sar Punti	Olive Barb	*Pontius sarana* (Hamilton, 1822)	+	+	+
		Jat Punti	Pool Barb	*Puntius sophore* (Hamilton, 1822)	+	+	+
		Tit Punti	Ticto Barb	*Pethia ticto* (Hamilton, 1822)	+	+	+
		Katari	Large Razor Belly	*Salmostoma bacaila* (Hamilton,	+	+	+

		Darkina	Flying Barb	*Esomus danricus* (Hamilton, 1822)	+	+	+
	Cobitidae	Rani	Queen Loach	*Botia dario* (Hamilton, 1822)	+	+	+
		Rani	Queen Loach	*Botia lohachata* (Hamilton, 1822)	+	+	+
		Gutum	Guntea Loach	*Lepidocephalichthys guntea* (Hamilton, 1822)	+	+	+
		Poia	Gongota Loach	*Somileptes gongota* (Hamilton, 1822)	+	+	+
Osteoglossiformes	Notopteridae	Chital	Humped Featherback	*Notopterus chitala* (Hamilton, 1822)	+	+	+
		Foli Kanla	Gray Featherback	*Notopterus notopterus* (Pallas, 1769)	+	+	+
Perciformes	Anabantidae	Koi	Climbing Perch	*Anabas testudineus* (Bloch, 1775)	+	+	+
	Ambassidae	Nama Chanda	Glass-Perchlet	*Chanda nama* (Hamilton, 1822)	+	+	+
		Ranga Chanda	Indian Glassy Fish	*Pseudambassis ranga* (Hamilton, 1822)	+	+	+
	Gobidae	Bailla	Tank Goby	*Glossogobius giuris* (Hamilton, 1822)	+	+	+
		Chuno Bele	Glass Goby	*Gobiopterus chuno* (Hamilton, 1822)	+	+	+
	Mastacembelidae	Tara Baim	Lesser Spiny Eel	*Macrognathus aculeatus* (Bloch, 1786)	+	+	+
		Guchi	Sriped Spiny Eel	*Macrognathus pancalus* (Hamilton, 1822)	+	+	+
		Sal Baim	Tire-Track Spiny Eel	*Mastacembelus armatus* (Lacepede, 1800)	+	+	+
	Nandidae	Bheda, Meni	Mud Perch	*Nandus nandus* (Hamilton, 1822)	+	+	+
	Osphronemidae	Khalisha	Giant Gourami	*Colisa fasciata* (Bloch, 1801)	+	+	+
		Lal Khalisha	Dwarf Gourami	*Colisa lalia* (Hamilton, 1822)	+	+	+
	Pristolepidae	Napte Koi	Badis	*Badis badis* (Hamilton,1822)	+	−	+
	Sciaenidae	Poa	Pama	*Otolithoides pama* (Hamilton,1822)	−	+	−
Siluriformes	Bagridae	Air	Long Whiskered Catfish	*Mystus aor* (Hamilton, 1822)	+	+	+
		Tengra	Day's Mystus	*Mystus bleekeri*(Hamilton, 1822)	+	+	+
		Gulsha	Gangetic Mystus	*Mystus cavasius* (Hamilton, 1822)	+	+	+
		Gulsha Tengra	Stripped Dwarf Catfish	*Mystus vittatus* (Bloch, 1797)	+	+	+
		Rita	Rita	*Rita rita* (Hamilton, 1822)	+	+	+
	Siluridae	Kani Pabda	Butter Catfish	*Ompok bimaculata* (Bloch, 1797)	+	+	+
		Pabda	Pabda Catfish	*Ompok pabda* (Hamilton, 1822)	+	+	+
		Boal	Boal	*Wallago attu* (Schneider, 1801)	+	+	+
		Kajuli	Gangetic Ailia	*Ailia coila* (Hamilton, 1822)	+	+	+

		Ghaura	Garua Bacha	*Clupisoma garua* (Hamilton, 1822)	+	+	+
		Bacha	Indus Garua	*Eutropiichthys vacha* (Hamilton, 1822)	+	+	+
		Shilong	Silond Catfish	*Silonia silondia* (Hamilton, 1822)	+	+	+
	Sisoridae	Baghair	Gangetic Goonch	*Bagarius bagarius* (Hamilton, 1822)	+	+	+
		Gogni	Indian Gagata	*Gagata cenia* (Hamilton, 1822)	+	+	+
	Clariidae	Magur	Walking Catfish	*Clarias batrachus* (Linnaeus, 1758)	+	+	+
	Heteropneustidae	Shing, Jiol	Stinging Catfish	*Heteropneustes fossilis* (Bloch, 1794)	+	+	+
	Chacidae	Chaka	Squarehead Catfish	*Chaca chaca* (Hamilton, 1822)	–	–	+
Synbranchiformes	Synbranchidae	Kuchia	Cuchia	*Monopterus cuchia* (Hamilton, 1822)	+	+	+
Tetrodontiformes	Tetradontidae	Potka	Common Pufferfish	*Tetraodon cutcutia* (Hamilton,1822)	+	+	+

Table 5. Analysis of species diversity indices.

Variables	Charigram haor	Dingapota haor	Tanguar haor
Sampled Spp	71	79	109
Sampled Individuals	6435	5312	5854
Shannon_Diversity Index (H')	3.24	3.31	3.44
Simpson's Diversity Index (D')	0.87	0.91	0.95
Margalef Richness Value (d)	5.37	5.77	6.02

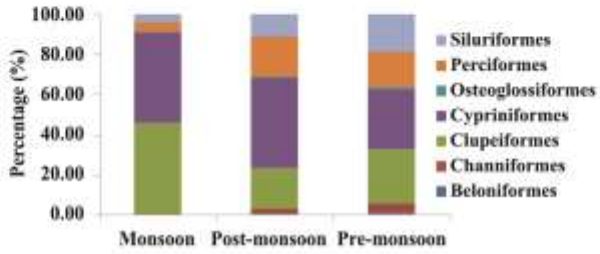


Figure 6. Order wise catch composition of fish at different locations of the studied haors.

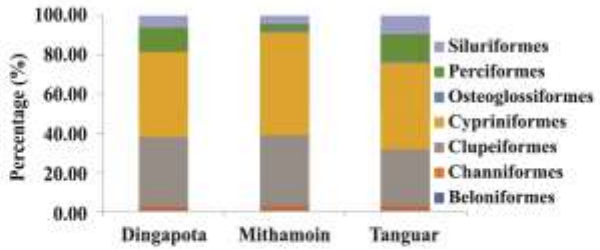


Figure 7. Order wise catch composition of fish in different seasons of the studied *haor*s.

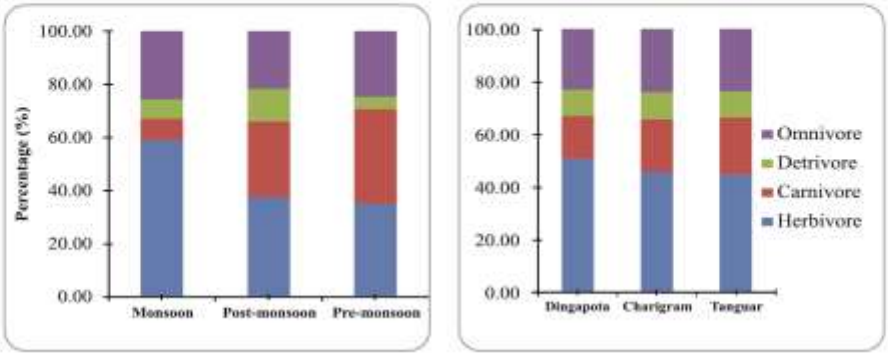


Figure 8. Tropic level of fish at different location and seasons of the studied *haors*.

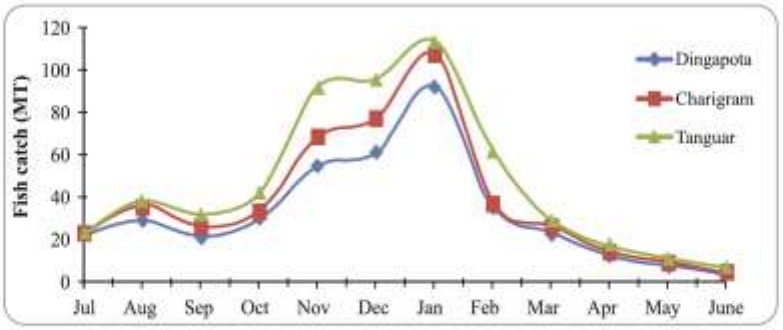


Figure 9. Monthly fluctuation of fish catches at selected haors.

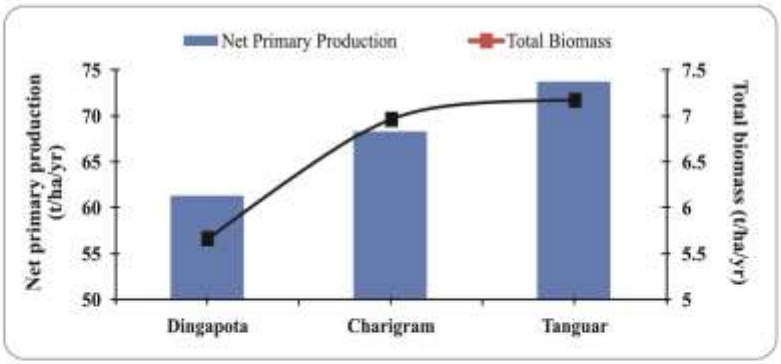


Figure 10. Productivity and biomass status of fish at selected haors.

Study 3. Assessment of net primary productivity, biomass and yield (Yield per Recruit) of different haors areas of Kishoreganj and Netrokona district

Table 4. Biomass and Productivity of different ecological groups of the selected haors during the study period.

SL No	Feeding habit-based groups	Tanguar *haor*		Dingapota *haor*		Charigram *haor*	
		Biomass (t/ha)	Productivity (t/ha)	Biomass (t/ha)	Productivity (t/ha)	Biomass (t/ha)	Productivity (t/ha)
1	Planktivores	6.63	0.86	5.71	0.81	5.24	0.75
2	Herbivores	1.61	0.49	1.38	0.43	1.41	0.41
3	Carnivores	0.93	0.31	0.88	0.29	0.92	0.31
4	Omnivores	0.52	0.19	0.51	0.18	0.49	0.15

Table 5. Estimates of yield, effort and remaining biomass proportion for EMY (Effort corresponding to maximum yield) and E0.1 strategy for selected haor areas.

Parameter	Unit	Charigram, (Charigram)	Dingapota *haor* (Mohanganj)	Tanguar *haor* (Sunamganj)	Habitat avr.
Net Primary Production	t/ha/yr	63.4	69.05	72.36	66.93
Total Biomass	t/ha	6.67	6.86	7.19	6.79
Ratio between Net Primary Production/ Total Biomass	-	9.5 . 1	10.2 . 1	10.37 . 1	11.03 . 1
Carrying capacity or MY (Maximum Yield)	t/ha/yr	1.83	1.87	2.96	2.21
Actual yield (E_{MY} . Effort corresponding to maximum yield)	kg/ha/yr	536	592	611	584
Predicted yield (Prediction model)	$Y_{E0.1}$ (kg/ha)	1622	1734	1779	1695
CPUE (Prediction model)	$CPUE_{E0.1}$ (kg/day)	0.81	0.88	0.93	0.87
	$CPUE_{max}$ (kg/day)	1.83	1.92	2.08	1.94
	$CPUE_{E0.1}$/ $CPUE_{max}$	0.44	0.46	0.45	0.45

Major findings

From the above statistics, it can be concluded that post- monsoon (Oct-Feb) is the pick time of fishing and exploitation and sampling station Tanguar haor is comparatively higher productive than the other two sampling station.

Development of Induced Breeding and Culture Techniques for Mekong Giant Catfish, *Pangasianodon gigas*

Researchers
Dr. Md. Khalilur Rahman, Director (Research and Planning)
A.K.M Saiful Islam, SSO
A.N.M Rezvi Kaysar Bhuiyan, SO

Objectives

- Study on application of vitamin A, D, and E and diet containing vitamin C for augmenting gonadal maturation of *P. gigas*
- Development of induced breeding techniques for *P. gigas*
- Development of rearing techniques for *P. gigas*

Achievements

Experiment 1. Studies on augmenting gonadal maturation of P. gigas through intramuscular injection of vitamin A, D, E and diets containing vitamin C

Fish rearing

Two ponds with an area of 40 decimals (Pond 1) and 150 decimals (Pond 2), respectively, were used to rear P. gigas. After preparing the ponds properly, water from deep tube wells were poured into the ponds. In pond-1 (P1), 7 fish were stocked and raised, whereas pond-2 had 40 fish (P2). Fish were fed a homemade diet consisting of readily available, approximately 28% protein materials at a rate of 1-3 % body weight. Feeding rates were 2-3% of body weight during the summer and 1% during the winter. Tamarind (Tetul) were incorporated into the meal at a dosage of 200 g/10 kg to inhibit the buildup of body fat. Ascorbic acid, a form of vitamin C, were also included to the standard diet (Table 2). In addition, vitamins A, D, and E (Tocopherol) were administered in order to speeding up egg development.

Table 1. Ingredients and composition of non-pelleted home-made feed for P. gigas.

Feed ingredients	Composition (%)	Protein (%)
Fish meal	20	12.30
Mustard oil cake	15	6.36
Rice bran	35	3.36
Wheat bran	10	2.96
Maize powder	20	2.52
Total	**100**	**28.00**
Vitamin premix	1 g/10 kg	
Tamarind (Tetul)	200 g/10 kg	
Salt	2.50 g/kg	

Table 2. Application of vitamins for augmenting gonadal maturation of *P. gigas*

Pond No.	No. of fish stocked	Vitamin A, D, E (mg/kg feed)	Vitamin C (mg/kg feed)
01	07	6	75
02	40	3	50

From February on-wards, health and ovarian development of fish were monitored. Maturation of *P. gigas* gonads were assessed using ultrasound technique.

Ultrasonography

A bamboo made temporary stage was made on the bank of the experimental pond where a dark environment was created by surrounding the stage with black cloth. A portable B-mode ultrasound machine (esaote, MyLab 5) was placed on the stage to examining the gonads of fish. Several P. gigas were collected by netting the experimental ponds. Before examination, fish were kept in lying position. A 2.5 MHz waterproof convex transducer prob fitted with a 3m long cable was placed on both the lateral sides of fish and moved gently from the genital opening towards head of fish to produce an image in partial transverse section. Ultrasound transmission jelly was used between the probe surface and fish' skin to improve imaging. A consensus sex determination was obtained by two observers for each fish. Accuracy of ultrasound derived sex determination were verified by visual examination of gonads. Several ultrasound images of the gonad were captured in the monitor for each fish.

Results

After scanning the gonads of fish with waterproof transducer prob, an image was appeared in the monitor of the ultrasound system. The image of testes appeared as dark, lobed regions beneath the vertebral column (Figure 1) whereas the image of egg appeared as large, grainy, bright structure in the coelomic cavity on the ventral side of the fish (Figure 2).

Figure 1. Ultrasound image of male *P. gigas*.

Figure 2. Ultrasound image of female *P. gigas*

Experiment. 2. Development of induced breeding techniques of P. gigas

Depending on the maturity of the fish, breeding trials of *P. gigas* were supposed to be conducted from June onwards. From February on-wards, male and female fish were identified, and maturity of the fish was evaluated using ultrasound technique. After ultrasonography, it was revealed that ova and sperm of *P. gigas* was in developing conditions. According to Nikolsky (1963), development stage 1 was observed in sperm and eggs of *P. gigas.*

Water quality parameters

Water quality parameters such as temperature, dissolved oxygen (DO), pH, and ammonia of the experimental ponds were measured fortnightly and summarized in Table 3.

Table 3. Water quality parameters (mean±sd) of the rearing ponds of *P. gigas*

Parameters	Pond-1	Pond-2
Temperature (°C)	26.64 ± 2.17	26.37 ± 2.24
Dissolved oxygen (ppm)	6.57 ± 0.55	6.74 ± 0.53
pH	8.36 ± 0.97	8.17 ± 0.22
Ammonia (ppm)	0.04 ± 0.02	0.038 ± 0.01

According to Boyd and Pillai (1984), all of the water quality parameters of the experimental ponds were in suitable range.

Breeding Biology of Commercially Important Freshwater Mollusk and Development of Culture Technique with Fish

Researchers

Sonia Sku, SSO
Mohammad Ferdous Siddique, SSO
Md. Nazmul Hossen, SO

Objectives

- To investigate breeding biology of commercially important mollusk (mussel and snail) available in Bangladesh
- To develop breeding technique and culture system of mollusk with fish in confined condition and pond ecosystem

Achievements

Experiment 1. Estimation of gonad maturation stages of mussel (Lamellidens jenkinsianus) Collection of mussels

Methodology

One pond having sandy soil was selected in Freshwater Station, BFRI. Pond was dried and lime was applied at the rate of 1kg per decimal and filled with water. After pond preparation about 1500-2000 specimen of year-round collected mussels from different aquatic habitat were stocked. Both organic manure (at 5 kg) and inorganic fertilizers (Urea 125 g + TSP 100 g) were applied fortnightly for ensuring sufficient feeds for mussel. Twenty individual samples were brought to laboratory and processed for histological study within 24 hrs. Shell height, length and weight were measured with Vernier calipers and electric balance and data were recorded. Sample specimens were dissected for gonadal tissue collection and fixed in Davidsons's Fixative for 48 hrs. After that the sample were preserved in 70% ethyl alcohol until histological procedure. Following steps were followed for histological study.

Step 1. Slicing
Step 2. Dehydration, clearing and infiltration
Step 3. Embedding
Step 4. Trimming
Step 5. Sectioning
Step 6. Staining
Step 7. Mounting
Step 8. Microscopic observations

Developmental stages of male and female gametic cells were differentiated, as described by Peredo and Parada (1984). Development of ova and spermatozoa was summarized as mentioned by Chatchavalvanich et al. (2006).

Results

Five gametogenic stages of the mussels were identified during study period from August 2021 to April 2022. The results are as follows:

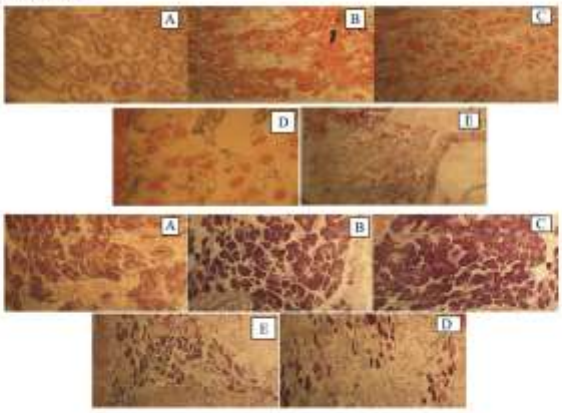


Figure 1. Five developmental stages of female and male mussel (*L. jenkinsianus*), A. Early Developing stage, B. Late Developing stage, C. Ripe stage, D. Spawning stage, E. Spent stage.

a. Early Development stage

In the female, oocytes remained in attached condition inside the follicles. In the male, spermatozoa were observed in attached condition inside the follicles.

b. Late Development stage

In this stage, most oocytes and spermatozoa were ready to release within the follicles.

c. Ripe stage

In the female, most oocytes were free within the follicles, but some oocytes were attached to the follicle wall. In the male, follicles filled by spermatozoa arranged in characteristic bands.

d. Spawning

In the female, large spaces inside the follicles and between free oocytes were present. Some follicles were completely devoid of oocytes. In the male, a marked decrease in the quantity of spermatozoa was observed in the follicles. Large spaces inside the follicles occurred. In some follicles, only a few residual spermatozoa were present. Highest value of spawning stage was found in August followed by December and January.

e. Spent

At this stage, most oocytes and spermatozoa were spawned and only some observed unspawned within follicles.

Within the study period (August 2021 to April 2022), the highest ripe ova were found in November followed by December while, highest percentage of spawning ova was found in August followed by December and January. The highest percentages of spent mussels were found in September followed by February till now. The peak breeding season can be identified after the end of the study period.

On the other hand, after the histological identification the male female ratio was found 1.1.30 (Table 1). Sex ratios are among the most basic of demographic parameters and provide an indication of both the relative survival of females and males and the future breeding potential of a population. So, it can be said that male female ratio was in good condition.

Table 1. Male and female ratio of mussel during the study period.

Month	Sample	Male	Female	Undifferentiated
August	20	7	11	2
September	20	8	12	0
October	20	8	12	0
November	20	11	8	1
December	20	8	12	0
January	20	8	11	1
February	20	10	10	0
March	20	6	14	0
April	20	9	8	3

Experiment 2. ***Determination of gametogenic cycle through Condition Index (CI) for mussel (L. jenkinsianus)***

Methodology

A total of 10 specimens were collected per month from stocking pond. Before dissection, shell length, width, height, and weight were measured. Weight of dry tissue and weight of dry shell was measured with an electric balance. Condition index is applied in bivalves to express the overall state of the animal. It often follows the gametogenic cycle of bivalves. CI was measured by using the following formula (Uddin et al. 2010).

$$\text{Condition Index} = \frac{\text{Wet tissue weight (g)}}{\text{Dry shell weight (g)}}$$

Results

After the measurement of CI, it was found that highest CI observed in August and second highest data found in December, January, and October also. The peak breeding season can be identified after the end of the study period.

Table 2. Condition Index (mean±sd) of freshwater mussel *L. jenkinsianus*.

Month	Wet tissue weight (g)	Dry shell weight (g)	Condition Index (CI)
August	25.43±4.2	35.44±10.68	0.71±0.19
September	20.05±2.33	49.45±12.22	0.40±0.11
October	17.09±2.64	27.02 ±6.57	0.63±0.15
November	17.98±2.01	39.43±7.62	0.45±0.08
December	26.56±3.96	40.06±11.94	0.66±0.13
January	24.17±2.77	37.42±7.18	0.66±0.14
February	19.92±3.05	54.14±8.91	0.37±0.06
March	11.32±1.79	22.91±5.79	0.51±0.13
April	17.66±4.36	40.13±4.75	0.44±0.13
May	15.82±2.74	32.33±3.25	0.48±0.09
June	26.89±5.58	45.53±1.37	0.59±0.07

Experiment 3. Impact of mussel's larvae (glochidia of L. marginalis) on carp production under poly culture management

Methodology

Pond was prepared following the standard procedure (drying, liming and fertilization). After pond preparation mussels and multi-species of fish were stocked as per design below.

Table 3. Experimental design.

Treatment	Species combination (Per decimal)		
	Mussel (*L. marginalis*)	Rajputi (*Barbodes gonionotus*)	Monosex Tilapia (*Oreochromis niloticaus*)
T_1	-	120	25
T_2	150	120	25
T_3	200	120	25
T_4	250	120	25
T_5	300	120	25

Fish was fed with commercial feed at the rate of initially 10% and down to 3% of the total biomass of the fish twice daily (half ration morning and rest half evening). Organic and inorganic fertilizer was applied fortnightly at the rate of 3 kg 0.1 kg TSP and 0.1 kg urea per decimal. Lime was applied fortnightly at the rate of 0.5 kg/decimal.

Results

During the study period (December 2021 to June 2022) we found the following result that is given in table 4.

Table 4. Weight performance (mean±sd) of *B. gonionotus* and *O. nilotica*.

Treatment	Species combination				
	Density of Mussel (*L. marginalis*)	Rajputi (*B. gonionotus*)		Tilapia (*O. niloticaus*)	
		Average initial weight (g)	Average final weight (g)	Average initial weight (g)	Average final weight (g)
T_1	-	23.1±3.64	147.5±7.3	25.4±2.41	193.9±1.24
T_2	150	21.3±1.61	146.5±4.4	22.8±1.96	198.4±2.15
T_3	200	25.7±3.15	156.6±6.3	25.7±3.15	200.8±7.05
T_4	250	25.5±3.18	156.4±9.56	25.5±3.18	197.7±6.01
T_5	300	24.9±3.32	141.7±4.02	24.9±3.32	194.3±6.01

From the above result it was observed that the average initial weight in T_1 were 23.1±3.64 g and 25.4±2.41 g and the average present weight 147.5±7.3 g and 193.9±1.24 g of *Barbodes gonionotus* and *Oreochromis nilotica,* respectively. The average initial weight and present weight of others Treatment (T_2, T_3, T_4, T_5) were like T_1. So, it can be said that till now the presence of glochidia in fish body does not make any problem on fish growth. Water temperature, pH, and alkalinity, ammonia, DO were recorded fortnightly.

Table 5. Water quality parameters (mean±sd).

Parameters	T_1	T_2	T_3	T_4	T_5
Temperature (^{0}C)	26.15±4.84	25.93±4.64	25.84±4.57	25.83±4.57	25.83±4.5
pH	7.92±0.34	7.87±0.22	7.87±0.42	7.91±0.25	7.74±0.29
DO (mg/l)	5.3±0.69	5.1±0.59	5.54±0.48	5.3±0.53	5.18±0.56
Ammonia (mg/l)	0.01±0.01	0.02±0.01	0.01±0.01	0.02±0.01	0.02±0.00
Alkalinity (mg/l)	128±21.00	115.14±6.20	117±12.24	127±14.53	117±7.55

Expt. 4. Natural propagation and seed production of freshwater apple snail (Pila globosa)

The ponds were prepared by following standard procedure. Lime was applied at the rate of 1-1.5 kg/decimal and filled up with water up to 0.3m. After that compost (mixture of cow dung, mustard oilcake and urea at the ratio of 1.0 kg, 1.0 kg and 0.5 kg/decimal) were applied throughout the pond at noon. This mixing was manually kept in water for three days and mixed to the pond water. After 07 days, pond was filled up with 0.9m -1.2m water and then collected snails were stocked at the prepared pond. Brood snail was collected from different aquatic habitats in July and August for rearing in pond. Snail was nourished by the natural and commercial food. Organic and inorganic fertilizers were applied fortnightly to the pond at the rate of 3 kg organic manure, 0.1 kg T.S.P and 0.1 kg urea per decimal. Lime was applied fortnightly at 0.5 kg/decimal.

Table 6. Experimental design.

Treatment	Stocking Density/decimal
T_1	400
T_2	600
T_3	800

Results

During breeding season snail eggs were collected from breeding ponds and kept in controlled condition in different techniques. So far, we have collected about 150 eggs cluster and maximum hatching rate was 45%.

Table 7. Collected egg cluster and hatching rate of snail.

Treatment	Stocking Density/decimal	Collected egg cluster number	Hatching rate (%)
T_1	400	30	37
T_2	600	40	41
T_3	800	80	45

Development of Breeding Technique of Snakeheads Fish

Researchers

Md. Rabiul Awal, SO
Maliha Khanom, SO
Md. Saiful Islam, SO

Objectives

- Collection and domestication of the Shol, *Channa striata*
- To study the reproductive biology of *C. striata* and *C. punctata*
- To develop controlled and induced breeding technique of Shol fish

Achievements

Experiment 1. Collection and domestication of the Shol, C. striata

The experiment was carried out in three experimental ponds in Freshwater Station, BFRI from November 2021 to May 2022. The present experiment was conducted with three Treatments namely T1, T2, and T3 each with three replications. The experimental designs are given below.

Table 1. Experimental design for the domestication of Shol fish fry in ponds.

Treatment s	Feed	Stocking density (nos./ha)
T_1	Commercial feed at 08-05% BW.	10,000
T_2	Fish muscle paste and commercial feed at 08-05% BW	
T_3	Live fish fry and trash fish at 08-05% BW.	

Pond preparation

The ponds were prepared by draining out the water and being exposed to sunlight for about 2 weeks. Lime was applied at the rate of 250 kg/ha. Two days after liming, the ponds were filled up with water. The research ponds were fenced by nylon net with bamboo sticks to make more replicable ponds as shortages of ponds facilities. Urea (50 kg/ha) and TSP (50 kg/ha) were applied uniformly into the ponds after 5 days of liming by throwing. To maintain water quality, the pond water was changed at regular intervals using water from a deep tube-well.

Collection and stocking of fish

Required no. of fry of Shol was collected from haor of Netrokona and Kishoreganj districts. Fish were transported to the experimental sites through plastic bags with proper aeration. Stocking was done according to their feeding habits. The initial average length and weight of fish were taken before stocking in the ponds.

Feed preparation and feeding

For T_1 Treatment, diets containing 40-35% protein, with a mixture of raw materials such as fish meal, MOC, rice bran, and wheat bran were applied at 8-5% of estimated BW. For T_2, fish were provided fish muscle paste mixed with commercial feed at 8-5% of estimated BW and for T_3 Treatment, live fish fry and trash fish were supplied at 8-5% of estimated BW.

Growth sampling of fish

Fish were sampled every 15 days intervals in the morning. The length and weight were recorded by random sampling from each pond.

Fish harvesting

Harvesting was done in the month of May-2022 by dewatering after completion of the experiment.

Water quality monitoring

The physico-chemical parameters of pond water were monitored fortnightly, and data recorded.

Analysis of experimental data on the growth parameters

Fish was weighed to grams using an electronic balance. All fish growth parameters were recorded and analyzed such as length and weight gain, survival, and specific growth rate (SGR). The procedure of calculation is as follows.

Length gain (cm) = Average final length – Average initial length
Weight gain (g) = Mean final weight (g) - Mean initial weight (g).
SGR (%/bwd) = {(ln final wt. - ln initial wt.)/ Culture period in days}x 100 (Brown, 1957)
Survival rate (%) = (No. of fish harvested/ No. of fish stocked) x 100 (De Silva, 1989)

Results

The experimental ponds were harvested after 180 days of culture. In this experiment, feed showed a clear effect on the final weight, survival, FCR, and production of *C. striata*.

Physico-chemical condition of water

The water quality parameters i.e. water temperature (°C), pH, dissolved oxygen, ammonia, and total alkalinity (mg/l) of all the treatments were recorded at bi-weekly intervals during the experimental period. The water temperatures were more or less similar throughout the experimental period. The values of water temperature ranged from 22.21 to 31.41°C, 22.8 to 30.41°C, and 22.2- to 30.54°C in T_1, T_2, and T_3, respectively. The lowest mean water temperature of 26.93°C was found in T2 and the highest water temperature 27.39°C was found in T_1 (Table 2). The values of dissolved oxygen ranged from 3.6 to 6.29 ppm, 3.32 to 5.79 ppm, and 3.15 to 5.8 ppm in T_1, T_2, and T_3, respectively (Table 2). The lowest mean value of dissolved oxygen 5.09 ppm was found in T_1, whereas the highest 5.32 ppm was found in T_3. The values of pH ranged from 7.5 to 8.76, 6.31 to 8.22, and 6.5 to 8.3 in T_1, T_2, and T_3, respectively. The lowest value of pH of 7.69 was found in T_2 and the highest value 7.99 was found in T_1 (Table 2). There was no remarkable variation in pH throughout the experiment. The results showed that the values of total alkalinity ranged from 110 to 130, 100 to 130 and 110 to 130 mg/l in T_1, T_2, and T_3, respectively. The lowest mean total alkalinity value of 116.7 mg/l was found in T_2 and the highest 118.3mg/l was found in T_1 (Table 2). The result showed that the values of NH_3-N ranged from 0.001 to 0.11, 0.001 to 0.09, and 0.002 to 0.10 mg/l in T_1, T_2, and T_3, respectively. The lowest mean value of 0.016 mg/l was found in T_2 and the highest at 0.027 mg/l was found in T_1 (Table 2).

Table 2. Water quality parameters (mean±sd) of the experimental ponds.

Parameters	Treatments			Suitable range
	T_1	T_2	T_3	
Water temperature (°C)	27.39±0.83 (22.21-31.41)	26.93±0.58 (22.8-30.41)	27.3±0.61 (22.2-30.54)	26-31°C
pH	7.99±.07 (7.5-8.76)	7.69±.09 (6.31-8.22)	7.78±.07 (6.5-8.3)	6.5-8.5
Dissolved oxygen (DO) (mg/l)	5.09±0.12 (3.6-6.29)	5.18±.13 (3.32-5.79)	5.32±.65 (3.15-5.8)	4-7mg/l
Total Alkalinity (mg/l)	118.3±1.31 (110-130)	116.7±1.52 (100-130)	117.6±1.7 (110-130)	70-190mg/l
NH_3(mg/l)	0.027±.008 (0.001-0.11)	0.016±0.005 (0.001-0.09)	0.022±0.006 (0.002-0.10)	0.0-0.04mg/l

*Values in parenthesis indicated range of the parameters

Growth performance of *C. striata*

The initial mean weight and length of the *C. striata* were 0.161±0.001 g and 2.83±0.01 cm, 0.162±0.001 g and 2.84±0.01 cm, and 0.162±0.001 g and 2.83±0.01cm in T_1, T_2, and T_3, respectively.

Table 3. Growth performance, survival (mean ± se), and FCR of *C. striata* fry.

Parameters	Treatments		
	T_1	T_2	T_3
Initial mean weight (g)	$0.161±0.001^a$	$0.162±0.001^a$	$0.162±0.001^a$
Initial mean length (cm)	$2.83±0.01^a$	$2.84±0.01^a$	$2.83±0.01^a$
Final mean weight (g)	$128.9±3.15^c$	$140.9±4.25^b$	$162.5±4.65^a$
Final mean length (cm)	$26.16±2.12^b$	$26.76±2.96^{ab}$	$27.66±3.12^a$
Av. daily weight gain (g)	$0.64±0.01^c$	$0.70±0.02^b$	$0.81±0.03^a$
Weight gain	$128.7±2.44^c$	$140.7±3.39^b$	$162.3±4.21^a$
Weight gain (%)	$81279±439^b$	$87867±467^{ab}$	$100999±952^a$
SGR (%/day)	$3.34±0.02^c$	$3.37±0.01^{ab}$	$3.45±0.02^a$
FCR	$2.26±0.04^c$	$2.05±0.035^b$	$1.79±0.028^a$
Survival (%)	$45.5±3.04^c$	$71.8±3.51^b$	$82.67±4.14^a$

The final mean weight of *C. striata* was 128.9±3.15 g, 140.9±4.25 g, and 162.5±4.65 g and the final mean length was 26.16±2.12 cm, 26.76±2.96 cm, and 27.66±3.12 cm, respectively in T_1, T_2, and T_3. The growth rates of the fry in different treatments are shown as weight gain and weight gain percent (Table 3). The weight gain was found 128.7±2.44 in T_1, 140.7±3.39 in T_2, and 162.3±4.21 in T_3. The survival rate was found to be 45.5±3.04, 71.8±3.51, and 82.67±4.14 in T_1, T_2, and T_3, respectively. Statistical analysis indicated that weight gain and survival of fry of T_3 were significantly higher ($p< 0.05$) than those of T_1 and T_2. The mean specific growth rates were found at 3.34±0.02%, 3.37±0.01%, and 3.45±0.02% per day in T_1, T_2, and T_3, respectively.

Experiment 2. *Study of the reproductive biology of Channa striata and C. punctata*

The experiment was carried out for 12 consecutive months from July 2021 to June 2022 in the Freshwater Station, Bangladesh Fisheries Research Institute, (BFRI) Mymensingh. In each month at least 05-10 pairs of fish were collected from the local market of Mymensingh and haor of Kishoreganj district for the determination of gonado-somatic index and fecundity.

Laboratory studies

Individual fish was measured for total length to the nearest cm with a measuring scale and body weight to the nearest g by an electronic balance.

Gonadosomatic Index (GSI)

The total body and gonad weight of collected fish in each month was considered to calculate the mean Gonadosomatic Index (GSI). Gonadosomatic Index (GSI) was calculated according to the formula.

$$GSI = \frac{\text{Weight of gonad}}{\text{Weight of fish}} \times 100$$

Fecundity estimation

The gravimetric method was used for the estimation of the fecundity of fish which is more efficient and gives a fairly accurate result. In this method, the ovaries were dissected with a pair of scissors. The gonad samples were swollen heavily, their membrane became transparent and the number of mature and maturing eggs from each portion was found out separately by actual counting. The mean number of eggs in 1.00 g was determined and then multiplied by the total weight of the ovary, which gave the total number of eggs *i.e.* the fecundity of the respective fish. The fecundity of the fish was obtained by using the following formula:

$$F = \frac{\text{Overy weight}}{\text{Sub-sample of ovary}} \times \text{No.of eggs}$$

Histology of Gonad

To study the sequential histological changes, the ovaries tissues were fixed in aqueous Bouin's fluid and the then routine histological process was done for slide preparation. Ovaries and testis belonging to a range of developmental stages were prepared for histological study by fixing in 10% formalin, washing in running water and storing in 70% alcohol until sectioning. Examination of a cross-section from different parts of ovaries and testis were prepared and finally observed microscopically to take a perfect picture. Those sections were dehydrated in a graded series of alcohol, cleared in chloroform, embedded in paraffin, trimmed by using a sharp knife and sectioned by using a microtome blade in a microtome machine at 3-4μ m. The sections were fixed on glass slides with Mayer's fluid, stained routinely with hematoxylin and eosin following the standard histological procedure, mounted in a mounting agent like DPX and examined to identify the gametogenic cell types.

Results

Reproductive biology of *C. striata*

Gonado-somatic indices (GSI) were recorded each month and variations were found. The highest GSI value for the ovary was recorded in July (5.67) while it was found to be 0.32 in the case of the testis. The lowest GSI value for the ovary was recorded in October (0.17) while it was found to be (0.011) in the case of the testis and gradually increased from February to May and reached a maximum value of 5.67 and 0.32 for females and males respectively. Later in October, there was a fall in the GSI values for both males and females. This tendency continued up to January (Figure 1). Therefore, June-July was the peak breeding season of *C. striata* during study areas.

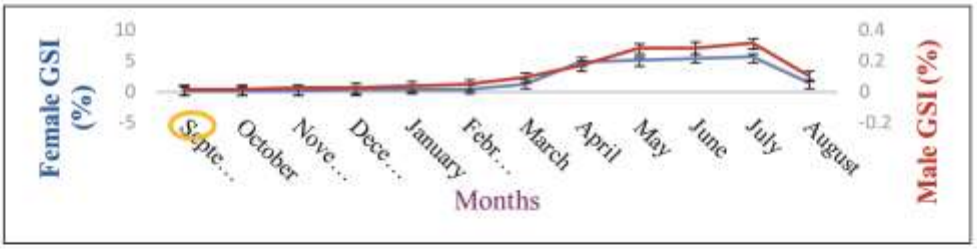


Figure 1. Monthly variation of GSI (%) of *C. striata* during the study period.

The fecundity of the fish also showed variation in different months. Figure (2) shows the absolute fecundity of mature female *C. striata*. From the study, the highest absolute fecundity (number of oocytes per female) was found to be 28768 in July. The absolute fecundity value was found to be lowest in October at 3128 with an average value of about 12049 throughout the study period.

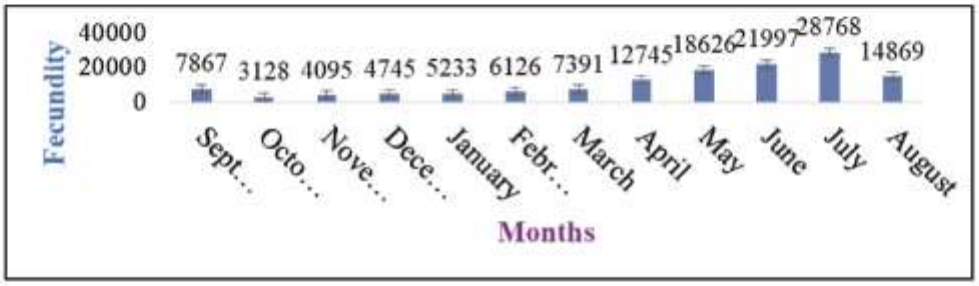
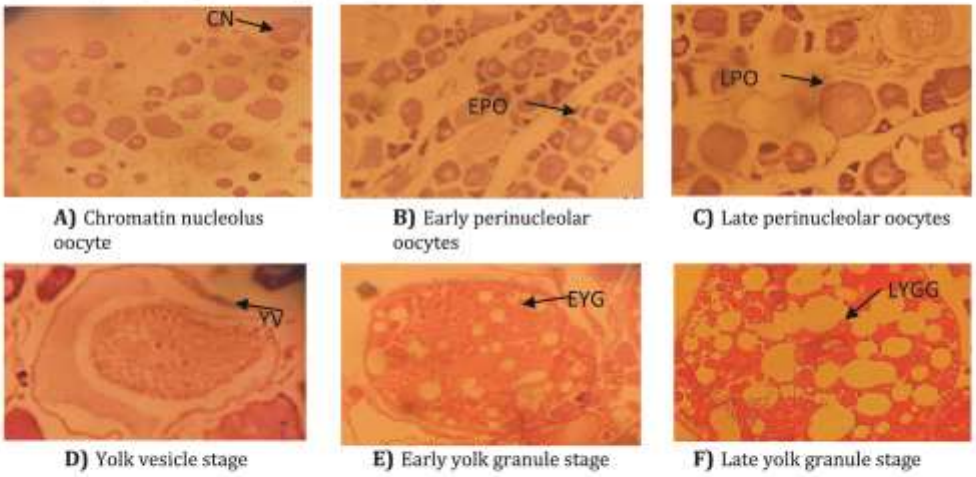


Figure 2. Monthly variation of fecundity (number of oocytes per female) of *C. striata* during the study period

Histological studies on gonads deliver more dependable and direct indications of the reproductive status of the fish. Based on histological criteria, in the present study the development of the oocyte stages was divided into chromatin nuclear stage, early perinuclear stage, late perinuclear stage, yolk vesicle stage, early yolk granule stage and late yolk granule stage. Presence of oocytes at various maturation stages viz. Chromatin nuclear stage (CN) in December, early perinuclear oocytes (EPO) in January, late perinuclear oocytes (LPO) in February, yolk vesicle stage (YV) in March, early yolk granule stage (EYG) in April-May, late yolk granule stage (LYG) in July were observed..



A) Chromatin nucleolus oocyte **B)** Early perinucleolar oocytes **C)** Late perinucleolar oocytes

D) Yolk vesicle stage **E)** Early yolk granule stage **F)** Late yolk granule stage

Figure 3. Developmental stages of female *C. striata* oocytes during the study period

Reproductive biology of *C. punctata*

Gonadosomatic indices (GSI) were recorded each month and variations were found. The highest GSI value for the ovary was recorded in July (8.32) while it was found to be 0.42 in the case of the testis. The lowest GSI value was recorded in October (0.40) for females and gradually increased from February to May and reached a maximum value of 8.32 and 0.42 for females and males respectively. Later in October, there was a fall in the GSI values for both males and females. This tendency continued up to January (Figure 3).

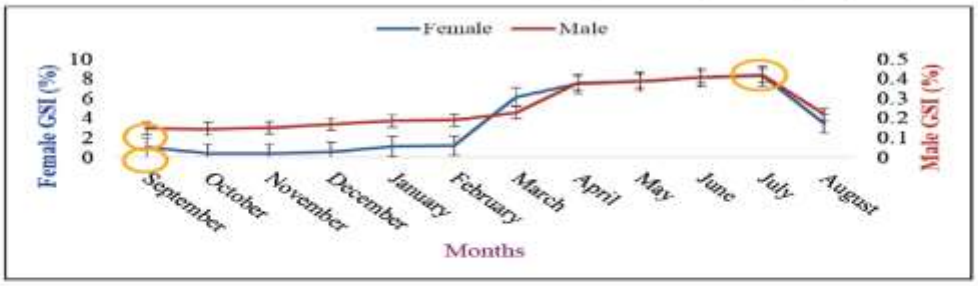


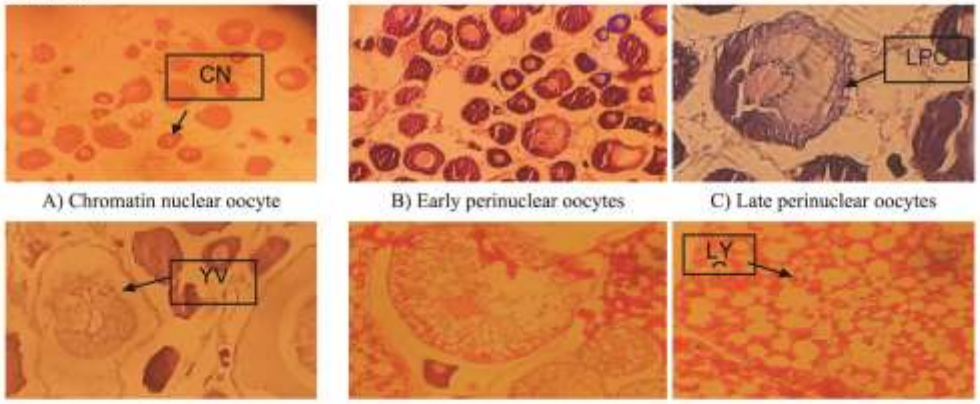
Figure 3. Monthly variation of GSI (%) of *C. punctata* during the study period.

The fecundity of the fish also showed variation in different months. Figure (4) shows the absolute fecundity of mature female *C. punctata*. From the study, the highest absolute fecundity (number of oocytes per female) was found to be 23847 in July. The absolute fecundity value was found to be lowest in October 1998 with an average value of about 9597 throughout the study period (Figure 4).

Figure 4. Monthly variation of fecundity (number of oocytes per female) of *C. punctata* during the study period.

The highest mean GSI value of females during July was found to be 8.32. Similarly, the mean highest GSI value of *C. punctata* male in July was 0.32 (Figure 4). Therefore, June-July was the peak breeding season of *C. punctata* during study areas.

In the present study the development of the oocyte stages was divided into chromatin nuclear stage, early perinuclear stage, late perinuclear stage, yolk vesicle stage, early yolk granule stage and late yolk granule stage. Presence of oocytes at various maturation stages *viz*. Chromatin nuclear stage (CN) in December, early perinuclear oocytes (EPO) in January, late perinuclear oocytes (LPO) in February, yolk vesicle stage (YV) in March, early yolk granule stage (EYG) in April-May, late yolk granule stage (LYG) in July were observed.



A) Chromatin nuclear oocyte B) Early perinuclear oocytes C) Late perinuclear oocytes

D) Yolk vesicle stage E) Early yolk granule stage F) Late yolk granule stage

Figure 5. Developmental stages of female *C. punctata* oocytes during the study period.

Experiment 3. Development of controlled and induced breeding technique of Shol fish

The experiment was conducted in the hatchery complex of Freshwater station of Bangladesh Fisheries Research Institute, Mymensingh during April-July, 2020.

Collection of fish

A total of 200 *C. striata* fish were collected from the haor of Netrokona and Kishoriganj districts from November-December/2021. After transportation, fish were acclimatized for 30 minutes in the selected pond. Then collected fish were stocked in a pond having an area of 10 decimals. The average initial length and weight of collected fish were recorded at 32.5±9.09 cm and 573.1±89.18 g, respectively. The stocked fish were reared with live fish fry and commercial feed at 5-3% body weight twice daily.

Selection of brood fish

For breeding trials, matured and healthy brood fish were selected randomly by netting from the pond and weighted. A total of 54 gravid male and female fish (36 males and 18 females) were selected and kept in cisterns for breeding purposes.

Acclimatizing of fish

At first, selected matured female and male fish were weighed and kept in separate cisterns for 5-6 hours for conditioning before being treated with PG extract solution. For conditioning, continuous water flow was supplied to ensure sufficient dissolved oxygen for fish through a porous PVC pipe on top of the cistern.

Experimental design

Three treatments were conducted for induced breeding of *C. striata*. Three different doses of PG *viz*. 70, 80, and 90 mg/kg body weight of female fish were applied which is considered as T1, T2, and T3, respectively in the month of May 2022 to develop the induced breeding technique of *C. striata*. Eighteen females and thirty-six males were divided into three treatments and designated as T1, T2, and T3, respectively having three replications indicated as R1, R2, and R3 and kept in separate glass nylon hapa which was previously set up in a cistern. For males under T1, T2, and T3 the dose of 35, 40, and 45 mg/kg body weight were administrated.

Preparation of PG extract

For hormonal injection, commercially available dry carp pituitary glands (PG) which are available on the market were collected first. At first, a required amount of PG was carefully weighed out in an electronic balance. The amount to be weighed out was calculated on the body weight of all the fish of a particular treatment to be injected on a particular day using the following formula.

Weight (mg) of the required amount of PG (Wt) = Wb x Pt

Where, Wb represents the total body weight (kg) of all the fish to be injected and Pt represents the rate in mg of PG to be injected/kg body weight under a particular treatment.

The total volume of the extract required was calculated by the following formula:

Vol. of extract (ml) = Wt x 1.0 [Wt= Weight of PG (mg)]

Where, 1.0 represents the volume of the extract in ml to be injected/kg body weight. The weighed PG was homogenized with a small volume of distilled water and the homogenate was carefully transferred to a centrifuge tube by using distilled water to ensure complete transfer. The mixture was centrifuged for 6 min at 6000 rpm. The clear supernatant was transferred to a vial and it was made pre-determined volume with distilled water.

Injecting the PG extract into fish

Based on the body weight of the gravid female fish the required volume of extract was taken in a graduated 3.0 ml hypodermic syringe. The selected brood fish were carefully taken for injecting the PG extract. The extract was injected intramuscularly into the fish on the dorsal side above the lateral line.

Indices of the effectiveness of PG dose

The following parameters were recorded as indices of the effectiveness of different PG doses:
Percent ovulation, percent fertilization; and percent hatching were calculated using the following formula.

$$\% \text{ ovulation} = \frac{\text{No. of fish ovulated}}{\text{Total no. of fish injected}} \text{X } 100$$

$$\% \text{ fertilization} = \frac{\text{No. of fertilized eggs}}{\text{Total no. of eggs (fertilized + unfertilized)}} \text{ X } 100$$

$$\% \text{ hatching} = \frac{\text{No. of eggs hatched}}{\text{Total no. of eggs}} \text{X } 100$$

For calculating percent fertilization, several egg samples were taken from each group and the number of fertilized and unfertilized eggs were counted under a microscope.

Results

Three treatments were conducted for induced breeding of *C. striata* with PG extract during May with a temperature around 28-31°C. To develop the PG dose for inducing ovulation in female *C. striata*, three different doses of carp PG extract were applied. Data representing the effects of PG doses on ovulation of female fish and the rates of fertilization and hatching of eggs are presented in Table 2. For developing PG dose in female *C. striata*, three different doses of PG *viz*., 60, 70, and 80 mg/kg body weight were applied in treatments T_1, T_2, and T_3, respectively in May. Fish showed minimum response with the 1st dose applied in T_1 but with 3rd doses applied in T_3 showed good response in consideration of ovulation, fertilization, and hatching.

Table 04. Response of different doses of Pituitary Gland (mean±sd) on induction of spawning of *C. striata.*

Hormone	Treatments	Body weight (g)		Dosage of hormone/ (kg)		Spawning period (hrs.)	Spawning rate (%)	Fertilization rate (%)	Hatching period (hrs.)	Hatching rate (%)	Incubation Temp. (°C)
		Female	Male	Female	Male						
PG	T_1	527.2± 82.2	446.9 ±57.5	70	35	14-16	34.4 ±4.5^c	41.5± 5.89^c	32-36	39.6± 4.9^c	28-31
	T_2	511.5± 56.5	432.3 ±43.1	80	40	14-16	57.6 ±7.7^b	55.6± 5.73^b	32-36	53.1± 4.5^b	
	T_3	551.3± 61.3	466.4 ±67.4	90	45	14-16	85.6 ±8.5^a	79.3± 4.89^a	32-36	64.3± 6.6^a	

** Figures with the same letter are not significantly ($p<0.05$) different.

From the study, the spawning rate showed noticeable differences in effectiveness among three doses in inducing ovulation. The spawning rate was recorded as 34.4%, 57.6%, and 85.6% in the Treatments of T_1, T_2, and T_3, respectively (Table 6). The time interval between the injection of PG extract and ovulation (latency period) varied between 14-16 hours of injection in all cases. Among three doses of PG in consideration of the spawning rate, T_3 showed the highest result followed by T_2 and T_1. The results from the ANOVA test indicated that there was a significant difference among the three doses of Treatments whereas T_3 was significantly ($p<0.05$) higher than that of Treatments T_1 and T_2 (Table 6). The average fertilization rate was recorded as 41.5±5.89, 55.6±5.73, and 79.3±4.89 in Treatments T_1, T_2, and T_3, respectively. The highest fertilization rate (79.3%) was recorded in T_3 while the lowest (41.5%) was found in T_1. The results from the ANOVA test indicated that there was a significant difference among the three doses whereas T_3 was significantly ($p<0.05$) higher than that of Treatments T_1 and T_2 (Table 1). Some little variation might be due to water quality, water temperature, brood management, etc. The hatching rate was found 39.6%, 53.1%, and 64.3% in Treatments of T_1, T_2, and T_3, respectively. The highest hatching rate was recorded 64.3±6.6 in T_3 and the lowest hatching rate was recorded 39.6±4.9 in T_1. The result from the ANOVA test indicated that there was a significant difference between the three doses. It was found that the hatching rate in T_3 was significantly ($p<0.05$) higher than that of T_1.

Problems/constraints encountered if any.

C. striata is an active predator, so it is very difficult to adopt feed for this fish in captive conditions. Sex identification is also a big problem in this fish from outside observation.

Conservation and Seed Production of Indigenaus Fish Speccies in Bangladesh

Researchers
Dr. Salina Yeasmine, SSO
Md. Rabiul Awal, SO
Md. Shahin Alam, SO

Objectives

- To refine breeding technique of Dhela (*Osteobrama cotio*) and Rani (*Botia. dario*)
- To develop nursery and culture technique of Dhela (*O. cotio*) and Rani (*B. dario*) in ponds under different stocking density
- To collect and domesticate Hiralu (*Barilius bendelisis*), Gang tengra (*Gagata youssoufi*) and Garua (*Clupisoma garua*)
- To study gonado-somatic index of Hiralu (*Barilius bendelisis*), Gang tengra (*Gagata youssoufi*) and Garua (*Clupisoma garua*)
- To study histological observation of gonads to find out the breeding season of Hiralu (*Barilius bendelisis*), Gang tengra (*Gagata youssoufi*) and Garua (*Clupisoma garua*)
- To investigate the efficacy of different hormones for induced breeding of Gang tengra (*Gagata youssoufi*), Mohashoal (*Tor tor*) and Garua (*Clupisoma garua*)
- To collect indigenous freshwater fish species from different regions for live gene-bank

Achievements

The following research works were done under the project during July 2021 to June 2022.

Experiment 1. Refinement of breeding technique of Dhela (O. cotio) and Rani (B. dario)

Collection and domestication of Dhela (*O. cotio*) and Rani *(B. dario)* in the pond

A total of two thousand (2000) Dhela (*O. cotio*) were collected from Brahmaputra River of Mymensingh and stocked at the rate of 30-35/decimal in the pond at Freshwater Station, BFRI having an area of 10 decimal and 1m or 0.8 m water depth. Fish were reared with supplementary feed at of 4-5% body weight twice daily for raising gonad development. Supplementary feeds were used twice a day at 8.00 am and 4.00 pm. After gonadal development breeding program were done. A total of 4500 Rani (*B. Dario*) was collected from haor of Netrokona and Jamuna river and stocked at the rate of 30-35/decimal in the previously prepared pond at Freshwater Station, BFRI having an area of 10 decimal and 1 m water depth. Fish were reared by supplying commercial feed at of 4-5% body weight twice daily. After gonadal development breeding program were done.

Refinement of induced breeding technique of Dhela (*O. cotio*)

The present experiment was undertaken to refine induced breeding technique of Dhela (*O. cotio*). Induced breeding trials of Dhela (*O. cotio*) were conducted to adjust hormone doses during May-June 2022. In this experiment for the ovulation of female's different doses (2, 4 and 6 mg/kg body weight of fish) of PG was used to confirm the optimum PG dose. On the other hand, male fish were treated with 2mg/kg body weight. Single dose was applied in both male and female fish. Three different treatment doses viz. T_1, T_2, and T_3 were used, and each dose were triplicated. Matured male and female fish were collected from the pond early in morning and weighted. After five hours of conditioning, PG was used for induced breeding of Dhela (*O. cotio*) with three different doses. The efficacy of PG doses and observed and collected data on ovulation, fertilization, hatching, and survival rates are summarized and shown in Table 1.

Figure 1. Injecting PG hormone of Dhela (*O. cotio*).

Table 1. Details of PG doses (mean±sd) on Dhela (*O. cotio*)

Treatment	Mean Body weight (g)		1st Injection dose (mg/kg^{-1})		Ovulation period (hr)	Ovulation rate (%)	Fertilization rate (%)	Hatching period (hr)	Hatching rate (%)	Incubation Temp.(°C)	Remarks
	Male	Female	Male	Female							
T_1	5.02 ±0.51	10.14 ±2.2	2	2	7-8	-	-	-	-	-	No Ovulation
T_2	5.02 ±0.22	11.22 ±3.1	2	4	7-8	40.1 ±4.0	50.12 ±8.02	22	40.24 ±12.60	32	Partial ovulation and fertilization and hatching
T_3	4.81 ±0.84	11.05 ±4.0	2	6	7-8	95.9 ±.5.0	98.22 ±11.10	22	96.38 ±10.26	32	Complete ovulation, Successful fertilization and hatching

Fish did not show any response with the dose applied in T_1 but with the doses applied in T_2 and T_3 showed minimum and good response respectively in consideration of ovulation, fertilization, and hatching.

Refinement of induced breeding technique of Rani (*B. dario*)

Induced breeding trials of Rani (*B. dario*) were conducted during June-July 2022. In this trials, different dose of synthetic hormone *viz*. 0.8, 1.0 and 1.5 ml/kg body weight of female fish were used to refine the breeding trials. On the other hand, male fish were treated with 0.4, 0.5 and 0.75 ml/kg body weight. Single dose was applied in male and female fish. Matured male and female fish were collected from the pond early in the morning. After five hours of conditioning, different doses of synthetic hormone were used for induced breeding of Rani (*B. dario*). The efficacy of synthetic hormone doses on Rani (*B. dario*) were observed and collected data on ovulation, fertilization, hatching, and survival rates are summarized in Table 2.

Table 2. Details of synthetic hormone doses on Rani (*B. dario*) and corresponding data on ovulation, fertilization, hatching and survival rates during study period.

Treatments	Mean Body weight (g)		1st Injection dose (ml/kg)		Ovulation period (hr)	Ovulation rate (%)	Fertilization rate (%)	Hatching period (hr)	Hatching rate (%)	Incubation Temp. (°C)	Remarks
	Male	Female	Male	Female							
T_1	6.01 ±0.40	12.84 ±2.10	0.4	0.80	-	-	-	-	-	-	No Ovulation
T_2	5.90 ±0.82	11.61 ±2.90	0.5	1.0	6	70.4 ±7	75.4 ±7	22	63.54 ±.13.79	32	Ovulation, Successful fertilization and hatching
T_3	5.73±0 .24	11.30±4. 20	0.75	1.5	6	-	-	-	-	-	No Ovulation

Experiment 02. Rearing and nursery technique of Dhela (O. cotio) in pond at different stocking density

The experiments were conducted for evaluating the production and survivability of spawn or larvae of Dhela (O. cotio) with different stocking density in nursery pond for 6 weeks period. Three stocking density *viz*. 0.7, 0.8 and 0.9 million/ha which considered as T_1, T_2 and T_3, respectively were maintained. The larvae/spawn were fed with nursery feed (finely powdered) twice daily (1.1 by weight) for the first two weeks; after that starter feed were supplied for last four weeks. The fry were sampled at weekly interval to determine the change in their length and weight. The experiment was carried out for 6 weeks and the produced fry were harvested by drying the ponds. The final length and weight were recorded for determining growth and survival of fry which are shown in Table 3. Highest growth performances and survival of the fry were observed in T_1 Treatment in terms of length and weight where stocking density was 0.7 million/ha.

Table 3. Growth performances in length and weight of Dhela (*Osteobrama cotio*) fry after 6 weeks rearing under different stocking density.

Treatment	Stocking density (million/ha)	Length (cm)			Weight (g)			Survival rate (%)
		Initial	Final	Net gain	Initial	Final	Net gain	
T_1	0.7	1.0	3.70	2.18±0.42	0.002	2.82	2.82±0.03	72.80±1.72
T_2	0.8	1.0	2.60	1.60±0.12	0.002	1.65	1.65±0.01	65.80±1.6
T_3	0.9	1.0	2.10	2.38±0.11	0.002	1.20	1.20±0.02	50.12±0.90

Experiment 3. Collection of Hiralu (B. bendelisis), Gang tengra (G. youssoufi) and Garua (C. garua) from wild sources and domestication in pond

A total of 500 of each species of Hiralu (*Barilius bendelisis*), Gang tengra (*Gagata youssoufi*) and Garua (*Clupisoma garua*) were collected from natural sources. Collected fish were transported to Freshwater Station in oxygenated bag or drum. After transportation, the fish were acclimatized in a pond for 1 hour. After acclimatization, the collected fish were stocked separately in two ponds at Freshwater Station, BFRI Mymensingh having an area of 20-30 decimal. During stocking initial length and weight of the collected fish were recorded. Average weight of collected Garua (*C. garua*) was 20±5 g. Average weight of collected Hiralu (*B. bendelisis*) was 4±1 g. The brood development and breeding program of Garua (*C. garua*) and Hiralu (*B. bendelisis*) will be continued in the subsequent years. The stocked fish are being reared by supplying commercial floating feed at of 2-3% body weight once daily. Monthly sampling is being done for feed adjustment.

Experiment 04. Study of reproductive biology of Hiralu (B. bendelisis), Gang tengra (G. youssoufi) and Garua (C. garua)

Collection of fish samples

The experiment was carried out for a period of 4 consecutive months, in the Freshwater Station, Bangladesh Fisheries Research Institute, Mymensingh. Total number of 10 females and 10 males of Garua (*C. garua*) were collected from Brahmaputra River of Mymensingh in each month, through the fishermen for the determination of fecundity and Gonado-Somatic Index.

Laboratory studies

Individual fish was measured for total length to the nearest cm with a measuring scale and body weight to the nearest g by an electronic balance.

Gonado-Somatic- Index (GSI)

Total body weight and gonad weight of collected fish in each month was considered to calculate the mean Gonado-Somatic Index (GSI). Gonado-Somatic Index (GSI) was calculated according to the formula

$$GSI = \frac{\text{Weight of gonad}}{\text{Weight of fish}} \times 100$$

Gonado-Somatic Index (GSI) of female and male Garua (*Clupisoma garua*) was calculated during January 2022 to June 2022 and monthly changes in mean GSI values of female and male are presented in Table 4 and 5.

Table 4. Details data and GSI values (mean±sd) of female Garua (*C. garua*) from January 2022 to June 2022.

Month	No. of fish examined	Body Length of fish (cm)	Body wt. of fish (g)	Ovary wt. (g)	GSI (%)
January	10	17.61±1.35	44.09±10.18	0.25±0.09	0.61±0.28
February	10	14. 66±0.12	25.8 ±11.98	0.4 ±0.26	0.90 ±0.42
March	10	19.25 ±2.34	49.2 ±13.43	0.393 ±0.28	0.78±0.24
April	10	20.12 ±2.36	51.32 ±20.23	0.41 ±0.16	0.82±0.31
May	10	22.11 ±3.10	63.15 ±12.22	0.40 ±0.10	0.78±0.14
June	10	23.21+3.10	75.12+12.32	0.41+0.16	0.90+0.12

Table 5. Details data and GSI values of male Garua (*C. garua*) from January 2022 to June 2022.

Month	No. of fish examined	Body Length of fish (cm)	Body wt. of fish (g)	Gonad wt. (g)	GSI (%)
January	10	12.31±1.34	25.08±5.15	0.15±0.04	0.31±0.11
February	10	14. 23±1.11	33.23 ±6.98	0.21 ±0.03	0.35 ±0.12
March	10	16.25 ±2.03	33.89 ±11.43	0.293 ±0.01	0.38±0.10
April	10	17.92 ±2.36	36.32 ±8.12	0. 31 ±0.02	0.42±0.11
May	10	17.10 ±1.86	36.64 ±6.71	0.31 ±0.02	0.52±0.11
June	10	17.90 ±1.86	38.64 ±3.32	0.34 ±0.12	0. 72±0.03

Figure 2. Measurement length (cm) of Garua (*C. garua*).

Figure 3. Measurement weight (g) of Garua (*C. garua*).

Figure 4. Position of ovary of Garua *(C. garua)*.

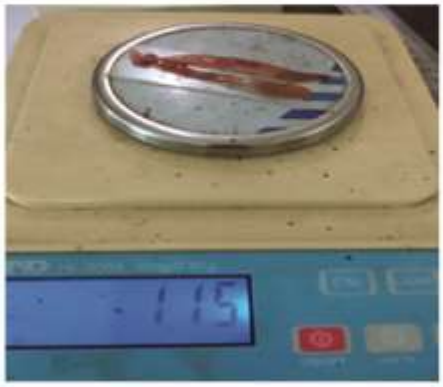


Figure 5. Collected ovary of Garua *(C. garua)*.

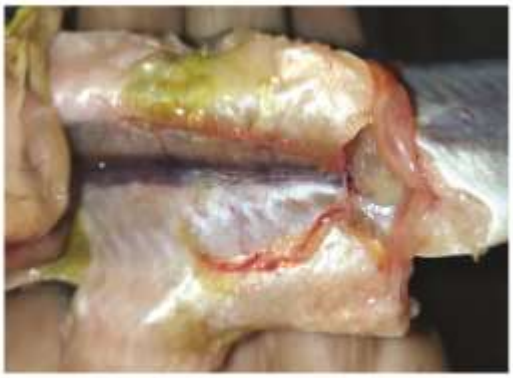


Figure 6. Position testis of Garua *(C. garua)*.

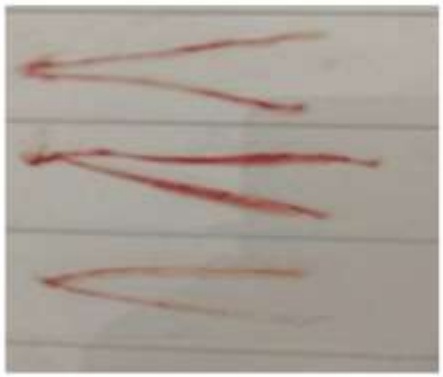


Figure 7. Collected testis of Garua *(C. garua)*.

Experiment 5. Study of the histological observation of gonad of Hiralu (B. bendelisis), Gang tengra (G. youssoufi) and Garua (C. garua)

The experiment was carried out for a period of 3 consecutive months (January to February) in the Freshwater Station, Bangladesh Fisheries Research Institute, Mymensingh. In each month, 10-15 female and male fish of Garua (*C. garua*) were collected from natural sources for the determination of histological observation to find out breeding season of the species. Histological process was performed maintaining standard protocol. The different stages were found in the ovaries of female of Garua (*C. garua*). The stages are Oogonial (O) stage; Chromatin nucleolar stage and early nucleolar stage were found in the month of January, February, and March respectively (Figure 8-10). In case of male Garua (*C. garua*) Primary germ cells and Spermatogonia stage of testis were found in February and March respectively (Figures11, 12).

Figure 8. Oogonial (O) stage in the ovaries of female, Garua *(C. garua)* in January.

Figure 9. Chromatin nucleolar stage in the ovaries of female Garua *(C. garua)* in February.

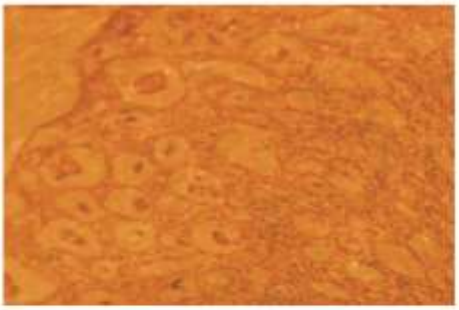


Figure 10. Early nucleolar stage in the ovaries of female Garua *(C. garua)* in March.

Figure 11. Spermatogonia stage of testis of male Garua *(C. garua)* in March.

Experiment 6. Development of induced breeding technique of T. tor

Collection and domestication of *T. tor* in pond

A total of 25 *T. tor* were collected from Sumessweri river of Netrokana district. Collected fish were transported to Freshwater Station in oxygenated drum. After transportation, the fish were acclimatized in a pond for 2 h. After acclimatization, the collected fish were stocked in a pond having an area of 40 decimals. During stocking initial length and weight of the collected fish were recorded and average body weight of collected fish was 1500±325 g. Fish are being reared by supplying commercially floating feed at of 2-3% body weight twice daily. Physico-chemical parameters of pond water *viz*. water temperature, pH, DO and ammonia are being monitored at monthly interval. Feed ration are being adjusted monthly. After maturing the fish, induced breeding technique will be carried out for seed production.

Figure 5. Sampling of *T. tor*

Experiment 5. Development of induced breeding technique of Titputi (Pethia ticto)

Induced breeding trials of *Pethia ticto* were conducted to optimize hormone doses during June 2022. In this experiment for the ovulation of female fish different dose (6, 8, 10 mg/kg body weight of fish) of PG were used to confirm the optimum dose. On the other hand, males were treated with (3, 4, 5 mg/kg body weight of fish) of PG. Single dose was applied in male and female fish. Three doses were used as three treatments T_1 T_2 and T_3. Mature male and female were collected from the stocking pond early in the morning and weighted. After 5-6 hours of conditioning, PG was used for induced breeding of *P. ticto* with different doses. Details of the effect of PG doses and corresponding data on ovulation, fertilization, hatching and survival rate are shown in the Table 6.

Table 6. PG doses (mean±sd) on Titputi (*P. ticto*)

Treatment	Mean Body weight (g)		Dose of Injection, PG) (mg/kg)		Ovulation period (hrs)	Ovulation rate(%)	Fertilization rate (%)	Hatching period (hrs)	Hatching rate (%)	Incubation Temp. (°C)
	Male	Female	Male	Female						
T_1	6.01±0.24	6.40±0.42	3	6	-	-	-	-	-	-
T_2	6.91±0.70	7.30±0.76	4	8	-	-	-	-	-	-
T_3	6.66±0.33	7.26±1.16	5	10	8-10	80.00±1.5%	55.22±8.10	14-16	35.02±12.20	30

Figure 6. Exhibition of *P. ticto*

Figure 7. Exhibition of *B. dario* to honorable secretary

Production Performance of Hairy River Prawn (*Macrobrachium rude*) with Feed and Fertilizer in Pond Condition

Researchers
Maliha Khanom, SO
Md. Rabiul Awal, SO

Objectives

- Explore the triggering factor of natural production of small prawn in pond without stocking of seeds
- Development of nursing technique of Gura chingri, *Macrobrachium dayanum*
- Development of poly-culture technique of Gura chingri, Mola (*Amblypharyngodon mola*) and Jait puti (*Puntius sophore*) fish
- To produce improved quality post larvae (PLs) of *M. rosenbergii* and distribute to the fish farmer/hatchery owners

Achievements

Experiment 1. Development of Poly-culture Techniques of Gura Chingri, Mola and Jait puti fish

The experiment was conducted with three Treatments namely T_1, T_2 and T_3 each with three replications at Freshwater Station of BFRI, Mymensingh for a period of 6 months during November 2021 to April 2022. The stocking density of small prawn, Mola and Jat puti fish were 250, 3 and 3 kg/ha, in T_1; 375, 3, and 3 kg/ha in T_2 and 500, 3, and 3 kg/ha in T_3, respectively. The initial average weight of the small prawn, Mola and Jait puti were 0.18±0.0049 g, 0.011±0.0012 g and 0.013±0.001 g, respectively for T_1; in T_2 small prawn, Mola and Jait puti were 0.21±0.004 g, 0.013±0.061 g and 0.011±0.002 g, respectively and finally for T_3 small prawn, Mola and Jait puti were 0.21±0.0065 g, 0.011±0.002 g and 0.014±0.003 g, respectively. The experimental designs are given in Table 1.

Table 1. Experimental layout of the poly-culture technique of small prawn, Mola and Jat puti fish

Treatments	Replications	Prawn and fish species	Stocking density (kg/ha)		
			Prawn	Mola	Jati puti
T_1	3	*M. rude* + Mola fish + Jait puti	250	3.00	3.00
T_2			375		
T_3			500		

Pond preparation

The ponds were equal in size and shape, depth and basin configuration including water supply facilities. The size of each pond was 300-400 m^2 each with an average depth of 1.5 m. The ponds were prepared by draining out the water. Lime was applied at the rate of 250 kg/ha. One week after lime application, the ponds were filled with water. To maintain water quality, the pond water was changed at regular intervals using water from a deep tube-well supply. Five days after fertilizer application, when the water turns green, small prawns were stocked. For increasing the primary productivity of water, 15 kg/ha. TSP and 25 kg/ha urea, MOC, and rice bran were applied at the fortnightly interval.

Collection of small prawns and fish

Small prawns, Mola and Jait puti fish were collected from the old Brahmaputra river and Fulpur Upazila, Mymensingh during September to October 2021. Healthy and vigorous small prawn and fish were collected and transported in oxy-polythene bags, then kept in cistern for acclimatization. After 6 h of acclimatization, small prawns and fish were transferred to the research ponds.

Feed and feeding

After stocking of prawns and fish, diets containing 28-32% protein were supplied with a mixture of raw materials such as fish meal (10%), mustard oil cake (30%), rice bran (40%), and wheat bran (20%) in all treatments at 8-3% of estimated body weight as per the experimental design.

Sampling

Sampling was done regularly at 30 days interval to know the growth performance of prawns and fish.

Harvesting

Harvesting was done in mid of May 2022 by dewatering after completion of the experiment.

Figure 1. Harvested chingri from the experimental pond.

Results

The experimental ponds were harvested after 180 days of culture. To evaluate the fish growth performance, weight gain (g), specific growth rate (SGR%/day), food conversion ratio, survival rate (%), individual and total production were measured after the end of the experiment. The following parameters were used to evaluate the growth performance of experimental fish:

Weight gain (g) = Mean final weight (g) - Mean initial weight (g)

$$\text{SGR (\%/day)} = \frac{\text{ln final weight-In initial weight}}{\text{Number of experimental days}} \times 100$$

$$\text{Food conversion ratio (FCR)} = \frac{\text{Feed fed (dry matter)}}{\text{Live weight gain}}$$

$$\text{Survival rate} = \frac{\text{No.of fish harvested}}{\text{No.of fish stocked}} \times 100$$

Growth performance and production of fish

The mean initial weight (g), final weight (g), weight gain (g), specific growth rate (SGR, % per day), food conversion ratio (FCR) and survival rate of fish during the study period were recorded and presented in Table 2.

Table 2. Growth performance and production (mean±sd) of Chingri and small fish

Parameters	Species	T_1	T_2	T_3
Initial mean weight (g)	*M. rude*	0.18±0.0049 ^{a}	0.21±0.0040 ^{a}	0.214±0.0065 ^{a}
	A. mola	0.02±0.0013 ^{a}	0.017±0.0042 ^{a}	0.019±0.015 ^{a}
	P. sophore	0.017±0.004 ^{a}	0.011±0.002 ^{a}	0.01±0.009 ^{a}
Final mean weight (g)	*M. rude*	0.94±0.3 ^{a}	0.92±0.21 ^{b}	0.87±0.13 ^{c}
	A. mola	3.27±0.11 ^{a}	3.14±0.01 ^{b}	3.08±0.09 ^{c}
	P. sophore	4.25±0.1 ^{a}	4.62±0.16 ^{a}	4.29±0.07 ^{a}
Mean weight gain (g)	*M. rude*	0.76±0.058 ^{a}	0.63±0.065 ^{c}	0.71±0.021 ^{b}
	A. mola	3.25±0.047 ^{a}	3.12±0.028 ^{a}	3.07±0.053 ^{b}
	P. sophore	4.23±0.032 ^{b}	4.60±0.019 ^{a}	4.28±0.113 ^{b}
Specific growth rate (%/day)	*M. rude*	0.93±0.02 ^{a}	0.76±0.018 ^{c}	0.81±0.02 ^{b}
	A. mola	3.01±0.05 ^{a}	3.02±0.042 ^{a}	3.03±0.21 ^{a}
	P. sophore	3.17±0.06 ^{a}	3.36±0.12 ^{a}	3.38±0.08 ^{a}
Survival rate (%)	*M. rude*	78.65±4.85 ^{ab}	75.6±2.16 ^{b}	83.38±3.52 ^{a}
	A. mola	81.72±6.04 ^{a}	78.7±3.2 ^{b}	78.5±5.28 ^{b}
	P. sophore	78.39±5.06 ^{b}	81.5±2.1 ^{a}	78.9±4.17 ^{b}
FCR		1.68±0.10 ^{b}	1.76±0.14 ^{a}	1.59±0.11 ^{c}
Species -wise production (g/dec./6 month)	*M. rude*	1654±54.69 ^{c}	3179±30.72 ^{b}	4807±96.14 ^{a}
	A. mola	4204±17.68 ^{a}	3889±18.5 ^{a}	3982±62.21 ^{a}
	P. sophore	5043±27.98 ^{b}	5288±52.96 ^{a}	5145±18.42 ^{b}
Total production (g/deci./6 month)		10,901	12,356	13,934

*Values in the same row having the same superscript are not significantly different ($p > 0.05$)

After 6 months of culture, the highest mean weight of chingri (0.94±0.3 g) and Mola (3.27±0.11 g) were found in T_1 Treatment; whereas the highest mean weight of Jait puti (4.62±0.16 g) was found in T_2 Treatment. Mola showed significantly lower growth performance in T_3 than T_3. On the other hand, chingri showed significantly higher ($p < 0.05$) growth performance in T_1 than T_2 and T_3. Significantly lower food conversion ratio was found in T_3 and the highest in T_1. At the end of the experiment, species-wise production of Mola and Puti were not found significantly different among the Treatments except chingri. The production of chingri was significantly higher ($p < 0.05$) in T_3 than T_1 and T_2. Total highest production of chingri and fish were recorded in T_3 (13,934 g/deci./6 months) followed by T_2 (12,356 g/deci./6 months months) and T_1 (10,901g/deci./6 months).

Water quality parameters

Physico-chemical parameters of pond water such as temperature, pH, transparency, alkalinity, and DO of all Treatments were monitored at fortnightly interval and recorded data are shown in Table 3. Water temperature of different ponds were same. The mean temperature was 24.59±2.60°C, 24.51±3.58 °C and 24.56±2.46 °C in T_1, T_2, and T_3, respectively. The mean values of pH were 7.83±0.31, 7.91±0.29 and 7.93± 0.34 in T_1, T_2 and T_3, respectively. The mean values of DO were 5.29±0.57, 5.37±0.5 and 5.64±0.36 in T_1, T_2 and T_3, respectively. The highest mean values of alkalinity were recorded in T_1 (131.2±6.0) and the lowest in T_3 (129.34±5.11). The highest mean values of ammonia were recorded in T_3 (0.002±0.0015) and the lowest in T_2 (0.0014±0.001).

Table 3. Water quality parameters (mean±sd) of experimental ponds during November-May/2022.

Parameters	Values (Mean±SD)		
Water Temp. (°C)	24.59 ± 2.60^a	24.51 ± 3.58^a	24.56 ± 2.46^a
pH	7.83 ± 0.31^a	7.91 ± 0.29^a	7.93 ± 0.34^a
DO (mg/l)	5.29 ± 0.57^a	5.37 ± 0.5^a	5.64 ± 0.36^a
Total Alkalinity (mg/l)	131.2 ± 6.04^a	129.34 ± 5.11^a	130.62 ± 6.09^a
NH_3 (mg/l)	0.019 ± 0.0012^a	0.002 ± 0.0015^a	0.0014 ± 0.001^a

* Figures in the same row having the same superscripts are not significantly different ($p \geq 0.05$)

The economic benefit analysis of polyculture system

A simple cost-benefit analysis in polyculture system of Gura chingri with Mola and Jait puti fish from one decimal ponds over a culture period of 6 months was done to estimate the return against investment and profitability that had been generated proper combination and stocking densities in polyculture of Gura Chingri with Mola and puti were summarized in Table 4. The total costs of farming (BDT/deci.) was lower in T_1 (2940) than those of T_2 and T_3. The net benefits were calculated from three Treatments as BDT 1769, 2182 and 3018 per decimal for T_1, T_2 and T_3, respectively. The BCR was found 1.60, 1.64 and 1.79 for T_1, T_2 and T_3, respectively.

Table 4. Cost and benefits analysis of Gura chingri with Mola and Jait puti fish in polyculture.

Item wise expenditure / Operational costs	T_1	T_2	T_3
A. Cost			
1. Lime, fertilizer etc.	350.00	350.00	350
2. Seeds (chingri, Mola and Jait puti fry)	700.00	980.00	1250
3.Feeds	890	1045	1200
4. Human labor, transport etc.	1000.00	1000.00	1000
Total cost	2940	3375	3800
B. Incomes			
Gura Chingri (700 tk/ kg)	1158	2079	3365
Mola (350 tk/ kg)	1472	1362	1395
Jait puti (400 tk/ kg)	2017	2116	2058
Total return	4645	5557	6818
Net Profit (B-A)	1706	2182	3018
Benefit-Cost ratio	1.57	1.64	1.79

Experiment 2. Explore the triggering factor of the natural production of small prawns in the pond without stocking of seeds

The experiment was carried out in a small pond at Freshwater Station, BFRI, Mymensingh for a period of 06 months from November/2021 to April/2022

Pond preparation

For the experiment, at first, pond was dried completely and exposed to sunlight for about 2 weeks. Thereafter, a polyethylene sheet was set on the bottom of the pond. Then 0.5 m layer of mud soils were added on the polyethylene sheet and the pond was filled up with deep tube well water. After that, the pond was prepared by lime applying at the rate of 250 kg/ha and Urea (50 kg/ha) and TSP (50 kg/ha). The water depth of the ponds maintained at 1.0 m. To maintain water quality, the pond water was changed at regular intervals using water from a deep tube-well water.

Figure 2 (A, B) Pond preparation for exploring the triggering factor of natural production of gura chingri in the pond without stocking of seeds.

Stocking and management

Small prawns (1.9-3.3 cm total length and 0.15-0.29 g weight) have been collected from the old Brahmaputra River in the month of September to October 2021. Fish have been transported to the experimental sites through plastic bags with proper aeration. The collected prawns have been stocked at the rate of 2000 g/decimal. Diets containing 28-32% protein was supplied to the prawn with a mixture of raw materials such as fish meal, mustard oil cake, rice bran and wheat bran at 05-03% of estimated body weight.

Results

About 2 kg prawn has been harvested from the pond in the month of March 2022. After that, soil, the stalks of different trees and the roots of water hyacinth of the experimental pond have been collected to observe under the microscope to find out whether any cyst of prawn eggs are available or not. Some cysts have been found in soil. After completely 10 to 15 days of drying the research pond we refilled it with deep tube-well water. Observation was done at 15 days interval to find out any existence of Gura chingri, we found some gura chingri at the 45 days after water supply in the experimental pond. So, the formation of cyst of Gura chingri was the source of triggering factor of natural production of small prawns in the pond.

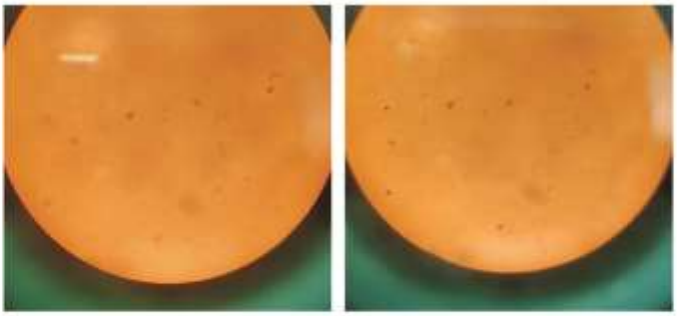


Figure 4. Observed cyst in soil under light microscope

Figure 5. Existence of gura chingri.

Water quality parameters

Physico-chemical parameters of pond water *viz*. temperature, pH, DO, and alkalinity were monitored at fortnightly interval during December-March/2021. The mean temperature was 23.25±3.04°C. The mean value of pH was 7.55±0.07. The mean value of DO was 5.43±0.06 and alkalinity was recorded 125±1.4. Water quality parameters were found suitable during the experimental period. Water quality parameters of experimental ponds are summarized in Table 5.

Table 5. Water quality parameters (mean±sd) of the experimental ponds

Parameters	Experimental pond
Temperature (°C)	23.25±3.04
pH	7.55 ± 0.07
DO (mg/l)	5.43 ± 0.06
Alkalinity (mg/l)	125±1.4

Experiment 3. Development of nursing technique of Gura Chingri, M. dayanum

The experiment was conducted into three different Treatments (T_1, T_2 and T_3) each having three replications. The experimental layout is shown in Table 6.

Experimental design

Table 6. Experimental layout of nursing of *M. dayanum* in pond

Treatments	Replications	Species	Stocking of larvae (nos./hapa)	Feeding	Nursing period (days)
T_1	3	*M. dayanum*	3000	Nursery feed (32% protein) at 20 -10% BW	90
T_2			4500		
T_3			6000		

Pond and hapa preparation

The experiment was conducted in ponds within 09 glass nylon fiber net. The hapa was equal in size and similar in shape, depth and basin configuration including water supply facilities. The size of each hapa is 2mx1mx1m. The ponds were prepared by draining out the water. Lime was applied at the rate of 250 kg/ha. After lime application, the ponds were filled up with water. The water depth was maintained to a maximum of 1.2 m. There was an inlet and out let system in each pond to maintain the water level.

Prawn stocking and management

After pond preparation, broods *M. dayanum* were stocked in T_1, T_2 and T_3 at the rate of 50, 75 and 100 nos./hapa, respectively. After stocking, supplementary feed (32% crude protein) was applied to the brooder at the rate of 3-4% of estimated body weight.

Figure 6. Collected broods of *M. dayanum*.

Nursing of *M. dayanum* in hapa

After hatching, the brood females *M. dayanum* were taken out and the newly hatched larvae were being reared at hapa. Larvae was reared in hapa about 90 days from hatching day. Golda nursery feed (32% crude protein) was applied to the hatchling at the rate of 20-10% of estimated body weight. Fortnightly sampling was carried out by a cast net to observe the prawn status.

The brood females *M. dayanum* released newly hatched larvae into the hapa and collected larvae are being reared carefully in the hapa condition. Sampling was done every 30 days interval. Growth performances are shown in Table 7.

Table 7. Growth parameters of Gura Chingri (mean±sd) in different Treatments during the experimental period.

Growth parameters	Treatments		
	T_1	T_2	T_3
Initial mean weight (g)	0.0018 ± 0.005^a	0.0019 ± 0.0030^a	0.0024 ± 0.006^a
Initial mean length (cm)	0.069 ± 0.004^a	0.065 ± 0.001^a	0.068 ± 0.006^a
Final mean weight (g)	0.49 ± 0.007^c	0.51 ± 0.023^b	0.55 ± 0.01^a
Final mean length (cm)	4.26 ± 0.098^b	4.16 ± 0.056^b	4.30 ± 0.04^a
Weight gain (g)	0.48 ± 0.02^c	0.5 ± 0.023^b	0.54 ± 0.025^a
SGR (%/day)	3.17 ± 0.12^b	3.19 ± 0.18^a	3.01 ± 0.09^c
Survival (%)	74.21 ± 4.68^c	77.15 ± 3.26^b	82.95 ± 2.01^a
FCR	1.84 ± 0.036^a	1.76 ± 0.083^b	1.63 ± 0.035^c

Values in the same row having the same superscript are not significantly different ($p > 0.05$)

After completion of the 3 months nursing period, the highest average length and weight of chingri were found (4.30±0.04 cm and 0.55±0.01 g) in T_3 and these were higher than T_1 and T_2. Significantly lower food conversion ratio (FCR) was found in T_3 and the highest value was found in T_1. SGR was significantly higher in T_2 than T_1 and T_1. Mola showed significantly lower growth performance in T_3 than T_1 and T_2. On the other hand, survival rate of chingri was significantly higher ($p < 0.05$) in T_3 than T_1 and T_2.

Water quality parameters

Water quality parameters of ponds such as temperature, pH, transparency, alkalinity and DO of all Treatments were recorded at bi-weekly intervals during the experimental period and summarized in Table 8. The mean temperature was 30.46±1.03°C, 30.19±0.73°C and 30.08±2.46°C in T_1, T_2, and T_3, respectively. The mean values of pH were 8.04±0.31, 8.07±0.21 and 8.2±0.26 in T_1, T_2, and T_3, respectively. The mean values of DO were 5.79±0.34, 5.56±0.32 and 5.77±0.15 mg/l in T_1, T_2, and T_3, respectively. The highest mean values of alkalinity were found in T_3 (136±1.89 mg/l) and the lowest was T_1 (134.7±2.37 mg/l). The highest mean values of ammonia were in T_1 (0.03±0.018 mg/l) and the lowest in T_2 (0.015±0.01 mg/l).

Table 8. Water quality parameters of the experimental ponds (March- May/2022).

Parameters	T_1	T_2	T_3
Water Temp. (°C)	30.46 ± 1.03^a	30.19 ± 0.73^a	30.08 ± 0.72^a
pH	8.04 ± 0.31^a	8.07 ± 0.21^a	8.2 ± 0.26^a
DO (mg/ l)	5.79 ± 0.34^a	5.56 ± 0.32^a	5.77 ± 0.15^a
Total Alkalinity (mg/ l)	134.7 ± 2.37^a	135 ± 1.79^a	136 ± 1.89^a
NH_3 (mg/ l)	0.03 ± 0.018^a	0.015 ± 0.01^b	0.013 ± 0.0013^b

* ঠধষঁবং রহ ŋযব ৎধসব ৎড়িি যধারহম ŋযব ৎধসব ংঁঢ়বৎংপৎরঢ়ঃ ধৎব হড়ঃ ংরমহরভরপধহঃষু ফরভভবৎবহঃ (ঢ়>০.০৫)

Experiment 4. Production of Macrobrachium rosenbergii post larvae (PLs) at the hatchery

Collection of brine water and brood Golda

Brine water (180 ppt) was collected from Pekua, Cox's Bazar. Forty-one Berried females were collected from Pirojpur (Kocha river) and transferred to the hatching tank.

PL production

About three to five thousand post-larvae were produced

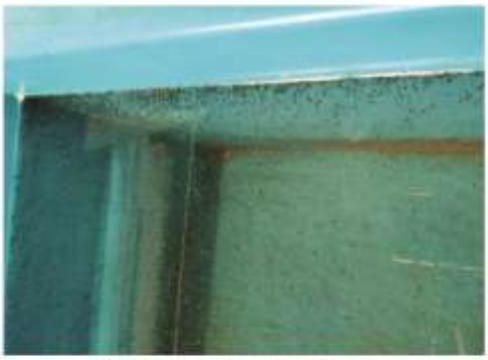


Figure 8 (A). 30 days Golda larvae.

Figure 8 (B). 60 days Golda post-larvae

Problems/constraints encountered if any

Increasing chingri production was very difficult due to their short life cycle and PL production of Golda is very challenging issue due to lack of pure brine water.

Development of YY GIFT Production Using Marker-assisted Selection and Quality Bi-sex Seed Production of GIFT Strain through Cohort Breeding

Researchers

Mohammad Ashaf-ud-Doulah, SSO
Dr. Jonaira Rashid, SSO
Md. Nahiduzzaman, SO
Nazia Naheen Nisheeth, SO
Md. Amdadul Haque, SO

Objectives

- To develop MAS-selected YY super-males of GIFT
- To produce of quality mass seed of GIFT strain using rotational breeding

Achievements

Experiment 1. YY GIFT production using Marker-assisted selection

The following steps are being followed for YY GIFT production

Pseudo female production

In 05 hapas having 2.0 m^3 have been set up in pond and single pair of mature GIFT Tilapia was stocked. Fertilized eggs were collected and kept in the hatching jar for incubation. After yolk sac absorption, 500 offspring of 5 full sib family were transferred in 5 mini cisterns. Diethylstilbestrol (DES) hormone was used at 0.50 g/kg and 1.0 g/kg feed for 21 days. The hormone treated feeds provided to the fry thrice a day up to 21 days. A total of 500 offspring of 5 full sib families each also transferred in another 05 hapa as control groups. After completed of hormone treatment, both groups of fries have been transferred to hapas in pond condition. We harvested 1420 fish from the three groups (Table 1). In group one all individuals were female 77% and group two found 350 of the 500 individuals were phenotypically female. In contrast, the female rate was only 40% in the control group. Variation in growth rates of Tilapia were found after trails (Table 2). Highest final weight gain (g) was found at T_1 (group 1). The survival rates (%) of fish were 82, 78, and 74 in T_1, T_2 and T_3, respectively (Table 2).

Table 1. Summary information on Sex Reversal of Tilapia

Group	DES (g)/feed (kg)	Individual number (n)	Female	Male	Intersex	Female rate (%)
T_1	1	500	390	90	0	77
T_2	0.5	500	350	120	0	70
Control (T_3)	N/A	500	200	270	0	40
Total			1420			

Table 2. Growth responses and survival of different treatments.

Sl no.	Treatment	Avg. length (cm)	Avg. weight (g)	% of survival
1.	T_1	22.68±0.91	196.9 ± 1.62	82
2.	T_2	20.01±0.66	158.6±1.78	78
3.	T_3	18.04±0.77	142.2±1.73	74

Hormone treatment completed for pseudo female production

2. Collection of fin sample for DNA extraction

For pseudo female identification, 10 fin samples from hormone treated fry having mean weight 36±1.68 g have been collected and kept in absolute ethanol for DNA extraction and genotyping.

Figure 1. Fin sample collection for pseudo female identification

Marker selection and PCR amplification

Sex determination (SD) marker closely linked to sex trait located on chrLG23 (forward primer. 50-TCCCATTTAGACC ACCACACCTCAACAACA-30; reverse primer. 50 -GTCAGAAT GCACTTTAACACAGAGATACCA-30; patent application no.. 2016107162044) are being used to genotype everyone. PCR amplifications are being performed and carried out in a 20-μL volume using 2× PCR master mix, 1 ng genomic DNA and 0.5μmol/L forward and reverse primers in a thermal cycler. The following program was applied (one cycle of 3 min at 94 °C, 38 cycles of 30 s at 94 °C, 30 s at 55 °C and 30 s at 72 °C, followed by a final extension of 5 min at 72 °C). The resulting PCR products were initially detected by electrophoresis with 6% agarose gel. PCR amplification of individuals during MAS using the sex-linked marker SD is shown in Figure 2.

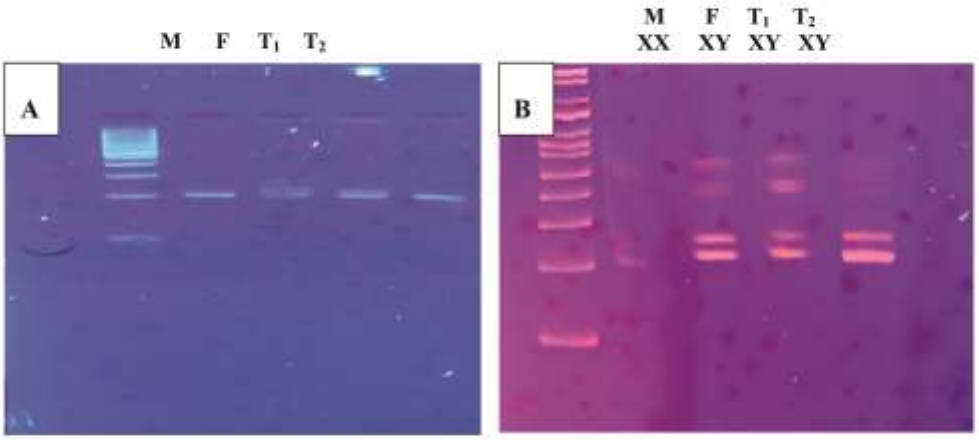


Figure 2. PCR amplification of T1 and T2 (M-Mother, F-Father, T_1 and T_2- Offspring) (a) Genotyping of individuals during marker-assisted selection using the sex-linked marker SD (M-Mother, F-Father, T_1 and T_2- Offspring) (b).

Experiment 2. Cohort breeding program of GIFT strain for quality seed production

The F-13 generations of GIFT fingerlings with a mean weight of 7.25±2.41 g were stocked in September 2021 in four (75 m^2) separate hapa in a pond having an area of 1000 m^2 area with an average depth of 1.25m for the period of five months. A total of 400 fingerlings have been stocked in each hapa for cohort breeding. The fish were fed with floating feed containing 30% crude protein at the rate of 4-8% based on body weight after stocking. After that, 200 males and 200 females were selected from each hapa for breeding. Production of 3.25 lakh fry through Cohort breeding was accomplished, out of that about 2.00 lakh fry were sold.

Development of Induced Breeding and Culture Techniques of Gangetic Endangered Fish Species

Researchers

Dr. David Rintu Das, PSO (Additional Charge)
Shishir Kumar Dey, SO
Md. Moniruzzaman, SO
Mahmudul Hasan Mithun, SO

Objectives

- Refinement of induced breeding techniques of the Batashi and Pialy.
- Determination of peak breeding season of Batashi, Pialy and Kajuli fish through gonadal histological observation.
- Development of induced breeding techniques of the Kajoli fish.
- Development of nursing and culture techniques of Batashi and Pialy.

Achievements

Experiment 1. Development of induced breeding of Neotropius atherinoides, Aspidoparia jaya and Ailia coila

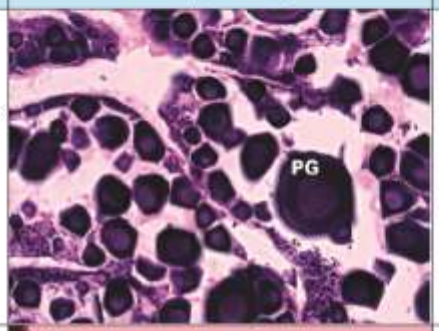
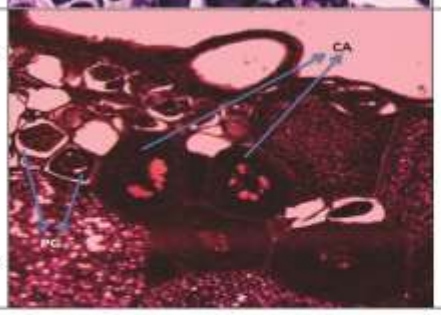
Collection of brood fish

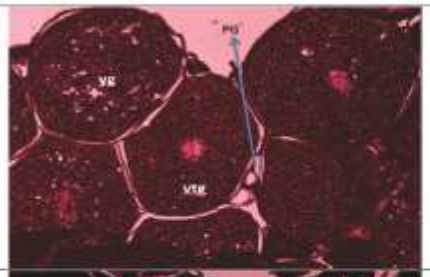
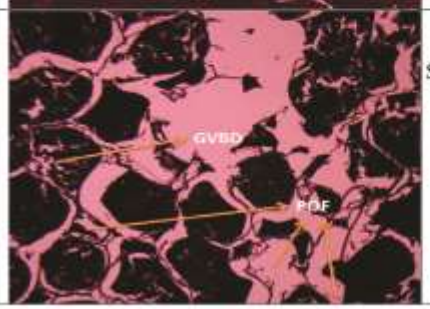
The sexually matured, strong, and diseased free broods were collected from the river Jamuna, river Atrai and Roktodaho beel of Sirajgonj, Naogoan, and Bogura districts of Bangladesh and were stocked in the ponds at the rate of 200-250 brood/decimal averaging 3-4 g each for domestication for breeding purposes.
GSI of *N. atherinoides*

For the calculation of GSI, every month about 10 fish were examined which mean body length was 7.12± 2.15 cm and mean body weight was 5.10 ± 1.95 g. In case of female *N. atherinoides*, it was found that the value of GSI gradually increased from February to April. Then it increased abruptly from April and became higher during the month of June, July, and August. Highest GSI value 13.55 ± 2.50 % was recorded in the month of July (Figure 1) which it indicated that July is the peak breeding season of *N. atherinoides*.

Figure 1. Monthly mean Gonado-somatic index (GSI) of female *N. atherinoides*.

Histological observation of *N. atherinoides*

Gonad stage	Macroscopic characteristics	Histological images	Months
Immature gonad	1. Only young females possess this stage. 2. Oogonia and PG follicles are well-organized in the ovigerous lamellae. 3. This stage does not present follicles in vitellogenesis.		April
Developing gonad	1. At this stage the females are considered entering the reproductive cycle. 2. Oogonia, PG oocytes, CA oocytes are predominant. 3. Follicles with CA and PG 4. As maturation progresses, the quantity of Vtg oocytes increases.		May and June

Mature gonad	1. Number of yolk granules was sharply increasing. 2. Mature and hydrated oocytes were numerous		June and July
Spent gonad	1. Abundant post ovulatory follicle (POFs) and germinal vesicle breakdown (GVBD) follicles.		August and September

Fecundity and Ova diameter range of *N. atherinoides*

Month	No. of fish examined	Fecundity range	Ova diameter (mm) (Mean±SD)
April	10	1100 -1800	0.10±0.01
May	10	26 70 -4200	0.21±0.03
June	10	3160 -6500	0.30±0.05
July	10	5100 -8900	0.50±0.07
August	10	2636 -5794	0.31±0.04
September	10	1300 -1900	0.15±0.02

GSI of Aspidoparia jaya

For the calculation of GSI, every month about 10 fish were examined which mean body length was 8.12±2.30 cm and mean body weight was 6.17±1.75 g. In case of female *A. jaya*, it has been found that the weight of the gonad gradually increased from April and then it increased abruptly from May and reached to a maximum value (12.15±1.50 %) in July. The higher GSI values were found in the month of June, July, August, December, and January. Two distinct peaks were observed during the month of July (12.15±1.50 %) and January (11.55±1.30 %) which indicated that they might be spawn twice in a year (from May to August and from December to January).

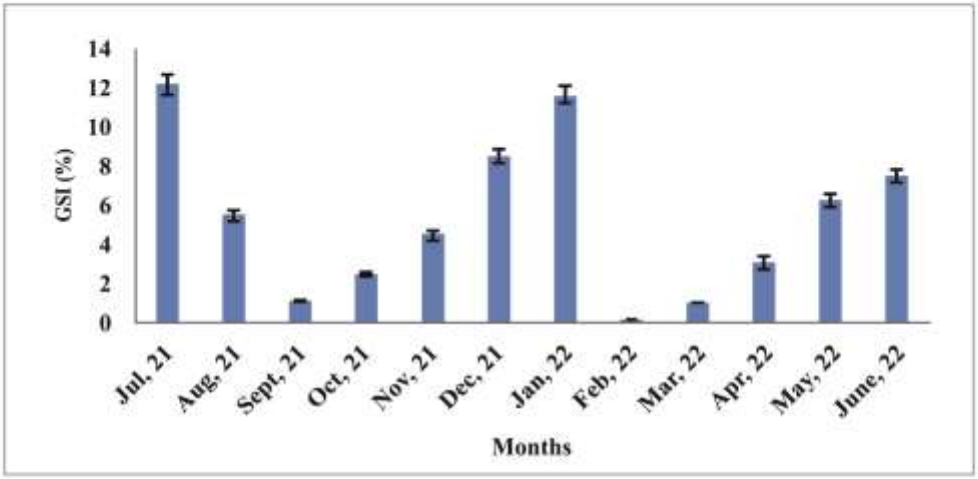
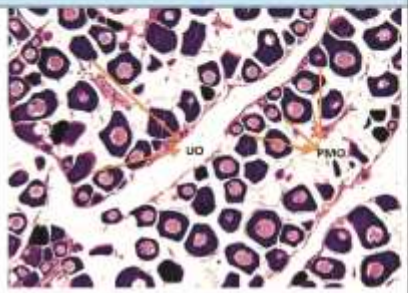
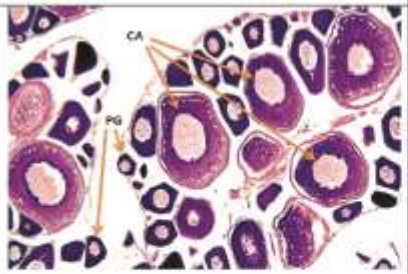
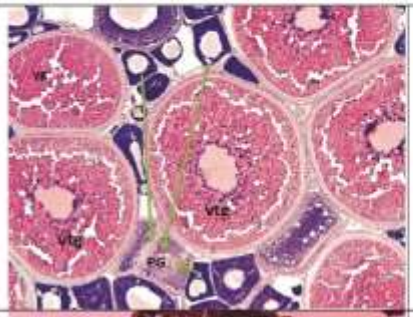
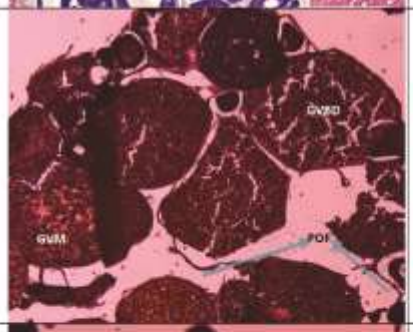


Figure 2. Monthly mean Gonado-somatic index (GSI) of female *Aspidoparia jaya.*

Histological observation of *A. jaya*

Gonad stage	Macroscopic characteristics	Histological images	Months
Immature gonad	1. Young females possess this stage. 2. Undeveloped oocyte and pre mature oocyte are well-organized 3. Oocyte mostly in CN stage, but some in PN stage. 4. This stage does not present follicles in vitellogenesis.		April
Developing gonad	1. Females are considered entering the reproductive cycle. 2. Oogonia, PG oocytes, CA oocytes are predominant. 3. As maturation progresses, the quantity of vtg oocytes increases.		May, June and December

Spawning capable gonad	1. Number of yolk granules was sharply increasing. 2. Mature oocytes were numerous		July and January
Partially spawned gonad	1. Abundant POFs and GVM follicles. 2. Initial stages of oocyte maturation may also be present, as well as GVBD follicles.		August
Post spawning gonad (Regenerating stages)	1. Post-spawning ovaries show the end of the reproductive period. 2. The "regressing" and "regenerating" stages 3. POFs absent 4. Constituted of undeveloped oocytes		September and October

Fecundity and Ova diameter range of *A. jaya*

Month	No . of fish examined	Fecundity range	Ova diameter (mm) (Mean±SD)
May	10	2100 -3560	0.22±0.01
June	10	3670 -7200	0.31±0.03
July	10	6160 -13500	0.55±0.05
August	10	1830 -3110	0.19±0.07
December	10	3970 -7300	0.35±0.02
January	10	5960 -12300	0.52±0.03

GSI of *A. coila*

For the calculation of GSI, every month about 10 fish were examined which body length were 4.12± 1.15 cm and body weight were 3.17 ± 1.05 g. Till now the highest GSI was found in the month of June and they are not matured enough for induced breeding.

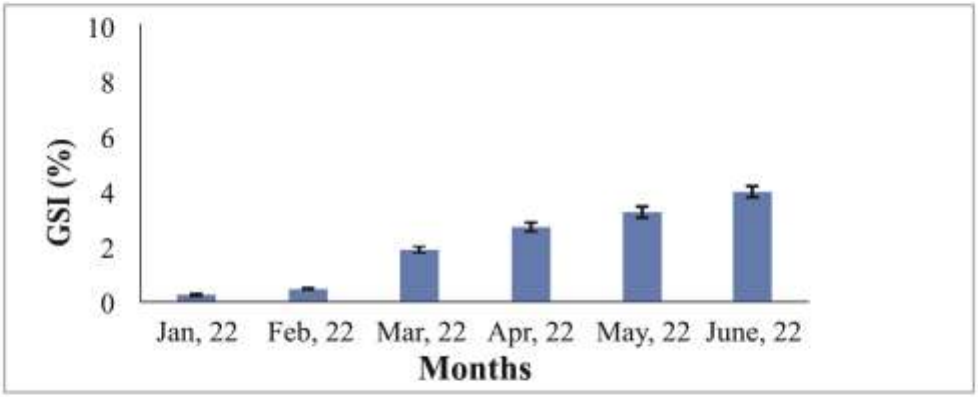


Figure 3. Monthly mean Gonado-somatic index (GSI) of female *Ailia coila*.

Histological observation of *A. coila*

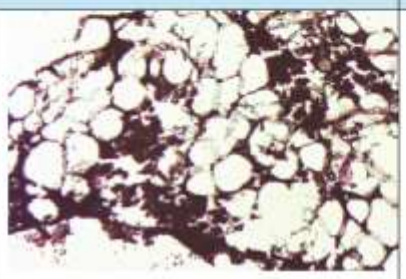
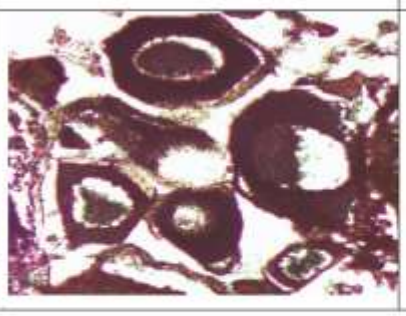
Gonad stage	Macroscopic characteristics	Histological images	Months
Immature gonad	1. Young females possess this stage. 2. Undeveloped oocyte and pre mature oocyte are well-organized 3. Oocyte mostly in CN stage, but some in PN stage.		March and April
Developing gonad	1. Females are considered entering the reproductive cycle. 2. Oogonia, PG oocytes, CA oocytes are predominant. 3. As maturation progresses, the quantity of vtg oocytes increases.		May and June

Table 2. Details of induced breeding (mean±sd) of *N. atherinoides* by applying PG doses.

Trial	Treatment	Weight of brood fish (g)		PG dose (mg/kg BW)		Latency period (h)	Ovulation rate (%)	Fertilization rate (%)	Incubation period (h)	Hatching rate (%)
		Female	Male	Female	Male					
Trial 1 (May)	T_1	5.4±0.26	4.1±0.19	14	7	12 to 15	52.12±1.21^a	59.24±1.12^a	23 to 26	65.54±2.11^a
	T_2	5.2±0.21	4.9±0.18	16	8	12 to 15	45.21±2.13^b	51.36±1.31^b	23 to 26	59.64±1.62^b
	T_3	5.6±0.25	5.3±0.20	18	9		No Ovulation			
Trial 2 (June)	T_1	7.2±0.16	5.5±0.24	10	5	12 to 15	61.12±1.21^b	59.24±1.12^b	23 to 26	75.54±2.11^b
	T_2	6.5±0.20	5.3±0.21	12	6		63.21±2.13^{ab}	61.37±1.31^{ab}		79.64±1.62 ab
	T_3	7.7±0.21	5.7±0.23	14	7		67.14±1.59^a	62.35±1.21^a		80.33±1.10^a
Trial 3 (July)	T_1	8.2±0.17	6.6±0.24	10	5	12 to 15	64.13±1.23^b	62.27±1.12^b	23 to 26	76.55±2.13^b
	T_2	7.4±0.22	7.1±0.22	12	6		66.22±2.15^{ab}	64.39±1.31^{ab}		80.65±1.64^{ab}
	T_3	7.9±0.23	6.9±0.20	14	7		70.15±1.61^a	65.37±1.21^a		81.34±1.12^a

*Values in the same row having the same superscript are not significantly different ($p > 0.05$)

The results in the present experiment indicated that induced breeding of *N. atherinoides* was successful by using different doses of PG extract and among all trials, comparatively better performances in terms of ovulation, fertilization, and hatching rates were found in Treatment T_3 in the month of July when 14 mg PG/kg body weight was applied.

Table 3. Details of induced breeding (mean±sd) of (*A. jaya*) by applying PG doses.

Trial	Treatment	Weight of brood fish (g)		PG dose (mg/kg BW)		Latency period (h)	Ovulation rate (%)	Fertilization rate (%)	Incubation period (h)	Hatching rate (%)
		Female	Male	Female	Male					
Trial 1 (May)	T_1	6.12±0.26	5.78±0.19	12	6	6 to 8	49.12±1.21	57.24±1.12	20 to 22	70.54±2.11
	T_2	6.81±0.21	5.97±0.18	14	7		No ovulation			
	T_3	6.54±0.25	5.61±0.20	16	8		No ovulation			
Trial 2 (June)	T_1	7.20±0.16	6.53±0.24	8	4	6 to 8	51.12±1.21^c	57.24±1.12^c	20 to 22	70.54±2.11^c
	T_2	7.53±0.20	7.34±0.21	10	5		56.21±1.13^b	63.36±1.01^b		74.64±1.62^b
	T_3	7.78±0.21	6.93±0.23	12	6		60.14±1.09^a	67.34±1.21^a		79.33±1.10^a
Trial 3 (July)	T_1	8.94±0.24	8.10±0.20	8	5	6 to 8	67.23±1.62^c	69.85±1.30^c	20 to 22	78.89±1.30^c
	T_2	8.79±0.26	8.36±0.29	10	6		72.59±1.80^b	74.74±1.51^b		82.48±1.12^b
	T_3	8.84±0.25	8.74±0.23	12	7		78.87±1.71^a	79.39±1.40^a		86.98±1.20^a

*Values in the same row having the same superscript are not significantly different ($p > 0.05$)

The results in the present experiment indicated that induced spawning of *A. jaya* was successful by using different doses of PG extract and among all trials, comparatively better performances interms of ovulation, fertilization, and hatching rates were found in T3 treatment in the month of July when we used 12 mg PG/ kg body weight.

Experiment 2. Development of nursing techniques of Batashi and Pialy

After breeding the five days old spawn stocking were reared with special attention for development of nursing techniques of Batashi and Pialy with different types of commercial feed and regular sampling was done to collect data on growth, FCR, etc.

Stocking of spawn

After five days of hatching, the spawn were transferred to the nursery ponds. The nursing period was 40 days. The stocking was done preferably during morning hours by acclimatizing them to the new environment. Here, the area of nursing ponds were 20 decimal in each. About 10,000 spawn/decimal were stocked in the nursing ponds. The feeding schedule during the experimental period was shown in Table 4.

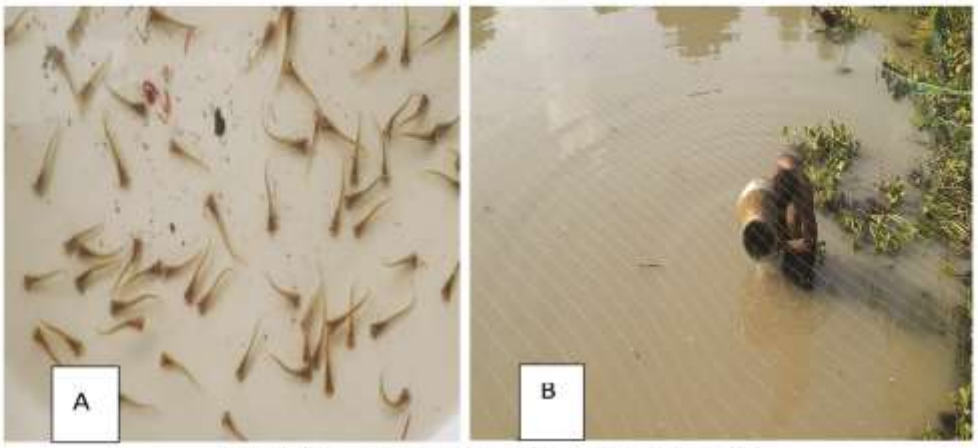


Figure 2 (A, B). Stocking of Spawn (A) after acclimatization (B).

Table 4. Feeding chart of 100 g spawn are given below.

Spawn/Fry age (Days)	Feed	Feeding rate	Feeding frequency (times/day)
1-3	Boiled egg yolk	2 egg	3
4-7	Boiled egg yolk + Flour mixing	50 g	3
8-15	Nursery feed (38-40 % protein)	100 g	3
16-30	Nursery feed (35 % protein)	200 g	3
31-40	Nursery feed (32 % protein)	300 g	3

Table 5. Water quality parameters during the nursing period.

Water quality parameters	Values	
	Mean ± SD	Range
Water Temperature (°C)	29.91 ± 1.55	28.36 - 31.46
pH	7.82 ± 0.11	7.71 - 7.93
DO (mg/l)	5.53 ± 0.38	5.15 - 5.91
Transparency (cm)	29.12 ± 1.22	27.9 - 30.34
Free CO_2 (mg/ l)	0.30 ± 0.06	0.24 - 0.36
Total ammonia (mg/l)	0.11 ± 0.05	0.06 - 0.16

Sampling

The stocked spawn were sampled four times at 10 days interval throughout the nursing period by netting to check development and physical conditions.

Growth performance

After 40 days of nursing period, the growth performance of both Batasi and Pialy was observed. In case of Batashi the average length and weight of fry were 3.1 cm and 2.3 g respectively, while in case of Pialy it was 1.8 cm and 1.9 g, respectively. The average survival rate was 73.19 % and 78.08 % in case of Pialy and Batashi, respectively.

Experiment 3. Effects of different stocking density on the growth and production of the Gangetic endangered fish species in earthen pond

Experimental Design

The trial was set up in a Completely Randomized Design (CRD) with three treatments (stocking density) having three replicates for each.

Table 6. Stocking ratio of Batashi and Pialy.

Species	Stocking density (Fry/dec)			Replication
	T_1	T_2	T_3	
N. atherinoides	500	750	1000	3
A. jaya	500	750	1000	
Total	1000	1500	2000	

Fish stocking

- 40 days old fry of Batashi and Pialy were stocked after acclimatization at 10-12 decimal sized ponds.

Feeding

Commercial floating feed of 0.3 mm size was given twice a day. Feeding adjustment was done every two weeks (25% down to 8% of BW). The amount of feeds used per treatment was recorded daily.

Sampling

The total culture period was 120 days where sampling of fish (n = 25) for length (cm) and weight (g) gain was done fortnightly, using measuring scale and digital balance. Water quality (temperature, dissolved oxygen, pH, alkalinity, ammonia) were measured using relevant equipment.

Data analysis

All the data collected during experiment were recorded and preserved on a computer spreadsheet. Growth and yield parameters of fish were analyzed statistically by one-way ANOVA and DMRT (Duncan Multiple Range Test) using the statistical software (Statistix 10).

Results and Discussion

Water quality parameters

Table 6. Water quality parameters (mean±sd) during the experimental period of 120 days.

Water quality parameters	T_1 Mean ± SD	T_2 Mean ± SD	T_3 Mean ± SD
Water Temperature (°C)	28.40 ± 1.30^a (27.10-29.70)	28.40 ± 1.30^a (27.10-29.70)	28.40 ± 1.30^a (27.10-29.70)
pH	7.50 ± 0.20^a (7.30-7.70)	7.40 ± 0.20^{ab} (7.40-7.60)	7.10 ± 0.40^b (6.70-7.50)
DO (mg/l)	5.40 ± 0.50^a (4.90-5.90)	5.30 ± 0.50^a (4.80-5.80)	5.10 ± 0.60^a (4.50-5.70)
Transparency (cm)	33.10 ± 2.10^a (31.00-35.20)	33.10 ± 2.40^a (30.70-35.50)	33.30 ± 2.30^a (31.00-35.60)
Total ammonia (mg/l)	0.10 ± 0.02^a (0.08-0.12)	0.14 ± 0.04^a (0.10-0.18)	0.17 ± 0.06^a (0.11-0.23)

*Values in the same row having the same superscript are not significantly different ($p > 0.05$)

Growth and production performance

- Results of the present study demonstrated that, no significant differences were observed among the Treatments in weight gain, SGR, and survival rates.
- A significantly higher ($p<0.05$) gross production of 13.33±0.19 kg/dec in 120 days was obtained in T_3 (2000 nos/dec) Treatment compared to T_2 and T_1.
- The results demonstrated that the polyculture of Batashi and Pialy at 2000 nos/dec stocking density may be suitable for higher growth, survival and production.

Table 7. Growth and production performace (mean±sd) of Batshi and Pialy in polyculture of 120 days.

Parameters	Species	Treatments		
		T_1	T_2	T_3
Initial length (cm)	*N. atherinoides*	2.52 ± 0.10^a	2.54 ± 0.08^a	2.56 ± 0.17^a
	A. jaya	$2.82\pm0.15_a$	2.44 ± 0.78^a	2.66 ± 0.19^a
Final length (cm)	*N. atherinoides*	8.63 ± 0.30^a	8.85 ± 0.07^a	8.99 ± 0.11^a
	A. jaya	8.93 ± 0.31^a	8.75 ± 0.57^a	8.89 ± 0.16^a
Initial weight (g)	*N. atherinoides*	0.99 ± 0.07^a	1.00 ± 0.11^a	1.01 ± 0.12^a
	A. jaya	1.02 ± 0.06^a	1.10 ± 0.11^a	1.01 ± 0.12^a
Final weight (g)	*N. atherinoides*	7.20 ± 0.07^a	7.44 ± 0.11^a	7.69 ± 0.14^a
	A. jaya	8.28 ± 0.24^a	8.22 ± 0.20^a	8.15 ± 0.17^a
Weight gain (g)	*N. atherinoides*	6.21 ± 0.03^a	6.44 ± 0.19^a	6.68 ± 0.11^a
	A. jaya	7.29 ± 0.20^a	7.22 ± 0.19^a	7.14 ± 0.11^a
SGR (% day^{-1})	*N. atherinoides*	1.66 ± 0.05^a	1.68 ± 0.10^a	1.70 ± 0.10^a
	A. jaya	1.77 ± 0.05^a	1.76 ± 0.10^a	1.74 ± 0.12^a
ADWG (g)	*N. atherinoides*	0.05 ± 0.00^a	0.05 ± 0.00^a	0.06 ± 0.00^a
	A. jaya	0.06 ± 0.00^a	0.06 ± 0.00^a	0.06 ± 0.00^a
Survival (%)	*N. atherinoides*	87.30 ± 6.60^a	85.71 ± 4.70^a	84.95 ± 5.40^a
	A. jaya	84.78 ± 4.22^a	84.65 ± 3.25^a	83.47 ± 3.48^a
Gross production (kg/dec)	*N. atherinoides*	3.14 ± 0.27^c	4.78 ± 0.20^b	6.53 ± 0.53^a
	A. jaya	3.51 ± 0.10^c	5.21 ± 0.08^b	6.80 ± 0.35^a
Total production (kg/dec)		6.65 ± 0.26^c	9.99 ± 0.30^b	13.33 ± 0.19^a
FCR		2.23 ± 0.06^a	2.17 ± 0.02^a	2.09 ± 0.06^a

*Values in the same row having the same superscript are not significantly different ($p > 0.05$)

Species Availability and Develop a Suitable Technology of Fermented Dried Fish Product (Shidol) in Floodplain Region of Bangladesh

Researchers

Shishir Kumar Dey, SO
Dr. David Rintu Das, PSO
Md. Moniruzzaman, SO

Objectives

- Survey of the available SIS species for Shidol production
- To analysis the proximate composition and nutritive value of Shidol
- To develop suitable technology of Shidol production

Achievements

Experiment. 1. Preparation of Shidol by using available SIS and ingredients found in the floodplain region of Bangladesh

i) Survey of the available SIS species and ingredients used in Shidol preparation.

We visited several local fish markets to know the availability of different SIS found in the floodplain region for the preparation of Shidol. Some locally available fish species are Puti, Taki, Kholisha, Mola, Chanda, Chela, Batashi and Dhela. Among them Puti was most available and cheap SIS. For development of suitable technology of Shidol Puti was used. In this experiment three different types of Shidol samples (BFRI-1, BFRI-2 and BFRI-3) was prepared by using different amount of ingredients and their special characters showed in Table 1.

- **Preparation of equipment required for Shidol processing**
  The equipment required for Shidol Processing like Smoking kiln, Ring tunnel fish drier, Net covered drying busket was already made and their picture are given below
- **Collection of raw materials**
  Raw materials (SIS) were collected from local fish market.
- **Dressing, Cutting, Gutting**
  After collection, raw materials (SIS) were gutted immediately. Generally, women workers are involved in dressing, cutting and gutting.

- **Salting**

  After gutting fish salting were done to protect fish from fly, insect, or their larval infestation. Sometime salts were used to get extra weight of fish. Generally, 125 g salt was used for1 kg of fish.

Figure 1. Salting of gutted fish

- **The Smoking Kiln**

  The Smoking Kiln was made with steel as a rectangular box of 105×75×45 cm^3 size. Horizontally, the chamber divided into two equal parts by placing a horizontal perforated iron net-frame and the bottom portion used as base for burning saw dust wooden logs/chips as smoke source. Temperature was maintained at 50-55^{0}C (external of Kiln) manually by controlling the outlet of the smoking chamber. During smoking operation fish were turned over in the middle period.

Figure 2. Smoking of salted fish in Smoking Kiln

- **Sun drying in ring tunnel**

  After smoking fish were sun dried (4-6 days) in ring tunnel fish dryer and net covered drying busket. Each fish turned over 8-10 times/day.

Figure 3. Sun drying of smoked fish

- **Processing of Shidol**

  At first mankochur data pulp mixed with dried Mola, Puti, Taki fish powder as required amount to make paste. After mixing, the paste was rubbed with turmeric and mustard oil and made the paste into round shaped by hand. To protect it from birds or flies during drying under sun for 8-10 days, the baskets were covered with nets.

Figure 4. Processing of Shidol

Table 1. Materials used in processing of Shidol.

Sample name	Origin/District	Materials	Special characters
BFRI-1	New	Aram greens (79.60%), Dry SIS (15.92%), Salt (1.99%), Garlic (1.99%), Ginger (0.99%), Turmeric powder (0.4%), Mustard oil (0.99%)	Salting, smoking, sun drying
BFRI-2	New	*Giant alocasia* greens (57%), Dry SIS (28%), Salt (1.99%), Garlic (7.2%), Ginger (3.6%), Turmeric powder (0.7%), Mustard oil (7.2%)	Salting, sun drying
BFRI-3	New	Aram green (79.60%), Dry SIS (15.92%), Salt (1.99%), Garlic (1.99%), Ginger (0.99%), Turmeric powder (0.4%), Mustard oil (0.99%)	Salting, smoking, sun drying
Traditional	Rangpur, Nilphamari, Kurigram, Dinajpur, Gaibanda, Lalmonirhat, Panchagarh	Aram green (70%), Dry SIS (17.54%), Salt (3.50%), Garlic (7.01%), Ginger (0.70%), Turmeric powder (0.35%), Mustard oil (0.70%)	Sun drying

Shidol samples were stored in different preservation unit and their preparation cost was shown in Table 2.

Table 2. Preservation unit and production cost of Shidol.

Sample name	Preservation methods	Qualitatively stable (month)	Sample preparation cost/market price (Tk.)
BFRI-1	Refrigeration	2	29.11 Tk/ 100 g
BFRI-2	Refrigeration	1	34.28 Tk/ 100 g
BFRI-3	In wooden ash	6	29.11 Tk/ 100 g
Traditional	Normal temperature	1	45 Tk/ 100 g

All samples were analyzed (Proximate composition) at nutrition laboratory at Freshwater Station in Mymensingh. All the data collected during experiment were recorded and preserved on a computer spreadsheet. Proximate composition of Shidol samples were analyzed statistically by one-way ANOVA and DMRT (Duncan Multiple Range Test) using the statistical software (Statistix 10). Proximate composition (% dry weight) of Shidol samples are shown in Table 3.

Table 3. Proximate composition (% dry weight) of Shidol samples.

***Shidol* Sample**	**Moisture (%)**	**Ash (%)**	**Lipid/fat (%)**	**Protein (%)**
BFRI-1	19.31±1.56^{a}	17.86±1.22^{b}	26.19±1.25^{ab}	36.82±1.29^{b}
BFRI-2	29.59±2.59b	21.48±1.09^{b}	27.33±1.55^{a}	38.97±1.32a^{b}
BFRI-3	18.78±1.29^{c}	20.44±1.35^{b}	14.26±1.02^{c}	41.98±1.05^{a}
Traditional/Local	37.36±2.98^{c}	30.18±2.21^{a}	22.40±1.98^{b}	31.48±2.01^{c}

Values in the same row having different superscript indicats significnat differences ($P < 0.05$)

Ecological Assessment of Inland Open Water Fisheries Population with Bio-physicochemical Properties to Frame EBFM Approach (Comp. B. FSS)

Researchers

Md. Moniruzzaman, SO
Dr. David Rintu Das, PSO (Additional Charge)
Md. Mehedi Hasan Pramanik, SSO
Shishir Kumar Dey, SO
Mahmudul Hasan Mithun, SO

Objectives

- To estimate population ecology and diet composition of some commercially significant inland open water fish (especially haor and beel resident fish)
- To assess bio-physicochemical properties of some selected inland water bodies (haor and beel) including seasonal variation and impact assessment of agro-chemicals level
- To assess stock and biomass of some important ecological fish groups i.e. Planktivores/Herbivores, Detrivores, Carnivores and Omnivores based on catch and CPUE data
- To formulate ecosystem-based management approach of some major inland open water bodies (especially haor and beel) with emphasizing to increase productivity, stock enhancement and conservation of the fisheries resources

(Objectives of this study comply with the sustainable development goal 14. Conserve and sustainably use the oceans, seas and marine resources for sustainable development)

Achievements

Study 1. Sampling of Bio-Physicochemical properties of inland open waters

Study Areas

- Roktodaho beel of Shantahar at Bogura and Naogaon
- Hasaigari beel of Naogaon

Collection of hydrological data

Water quality parameter such as transparency, temperature, dissolved oxygen, pH, CO_2, alkalinity, hardness, conductivity and TDS of sampling sites were recorded monthly basis.

Water quality of Roktodaho and Hasaigari beel

Surface water temperature ranged from 20 ^{0}C to 31 ^{0}C in Roktodaho beel and 21 ^{0}C to 32 ^{0}C in Hasaigari beel during the study period. The temperature decreases from July to January then increase. The pH ranged from 7.1 to 7.8 in Roktodaho beel and 7.3 to 8.1 in Hasaigari beel during the study period. pH remained in the optimum range although it was fluctuating in different months.

In the present study, DO ranged from 4.9 to 5.8 mg/l in Roktodaho beel and 4.7 to 5.7 mg/l in Hasaigari beel. Maximum DO was observed in the month of July and gradually decreased to 4.7-4.9 mg/l in the month of December. CO2 content ranged from 7.1 mg/l to 8.2 mg/l in Roktodaho beel and 8.0 to 9.1 mg/l in Hasaigari beel. It was minimum in December and maximum in July. The fluctuations of CO_2 values correspond directly with standing crop of phytoplankton. Free carbon dioxide was found to be least in winter months (Jul 21 to Dec 21) due to greater utilization of it for photosynthetic activity by the phytoplankton.

Total alkalinity content ranged from 105 mg/l to 121 mg/l in Roktodaho beel and 103 to 125 mg/l in Hasaigari beel during the study period. In the present study the lower alkalinity values were recorded during July (103-105 mg/l) which may be due to dilution effect. This indicates that water is soft. The total hardness in both Roktodaho and Hasaigari beel varied from 35 mg/l to 69 mg/l which indicated that water is soft and is suitable for drinking and irrigation purpose after the treatment.

The electrical conductivity value ranged between 202 µmhos/cm and 231 µmhos/cm in Roktodaho beel and 190 to 225 µmhos/cm in Hasaigari beel during the study period. The EC values showed marked seasonal variation being maximum during July and minimum during December. The TDS value ranged between 210 ppm to 231 ppm in Roktodaho beel and 225 to 290 ppm in Hasaigari beel. The TDS values showed marked seasonal variation being maximum during December and minimum during July. Higher values of TDS during winter season of both Roktodaho and Hasaigari beel can be attributed to decreased water volume . Transparency ranged from 28 cm to 48 cm in Roktodoho beel and 27 cm to 47 cm in Hasaigari.

Water quality standard

Almost all water quality parameters were in acceptable ranges according to Bangladesh standard which is suitable for fish and other aquatic animals (Table. 1).

Table 1. Water quality standards (Bangladesh).

Sl. No.	Parameter	BD Standard	Source
1	Water temperature (^{0}C)	30	EQs, 1997
2	Dissolved oxygen (mg/l)	6.5	DoE, 2001
3	Carbon-di-oxide (mg/l)	23	EPAUS, 1976
4	pH	8.5	EQs, 1997
5	Transparency (cm)	45	BARC/Hossain, 2011
6	Total alkalinity (mg/l)	100-200	Boyd and Tucker, 1998
7	Total hardness (mg/l)	500	DoE, 1997
8	TDS (mg/l)	1000	DPHE, BD Online
9	Conductivity (µS/cm)		

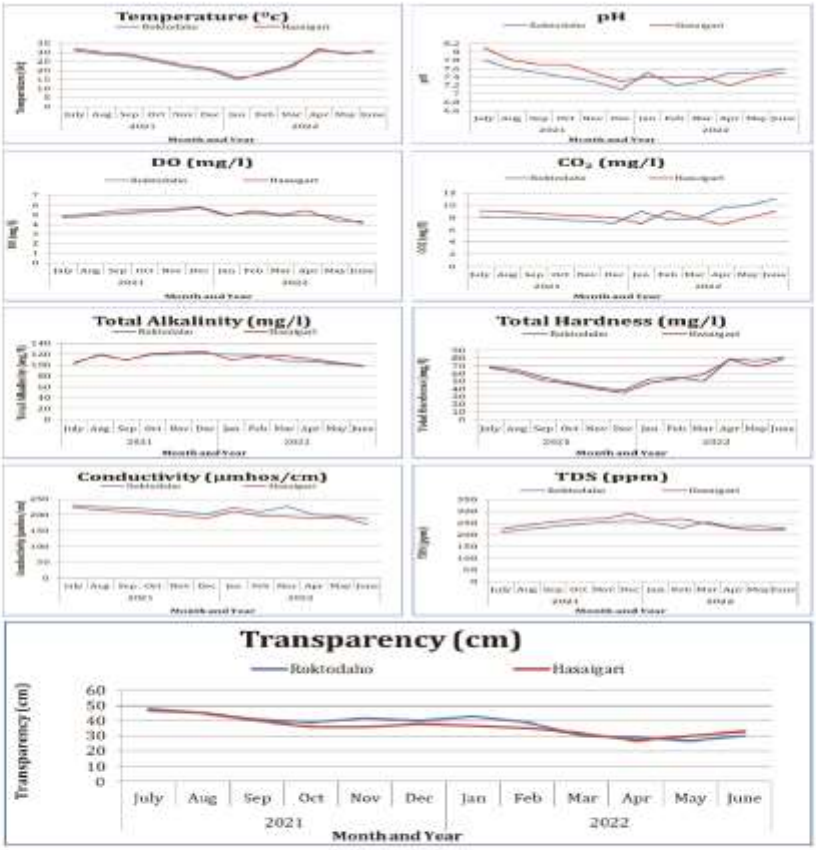


Figure 1. Water quality parameters of Roktodaho and Hasaigari *beel.*

Plankton identification

A total of 10 and 9 genera of phytoplankton and 4 and 5 genera of zooplankton were identified during the study period from Roktodoho and Hasaigari beel area. Of them, Chlorophyceae among phytoplankton and Cladocera among zooplankton population were dominant. The phytoplankton and zooplankton ratios in Roktodoho and Hasaigari beel were 87:13 and 89:11, respectively. Groupwise list of different plankton species list and nos./l of two sampling site has given on Table. 2-4.

Table 2. Group wise list of different plankton Species of Roktodoho *beel.*

Plankton Type	Plankton Groups	Genus
Phytoplankton (10)	Bacillariophyceae	*Melosira* sp. *Navicula* sp. *Asterionella* sp.
	Cyanophyceae	*Spirulina* sp.
	Chlorophyceae	*Closterium* sp. *Spirogyra* sp. *Tetraedron* sp. *Scenedesmus* sp.
	Euglenophyceae	Euglena sp. *Phacus* sp.
Zooplankton (4)	Copepoda	*Nauplius* sp.
	Rotifera	*Keratella* sp.
	Cladocera	*Brachionus* sp. *Bosmina* sp.

Table 3. Group wise list of different plankton of Hasaigari *beel.*

Plankton Type	Plankton Groups	Genus
Phytoplankton (9)	Bacillariophyceae	*Melosira* sp. *Navicula* sp.
	Cyanophyceae	*Spirulina* sp.
	Chlorophyceae	*Closterium* sp. *Spirogyra* sp. *Tetraedron* sp. *Pediastrum* sp.
	Ulvophyceae	*Ulothrix* sp.
	Euglenophyceae	*Phacus* sp.
Zooplankton (5)	Copepoda	*Nauplius* sp.
	Rotifera	*Keratella* sp.
	Cladocera	*Brachionus* sp. *Daphnia* sp. *Moina* sp.

Table 3. Assessment of plankton density.

Sampling Points	Total plankton (nos./l)	Phytoplankton (nos./l)	Zooplankton (nos./l)	Ratio (Phyto:Zoo)	Remarks
Roktodaho *beel*	56×10^4	49×10^4	7×10^4	87:13	Phytoplankton dominant
Hasaigari *beel*	61×10^4	52×10^4	9×10^4	89:11	Phytoplankton dominant

Study 2. Field data collection for estimating population ecology of commercially significant Haor and beel resident fish

Length weight range

Length-weight range of different fish species has been described in Table 4 according to the fishing gear/ trap operated in study area. This length-weight range varies seasonally (Table 4 and 5).

Table 4. Length-weight range of different fish species at Roktodoho *beel*

	Punti	Bele	Kholisa	Baim	Koi	Veda	Shing
Length (cm)	4.5-8.5	6.5-7.7	8.7-9	9-15	8-11	7-16.6	6.6-20
Weight (g)	1.5-10	2.2-3.5	13-16	2.8-10.3	8.6-23.8	9.5-63	6-57

	Napit Koi	Chanda	Taki	Tengra	Mola	Shol	Echa
Length (cm)	3.2-4.2	2.4-4.5	20-23	7.5-10.2	4-6.5	5-13.8	3-7.5
Weight (g)	0.8-1.4	2.4-1.5	78-169	4.4-7.5	0.8-2.6	1-20	0.2-4.7

Table 5. Length-weight range of different fish species at Hasaigari *beel*

	Punti	Bele	Kholisah	Baim	Koi	Veda	Shing	Mola	Tengra	Taki	Chana	Echa
Length (cm)	4.3-6	6.4-7.8	8-9	7-15	7-12	6-16	6-18	3-6.5	6.5-10	14-23	2-4.5	2-7.5
Weight (g)	1.5-9.8	2.2-3.6	10-12	2.1-10	8-24	10-65	6-49	0.5-2.7	4-7.7	44-169	2-1.5	0.1-4.7

Gear study

The various types of fishing gear/ traps are used in this study area. Types of gear used comply with the fishermen's benefit. Seine net, Cast net, Gill net and Fish trap of different mesh size are common in Roktodoho and Hasaigari beel area. Use of different fishing gear and traps can also serve as a rough indicator of the availability of different fish species. We observe that Vadai net used widely during the sampling period in those study areas. (Table 6).

Table 6. Location wise net list.

Sl. No.	Location	Net name and type
1.	Roktodoho *beel*	Fash/Current Jal (Gill net), China Doari Jal, Vadai jal, Jhaki jal (Cast net), Veshal Jal, Thela Jal, (6) Khoilsun , Vara, Chai (Fish trap) (3) Brush Shelter (Seasonal Trap) Kua (Seasonal Trap)
2.	Hasaigari *beel*	Ber jal (Seine net), Fash/Current Jal (Gill net), China Doari Jal, Vadai jal, Jhaki jal (Cast net), Veshal Jal, Thela Jal, (7) Khoilsun , Vara, Chai (Fish trap) (3) Kua (Seasonal Trap)

Study 3. Assessment of stock or biomass of commercially significant inland open water fish as well as water bodies

CPUE of different types of fishing gear

Use of different fishing gear and traps can also serve as a rough indicator of the availability of different fish species. Some gear is species selective such as gill nets, traps. We observe that Vadai net is used widely during the sampling period in those study areas. CPUE of Seine net was the highest in Roktodoho beel. The CPUE was 21-22 kg in August to October. The CPUE of Seine net at Hasaigari beel was higher at the massive level. It was 21-22 kg in August to October. There was not enough water in beel during the period from March to June. CPUE of different types of fishing gear has been presented in the following Table according to monthly basis. (Table 7-8).

Table 7. Monthly Gear-wise average CPUE (Catch Per Unit Effort) of Roktodoho beel (kg/hour/100 m net)

Net Name	Month Name												Total	Avg. (kg)
	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun		
Vadai jal/Ber jal (Seine net)	17	22	21	22	14	9	8	7	6	4	3	3.5	136	11.37
Jhaki jal (Cast net)	2	2	1	2	0.5	0.6	0.8	2	2	1.5	1	2	17.4	1.45
Fash/Current Jal (Gill net)	2	4	5	6	2	2	1	0.5	0.5	0.7	0.8	1.25	25.75	2.14
Veshal net	1.7	3	2.5	1.9	1.75	0	0	0	0	0	0.5	1	12.35	1.02
Fish trap	2.6	2.4	3.3	1.7	1.7	1.5	1.4	1.5	1.5	1.4	1.3	1.5	21.8	1.81
Brush Shelter (Seasonal)						220							220	220
Kua (Seasonal)						140							140	140

Table 8. Monthly Gear-wise Average CPUE (Catch Per Unit Effort) of Hasaigari beel (kg/hour/100 m net).

Net Name	Month Name												
	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Total	Avg. (kg)
Vadai jal/Ber jal (Seine net)	16.5	21	21	23	13	9	7.5	7	5	4	3.3	130.3	11.84
Jhaki jal (Cast net)	2	2.5	1	2.2	0.5	0.7	0.8	2.1	2.2	1.5	1	16.5	1.5
Fash/Current Jal (Gill net)	2	4.4	5	6.5	2	2.3	1	0.5	0.6	0.7	0.9	25.9	2.35
Veshal net	1.5	3	3	1.9	2	0	0	0	0	0	0.5	11.9	1.08
Fish trap	2.5	2.5	3.3	1.6	1.7	1.5	1.4	1.3	1.5	1.4	1.1	19.8	1.8
Kua (Seasonal)						150						150	150

Domestication and Conservation of Some Important Threatened Stream Fish in Northern Part of Bangladesh

Researchers
Dr. Khondaker Rashidul Hasan, PSO
Dr. Azhar Ali, PSO (Additional Charge)
Mr. Istiaque Haidar, SO
Mr. Srebash Kumar Saha, SO
Mr. Tashrif Mahmud Minhaz, SO

Objectives

- To collect the fry/semi-adult/adult of the selected fish from wild sources
- To study the reproductive biology *viz*; sex ratio, gonadosomatic index (GSI), fecundity and egg diameter of the fish
- To domesticate and brood development of the fish in captive condition
- To determine the reproductive response of the selected fish to different doses of natural and synthetic hormones in captive condition
- To develop the larvae and nursery rearing techniques of the selected fish in captive condition

Achievements

Collection of fry/fingerling/sub-adult/adult of target species

A total of 2000 fingerlings of *Barilius barila*, 400 sub-adults of *Labeo angra*, 500 sub-adults of *Chagunius chagunio*, 100 sub-adults of *Raiamas bola*, 40 sub-adults of *Labeo dero*, and 20 sub-adults of *Salmostoma bacaila* were collected from the Teesta, Atraii, Chikli and Burikhora river of Northern region of Bangladesh for the studies of reproductive biology and domestication in ponds at the Freshwater sub-station, Saidpur, Nilphamari.

Studies of reproductive parameters of *C. chagunio*

The sex ratio and gonadosomatic index (GSI) of *C. chagunio* have been studied to know the spawning season of this fish. A total of 213 fish were collected from the natural sources during July 2021 to June 2022 and studied monthly. The results are presented in Table 1.

Table 1. Sex ratio and GSI values of *C. chagunio* during July 2021 to June 2022.

Month	Individual	♂	♀	♂:♀	GSI% (♀)
July, 21	22	09	13	1.0.1.44	0.62±0.12
August, 21	23	08	15	1.0.1.87	2.18±0.23
September, 21	26	10	16	1.0.1.60	3.65±0.37
October, 21	22	09	13	1.0.1.44	3.95±0.30
November, 21	19	07	12	1.0.1.71	4.15±0.24
December, 21	19	09	10	1.0.1.11	5.24±0.20
January, 22	19	08	11	1.0.1.38	5.37±0.19
February, 22	25	11	14	1.0.1.27	6.03±0.26
March, 22	17	7	10	1.0.1.42	4.55±0.93
April, 22	9	8	1	1.0.0.13	0.73
May, 22	12	10	2	1.0.0.2	0.66±0.07

Studies of absolute and relative fecundity

The fecundity was estimated by gravimetric method. The method was used as described by Blay (1981). The fecundity was calculated using the following formula. The results are presented in Table 2.

F = (N x Gonad weight) / Sample weight

Where, F is the fecundity and N is the number of eggs in the sample.

Table 2. Mean values of relative fecundity of *C. chagunio.*

Class interval (cm)	Parameters			Fecundity (Per 100g bwt)
	TL (cm)	BW (g)	GW (g)	
15.1–18.0	17.05±1.04	67.74±24.85	2.61±1.5	3463±2639
Range	15.2-18.0	35.98-120.6	1.25-4.95	1423-7730
18.1–21.0	19.79±0.75	101.95±25.11	4.35±2.67	5586±2727
Range	18.5-21.0	69.9-182.0	1.13-12.61	1038-10607
21.1–24.0	22.1±0.87	132.09±22.96	5.03±1.42	6628±2487
Range	21.1-24.0	110.3-177.28	2.34-7.26	2848-10512
24.1–Above	29.4±2.66	352.67±106.53	14.88±7.42	19186±4226
Range	27.8-31.0	277.34-427.99	9.63-20.12	16198-22175
Mean ± SD	20.55±2.85	118.13±64.55	4.76±3.34	6201±4068
Range	15.2-31.0	35.98-427.99	1.13-20.12	1038-22175

Domestication and brood development of C. chagunio with carp in captive condition

Pond preparation and experimental design

The experiments were started at the BFRI, FSS, Saidpur for a period of 12 months, from late October 2021 to late September 2022 to observe the growth, gonadal maturation, and yield performance of *C. chagunio* in captive condition. For this experiment, the selected ponds had 15 decimals in size. The water depth was maintained at 1.5 meter. The ponds were prepared by drying, liming (1 kg/deci.) and fertilization (Urea 100

g/deci. and TSP 50 g/deci.). The fish were stocked at late October 2021 as per experimental design. The experimental design is presented in Table 3. The data on growth performances of *C. chagunio* and physico-chemical parameters of the experimental ponds were collected for 08 (eight) months and presented in Tables 4 and 5, respectively.

Table 3. Brood rearing of *C. chagunio* with carps in captive condition.

Treatment	Stocking density (indi./ha)	Stocking density (3000 indi./ha)			
	C. chagunio	Catla	Silver	Rohu	Rajpunti
T_1	3000				
T_2	4000	25% (750)	20% (600)	30% (900)	25% (750)
T_3	5000				

Table 4. Growth performance of *C. chagunio* under different stocking densities.

Parameters	Treatments		
	T_1	T_2	T_3
Culture period (days)	240	240	240
Initial length (cm)	14.1±1.0	14.7±1.3	14.2±1.2
Initial weight (g)	20.3±1.0	21.2±1.3	20.5±1.2
16^{th} sampling length (cm)	32.74±2.86	31.45±2.90	30.92±3.37
16^{th} sampling weight (g)	158.43±2.80	146.81±4.15	139.52±4.20
Weight gain (g)	138.13±2.10	125.61±3.65	119.02±3.26
SGR (%/day)	0.86±0.03	0.81±0.02	0.80±0.03
ADG (g/day)	0.58±0.02	0.52±0.02	0.50±0.02
HC (g/cm)	4.84±0.4	4.67±0.3	4.51±0.5

Table 5. Physiochemical parameters in three treatments of brood development of *C. chagunio*.

Water quality parameters	T_1	T_2	T_3
Water Temperature (°C)	26±1.0	25.67±.50	27.2±1.2
Water pH	7.50±.02	7.8±0.6	7.9±0.25
DO (mg/l)	6.2±0.10	5.9±0.2	5.3±0.24
NH_3(mg/l)	0.05±.03	0.07±.02	0.09±.03

Induced breeding of C. chagunio and S. bacaila using natural and synthetic hormone

Induced breeding of *C. chagunio* using natural and synthetic hormone

The experiment was conducted at the hatchery of BFRI, FSS, Saidpur to determine the reproductive response of *C. chagunio* using different types of hormones. The results are presented in Tables 6 and 7.

Table 6. Spawning response of *C. chagunio* to PG under natural method.

Treatments	PG (mg kg^{-1})		Latency period (hrs)	Incubation temperature (^{0}C)	% of egg release	% of fertilization	% of hatching	Remarks
	M	F						
T_1	3	6.0	-	25.73±0.23	-	-	-	No fertilization, ovulation, and spawning
T_2	4	8.0	-	26.86±0.20	-	-	-	
T_3	5	10	-	24.97±0.28	-	-	-	

Table 7. Spawning response of *C. chagunio* to synthetic hormone under stripping method.

Treatment	Ovuhom (ml/kg)		Latency period (hrs)	Incub. Temp. (°C)	% of egg release	% of fertilization	% of hatching	Remarks
	M	F						
T_1	0.2	0.5	-	18.0-24.0	-	-	-	No fertilization, ovulation and spawning
T_2	0.5	1.0	-	21.0-22.0	-	50	-	Stripping was conducted, Fertilized but not hatched

Induced breeding of *S. bacaila* using synthetic hormone

The experiment was conducted at the hatchery of BFRI, FSS, Saidpur to determine the reproductive response of *S. bacaila* using different types of hormones (See Figure 1). The results are presented in Table 8.

Table 8. Primary induced breeding success of *S. bacaila* in captivity.

Treatment	Ovuhom (ml/kg)		Latency period (hrs)	Incub. Temp. (°C)	% of egg release	% of fertilization	% of hatching	Remarks
	M	F						
T_1	0.3	0.6	-	26.5-27.0	-	-	-	No fertilization, ovulation and spawning
T_2	0.5	1.0	9.0	26.8-27.2	95	80	90	IP. 8-10 hrs; First Feeding. 36 hrs after hatching; Survival rate at 3 days after first feeding. 50%

Figure 1. Breeding operation of *S. bacaila*. (A) Injecting hormone to brood *S. bacaila* fish, (B) Hatchlings of *S. bacaila*, and (C) Hatchery produced fry of *S. bacaila*.

Development of nursery rearing technique of *Labeo angra* and *Labeo dero*

This experiment was carried out to develop the nursery rearing technique of hatchery produced *L. angra* and *L. dero* in captivity. Six mini ponds (1 decimal each) were selected for each fish species. Ponds were prepared by weeding, liming ($CaCO_3$, 1 kg/deci.) and fertilizing (TSP. 2 ppm and Urea 1.5 ppm). During culture period the fish were fed with 35% protein containing commercial feed at 25-10% of body weight. The spawn of both fish were stocked on late June 2022, at the age of 7 days after hatching and cultured up to late August 2022. The experimental design is presented in the Table 9. The growth performance of fish and water quality parameters of the nursery pond are presented in the Table 10-11 and 12-13, respectively.

Table 9. Experimental design for the nursery rearing of *L. angra* and *L. dero*.

Treatment	Stock. of spawn (g/deci.)	Culture period (days)
	L. angra/ *L. dero*	
T_1	8	
T_2	10	60
T_3	12	

Table 10. Growth performance of *L. angra* under different stocking densities.

Parameters	Treatments		
	T_1	T_2	T_3
Culture period (days)	60	60	60
Initial length (cm)	0.87±0.03	0.86±0.02	0.89±0.03
Initial weight (g)	0.0047±0.0001	0.0048±0.0001	0.0047±0.0001
Final length (cm)	4.13±0.03	4.05±0.02	3.99±0.02
Final weight (g)	1.58±0.02	1.52±0.02	1.51±0.02
Weight gain (g)	1.57±0.02	1.51±0.02	1.50±0.02
SGR (%/day)	9.67±0.08	9.60±0.07	9.62±0.08

Table 11. Growth performance of *L. dero* under different stocking densities.

Parameters	Treatments		
	T_1	T_2	T_3
Culture period (days)	60	60	60
Initial length (cm)	0.91±0.02	0.93±0.02	0.90±0.03
Initial weight (g)	0.0054±0.0003	0.0055±0.0002	0.0053±0.0003
Final length (cm)	5.46±0.07	5.12±0.06	5.17±0.07
Final weight (g)	2.03±0.03	1.97±0.03	1.98±0.03
Weight gain (g)	2.02±0.03	1.96±0.03	1.97±0.03
SGR (%/day)	9.88±0.13	9.80±0.11	9.87±0.12

Table 12. Physiochemical parameters in three treatment ponds of nursery rearing of *L. angra.*

Water quality parameters	T_1	T_2	T_3
Water Temperature (°C)	29.4±1.2	29.6±0.9	29.5±1.3
Water pH	7.50±0.06	7.60±0.07	7.50±0.08
DO (mg/l)	5.6±0.4	5.7±0.2	5.3±0.4
NH_3 (mg/l)	0.03±0.001	0.04±0.001	0.04±0.002

Table 13. Physiochemical parameters in three treatment ponds of nursery rearing of *L. dero.*

Water quality parameters	T_1	T_2	T_3
Water Temperature (°C)	29.7±1.2	29.8±0.9	29.7±1.3
Water pH	7.60±0.05	7.60±0.08	7.50±0.07
DO (mg/l)	5.9±0.4	5.7±0.3	5.6±0.3
NH_3 (mg/l)	0.02±0.001	0.02±0.001	0.03±0.002

Culture Suitability of *Barilius barila, Labeo angra* and *Colisa fasciatus* Under Polyculture in Farmers Pond of Northern Region of Bangladesh

Researchers

Dr. Azhar Ali, PSO (Additional Charge)
Mr. Md. Istiaque Haider, SSO and PI
Mr. Srebash Kumar Saha, SSO

Objectives

- To evaluate the production potentials of *Barilius barila, Labeo angra* and *Colisa fasciatus* under polyculture in the seasonal water bodies of farmer's field
- To assess the water quality parameters of cultural water bodies
- To assess the BCR of culture technologies
- To disseminate suitable culture techniques of *Barilius barila, Labeo angra* and *Colisa fasciatus* in different aqua-ecological zones in the northern part of Bangladesh

Achievements

Growth and yield of *Barilius barila* under polyculture in the farmer's ponds

For this study, a total of 6 (six) seasonal ponds were selected in 6 (six) different Upazilas of Rangpur region (Table 1) with the concern of relevant Upazilla Fishery Officer (UFO/ SUFO). The six (6) ponds were divided into three groups. Each group was treated as one treatment e.g. Treatment-I (T_1), Treatment-II (T_2), Treatment-III (T_3) and each pond was considered as one replication. The areas of ponds ranged between 12 and 15 decimal and depth between 1.0 and 1.5 m. The selected ponds were prepared by dewatering, liming and fertilization. The list of location wise selected ponds and experimental design are presented in Tables 1 and 2.

Table 1. Location wise list of selected farmer's ponds.

S.No.	Location (Upazila, district)	No. of farmer's pond	Pond area (decimal)
1	Domar, Nilphamari	01	12
2	Sadar, Nilphamari	01	14
3	Chirirbondor, Dinajpur	01	13
4	Debiganj, Panchagarh	01	15
5	Sadar, Nilphamari	01	13
6	Badarganj, Rangpur	01	12

Table 2. Polyculture of *Barilius barila* (Pattern-I) in the farmer's ponds.

Treatments	Species combination	Stocking density (indi./deci.)	Fingerlings size
T_1	Barali+Catla+Silver carp+Sarpunti+ GIFT	500+12+6+10+8	Over wintered
T_2	Barali+Catla+Silver carp+Sarpunti+ GIFT	600+12+6+10+8	
T_3	Barali+Catla+Silver carp+Sarpunti+ GIFT	700+12+6+10+8	

Pond preparation

The selected ponds were prepared by dewatering, liming, and fertilization.

Culture period

Culture period is 05 months (May- September).

Stocking of fingerlings

Average 7-10 cm size fingerlings were stocked following experimental design (Table 2, Figure 1).

A B C

Figure 1. Stocking of bairali fingerling. (A) Length measurement (B) Weighing, and (C) Stocking.

12.5 Feeding regime

Fish were fed containing 30-35% protein at 10-5% of body weight twice Daily.

12.6 Sampling

Length-weight data and water quality parameters such as temperature, water pH, dissolved oxygen (DO) and NH_3 of the experimental ponds were collected fortnightly (Figure 2).

A B C

Figure 2. Sampling of Barali in farmers pond. (A) Growth measurement (B) Water quality parameters observation, and (C) Pond observation.

Growth performance of Barali and water quality parameters under polyculture in the farmer's ponds

After 90 days, the growth performances of Barali and physico-chemical parameters of the experimental ponds are presented in Tables 3 and 4 respectively.

Table 3. Growth performances of *Barilius barila* under polyculture in the farmer's ponds.

Parameters	Treatments		
	T1	T2	T3
Stocking density of Barali (indi./deci.)	500	600	700
Culture period	90	90	90
Initial length (cm)	2.5±0.4	2.5±0.4	2.5±0.4
Initial weight (g)	1.25±0.3	1.25±0.3	1.25 ±0.3
6^{th} sampling weight (g)	10.4 ±1.20	11.2 ±1.0	10.7 ±1.10
Weight gain (g)	9.15 ±1.30	9.95 ±1.20	9.45 ±1.40
% weigh gain	732 ±15.04	796 ±20.04	756 ±16.40
SGRw (%/day)	2.35 ±0.23	2.44 ±0.23	2.38 ±0.20
ADGw (g/day)	0.80 ±0.10	0.11 ±0.20	0.105 ±0.23

Table 4. Physico-chemical parameters of the experimental ponds.

Parameters	T_1	T_2	T_3
Water Temp. (°C)	29.0±2.2	28.8±2.0	28.3±2.3
Water pH	7.4±0.20	7.5±0.24	7.7±0.30
DO (mg/l)	6.0±0.5	5.5±0.2	5.4±0.15
NH_3 (mg/l)	0.12±0.03	0.08±0.01	0.10±0.02

Culture of Indigenous Small Fish in Biofloc Aquaculture System (Component-B)

Researchers

Mrs. Maliha Hossain Mou, SSO
Dr. Azhar Ali, PSO (Additional Charge)
Mrs. Shaida Akter, SO
Mr. Tashrif Mahmud Minhaz, SO

Objectives

- To observe growth and survival rate of short cycle catfish *viz*; Shing, Magur, Pabda, Tengra under Biofloc aquaculture system
- To determine water quality parameters under Biofloc aquaculture system
- To analysis analyze the economic benefits of Biofloc system
- To disseminate the suitable culture trials of short cycle catfish under Biofloc aquaculture system in the northern part of Bangladesh

Achievements

Growth and yield performance of Pabda with different stocking densities in biofloc system

Pabda (*Ompok pabda*) were cultured with 3 different stocking densities in plastic circular tanks at BFRI, FSS, Saidpur. For conducting this experiment, three plastic circular tanks were prepared with 5 tons of water holding capacity of each. The plastic circular tanks were filled with under-ground water and then initiated the biofloc environment by adding lime (50 g/1000 L), raw salt (1 kg/1000 L), molasses (200 ml/1000 L) and probiotic (20 g/1000 L) with continuous aeration. The fingerlings of Pabda were stocked as per experimental design (Table 1) on 02 June, 2022. Culture period is 5 months (June to October). Length-weight data were collected fortnightly and water quality parameters *viz*; air temperature, water temperature, dissolved oxygen (DO), pH, ammonia and total dissolved solids (TDS) were collected once daily.

Table 1. The following experimental design in the plastic circular tank have been followed.

Treatment	Species	Stocking density (indi./m^3)	Stocking size
T_1	Pabda	600	8-10 cm
T_2		800	
T_3		1000	

Figure 1. Stocking of Pabda fingerlings.

Feeding. Fish are being fed at 5-2% BWday-1 twice daily. Locally available commercial fish feed containing 30-35% protein is being used. A pictorial view of feeding performance is showing as follows.

Figure 2. Feeding performance of pabda under biofloc system.

Growth performances of Pabda in the plastic circular tanks

The culture of Pabda is ongoing. After 75 days of rearing (Figure 3), the growth performances of Pabda and water quality parameters in the plastic circular tanks were collected. The data has been presented in Tables 2 and 3.

Table 2. Growth performances of Pabda in the plastic circular tanks.

Growth parameters	Tanks		
	T_1	T_2	T_3
Initial weight (g)	1.2±0.1	1.2±0.1	1.2±0.1
5^{th} sampling weight (g)	19.4±0.7	17.1±0.4	16.8±0.3
ADG (g/day)	0.24±0.01	0.21±0.01	0.21±0.01
SGR (%/day)	3.71±0.13	3.55±0.12	3.52±0.09

Figure 3. Length-weight observation during sampling.

Table 3. Water quality parameters in the plastic circular tanks.

Water quality parameters	Tanks		
	T_1	T_2	T_3
Air temperature (°C)	27.5±0.3	27.5±0.3	27.5±0.3
Water temperature (°C)	26.2±0.6	26.4±0.5	26.6±0.4
Water pH	7.7±0.1	7.8±0.1	7.8±0.1
DO (mg/l)	5.8±0.5	5.7±0.5	5.7±0.4
Ammonia (mg/l)	0.01±0.01	0.01±0.01	0.01±0.01
TDS (mg/l)	680±42.0	680±30.0	680±26.0
Floc density (ml/l)	11.5±0.8	12.4±0.7	11.7±0.4
C : N Ratio	15.1	15.1	15.1

Ecological Assessment of Open water Fisheries (Baor) Population with Bio-Physicochemical Properties to Frame EBFM Approach (Comp. C)

Researchers

Mohammed Ashraful Haque, SSO
Md. Shariful Islam, SSO
Anik Talukdar, SO
Md Raibul Islam, SO

Objectives

- To estimate population ecology and diet composition of some commercially important inland open water fish (especially baor resident fish)

- To assess bio-physicochemical properties of some selected baor including seasonal variation and agro-chemicals impact level
- To assess stock of some important ecological fish groups i.e., planktivores/herbivores, detrivores, carnivores and omnivores based on catch and CPUE data
- To assist for framing or formulating ecosystem-based management approach of some commercially significant inland open water bodies with emphasizing to increase productivity, stock enhancement and conservation of the fisheries resources.

Achievements

All water quality parameters were found within suitable ranges for fish culture in the Bukbhara baor of Jahore Sadar upazilla, Jhapa and Khedapara baor of Monirampur upazila, respectively (Table 1, Table 2 and Table 2).

Table 1. Monthly water quality parameters of Bukbhora *baor.*

WQ Parameters	Jul/ 21	Aug/ 21	Sep/ 21	Oct/ 21	Nov/ 21	Dec/ 21	Jan/ 22	Feb/ 22	Mar/ 22	Apr/ 22	May/ 22	June/ 22
Temperature (^{0}c)	32	32.5	30.5	30.2	26	22.3	21.5	24.3	30	32	31.9	32.3
Transparency (cm)	33	33	32	33	34	35	31	32	30	34	35	33
DO (mg/l)	5.5	5.7	5.5	5.7	5.9	6.1	6.5	6.0	6.1	6.0	6.2	5.9
CO_2 (mg/l)	7.49	7.43	7.39	7.39	7.36	7.33	7.32	7.30	7.34	7.35	7.23	7.42
pH	7.5	7.5	7.5	7.8	7.75	7.8	8.0	8.0	8.0	8.0	7.75	7.9
NH_3 (mg/l)	00	00	00	00	00	00	00	00	00	00	00	00
Total alkalinity	169	170	167	165	165	166	168	167	168	166	168	169
Total hardness	176	179	170	169	170	169	171	171	172	170	173	174

Table 2. Monthly water quality parameters of Jhapa *baor.*

WQ Parameters	Jul/ 21	Aug/ 21	Sep/ 21	Oct/ 21	Nov/ 21	Dec/ 21	Jan/ 22	Feb/ 22	Mar/ 22	Apr/ 22	May/ 22	June/ 22
Temperature (^{0}c)	32	32	30	29.8	25.5	22.2	21.6	25.9	30.5	32.5	31.7	31.5
Transparency (cm)	44	43	42	43	44	44	45	47	44	43	47	44
DO (mg/l)	5.5	5.8	5.4	5.4	5.5	6.4	6.1	6.0	6.1	6.0	6.1	5.9
CO_2 (mg/l)	8.00	8.25	7.95	7.75	7.40	7.37	7.35	7.32	7.35	7.34	7.33	7.36
pH	7.5	7.5	7.7	7.8	7.6	8.0	8.4	8.3	8.1	8.3	8.1	7.9
NH_3 (mg/l)	00	00	00	00	00	00	00	00	00	00	00	00
Total alkalinity	167	168	166	167	168	169	168	167	166	168	169	166
Total hardness	175	174	169	170	171	172	172	171	172	175	174	170

Table 3. Monthly water quality parameters of Khedapara *baor.*

Parameters	Oct/21	Nov/21	Dec/21	Jan/22	Feb/22	Mar/22	Apr/22	May/22	June/22
Temperature (^{0}c)	29.5	24.5	21.4	21.1	24.9	30.2	32.3	32.5	32.7
Transparency (cm)	43	44	42	43	40	41	43	44	42
DO (mg/l)	5.5	5.6	6.5	6.6	6.1	6.0	5.9	5.6	5.8
CO_2 (mg/l)	8.00	8.25	7.95	7.83	7.76	7.73	7.75	7.87	7.92
pH	7.8	7.5	8.7	8.3	8.4	8.2	8	8.4	8.2
NH_3 (mg/l)	00	00	00	00	00	00	00	00	00
Total Alkalinity	168	169	167	167	168	165	166	169	167
Total Hardness	170	172	170	171	172	170	172	173	171

The phytoplankton and zooplankton genera identified in all the baors are similar and belonging from seven groups and three groups, respectively (Table 4).

Table 4. Phytoplankton and zooplankton genera found in Bukbhora, Jhapa and Khedapara *baor.*

Group	Phytoplankton Genera	Total Species
Bacillariaophyceae	*Diatoma, Navicula, Nitzschia, Chaetoceros, Cyclotella, Gyrosigma, Synedra, Bacillaria, Fragilaria*	09
Chlorophyceae	*Oscillatoria, Eudorina, Gomphosphaeria, Pleodorina, Anabaena, Oedogonium, Microcystis, Ankistrodesmus, Oocystis, Chlamydomonas, Coelastrum*	11
Cyanophyceae	*Spirogyra, Anacystis, Ploycystis, Ulothrix, Volvox, Spirulina, Chlorella, Pediastrum, Microspora, Scenedesmus, Merismopedia*	11
Zygnematophyceae	*Zygnema, Staurastrum, Closterium, Euastrum, Desmidium, Mougeotia*	06
Euglenophyceae	*Euglena, Phacus, Trachelomonas*	03
Trebouxiophyceae	*Crucigenia, Actinastrum*	02
Dinophyceae	*Ceratium*	01
Total		**43**
Group	**Zooplankton Genera**	**Total Species**
Rotifera	*Brachionus, Keratella, Filinia, Asplanchna*	04
Cladocera	*Moina, Daphnia, Bosmina, Diaphanosoma*	04
Copepoda	*Cyclops, Mesocyclops, Macrocyclops, Diaptomus, Pseudodiaptomus, Nauplius*	06
Total		**14**

Month wise abundance of phytoplankton and zooplankton in Bukbhara, Jhapa and Khedapara baor are shown in Table 5. It shows that plankton abundance is higher in Jhapa baor and lower in Khedapara *baor.*

Table 5. Phytoplankton and zooplankton abundance in Bukbhora, Jhapa and Khedapara *baor.*

Name of the *baor*	Plankton density	Jul/ 21	Aug/ 21	Sep/ 21	Oct/ 21	Nov/ 21	Dec/ 21	Jan/ 22	Feb/ 22	Mar/ 22	Apr/ 22	May/ 22	June/ 22
Bukbhora	Phyto (No./l x10^2)	165	163	161	167	165	162	163	170	167	172	178	179
	Zoo (No./l x10^2)	40	38	35	37	40	41	39	46	44	45	47	46
Jhapa	Phyto (No./l x10^2)	208	203	200	205	201	202	204	216	206	218	216	219
	Zoo (No./l x10^2)	54	51	55	36	59	57	58	61	57	65	67	66
Khedapara	Phyto (No./l x10^2)	-	-	178	180	179	176	178	189	187	192	195	197
	Zoo (No./l x10^2)	-	-	43	44	45	43	42	48	45	47	49	51

In case of Bukbhora and Khedapara baor, phytoplankton and zooplankton comprises 80% and 20%, respectively. However, Jhapa baor belongs to 78% and 22% phytoplankton and zooplankton respectively. It's indicated that all of these baors are productive and good for fish production.

Table 6. Phytoplankton and zooplankton ratio in Bukbhora, Jhapa and Khedapara *baors.*

Bukbhora		Jhapa		Khedapara	
Phyto (%)	**Zoo (%)**	**Phyto (%)**	**Zoo (%)**	**Phyto (%)**	**Zoo (%)**
80	20	78	22	80	20

Among the stocked fish in Bukbhora baor, Silver carp was the dominant species. Around 1500 different fish are being stocked in Bukbhora baor with 0.5-1/kg fish per hactre. Total 187 metric ton fish are produced in this medium sized baor (153 ha area) where 44.55 metric tons are SIS and 142.20 metric tons were cultured fish and its production intensity was 1222 kg/ha (Table 7).

In Jhapa baor, Rui and Common Carp are the dominant species. Around 1800 different fish are being stocked in Jhapa baor with 1-2/kg fish per hactre. Total 429 metric ton fish are produced in this large baor (244 ha area) where 73.44 metric tons are SIS and 139.77 metric tons are cultured fish and its production intensity was 1758 kg/ha (Table 7).

In a small sized (57 ha area) Khedapara baor, Rui and Mrigal are the dominant species. Around 3100 different fish are being stocked in Khedapara baor with 0.5-1/kg fish per hactre. Total 145 metric ton fish were produced in this baor where 5.09 metric tons are SIS and 355.08 metric tons are cultured fish and its production intensity was 2544 kg/ha (Table 7). Total fish production including captured and cultured in Bukbhora, Jhapa and Jhedapara baor were 187, 429 and 145 metric tons (Table 7).

It was observed that average production in Jhapa baor is lower than Khedapara baor though Jhapa baor is around five times larger than khedapara baor. On the other hand, average production in Bukbhora baor is lower than Khedapara baor though Jhapa baor is around three times larger than khedapara *baor.*

Table 7. Overall Fish production Scenario in Bukbhora, Jhapa and Khedapara baor.

Name of the *baor*	Stocked fish ratio (per 100 fish)									Stocking density (ha^{-1})	Total area (ha)	Total production (Ton)			Production (kg ha^{-1})
	Silver Carp	Bighead Carp	Rui	Catla	Mrigal	Common Carp	Grass Carp	Black carp	Kalibaush			Captured SIS	Cultured fish	Total	
Bukbhora	45	20	7	10	4	10	2	1	1	1350-1500 (7-8/kg)	153	44.55	142.20	187	1222
Jhapa	15	5	30	5	15	25	5	0	0	1500- 1800 (10-12/ kg)	244	73.44	355.08	429	1758
Khedapara	15	1	40	2	30	10	1	1	0	2500-3100 (12-15/kg)	57	5.09	139.77	145	2544

There was only one carnivorous fish in the study area. Most of cultured fish are herbivore and omnivore and few are detrivore (Table 8 and Figure 1). In case of captured SIS, all the baors are carnivore dominat, few are herbivore and omnivore but there were no detrivore fish (Table 9 and Figure 1).

Table 8. Cultured fish classification according to their food habit in different *baors.*

Name of the *baor*	Number of Species			
	Herbivore	Carnivore	Omnivore	Detrivore
Bukbhora (9)	3	1	3	2
Jhapa (7)	3	0	2	2
Khedapara (8)	3	0	4	1

Table 9. Captured SIS classification according to their food habit in different *baors.*

Name of the *baor*	Number of Species			
	Herbivore	Carnivore	Omnivore	Detrivore
Bukbhora (18)	2	10	6	0
Jhapa (16)	1	11	4	0
Khedapara (14)	1	9	4	0

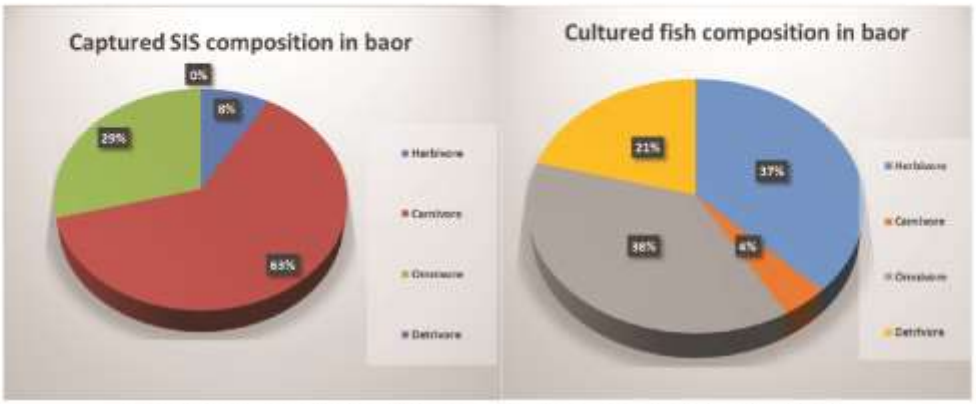


Figure 1. Captured and cultured fish composition in baor according to food habit of fish.

Different gears are used to harvest fish in baors which are nets (Table 10), traps (Table 11), hook and lines (Table 12) and wounding gears (Table 13).

Table 10. Fishing nets operated in *baors*.

Type of fishing	Gear		Description/Size		Mesh (cm)	Materials used	Nature of gear	Fishing period	Fishermen (FM) and boat required	Comments
	English name	Local name	Length (m)	Breadth (m)						
Kochal	Seine net	Ber jal	80-110	15-20	8-10	Nylon twine	Selective	Oct-Dec Feb-mar May-Jun	12-14 FM	Most widely used
Komar	Drag net	Komar jal	300-400	9-10	3-4	Nylon twine	Selective	Oct-Dec Feb-Mar May-Jun	FM depends on Komar size and group 2 boats	Most widely used
Chak	Lift net	Chak jal	8-10	7-8 (round diameter)	4-5	Cod twine with bamboo frame	Selective	Oct-Dec Feb-mar May-Jun	FM depends on Komar size	Minimum use
Cast	Cast net	Jhaki jal	8-10	8-10 (round diameter)	0.5-1.4	Nylon twine	Non-Selective	Round the year	2 FM and 1 boat	Minimum use
Lift	Lift net	Veshal jal	12-15	10-12	0.5 Centre; 1.5 Front	Nylon twine	Non-Selective	Rainy Season	1 FM and 1 boat	Minimum use
Gill netting	Gill net	Puti jal	10-12	0.5-1.0	0.5-1.0	Nylon twine	Selective	Round the year	3 FM and 1 boat	Most widely used
		Maya jal	10-12	0.5-1.0	0.5-1.0	Nylon twine	Selective		1 FM and 1 boat	
		Fash jal	10-12	2-5	5-8	Mono-filament nylon	Selective		1 FM and 1 boat	

Table 11. Fishing traps operated in *baors*.

Type of gear	Local name	Description			Mesh size (cm)	Materials used	Fishing period	Number of trap used/unit	Fisherman (FM) and boat needed
		Length (cm)	Height (cm)	Breadth (cm)					
Trap	Ramani	100-150	60-80	30-40	1.5-2.0	Split of bamboo and cane materials	Jun-Aug	5-10	FM 1 and Donga
	Arina	45	25	25	0.8	Steel wire	Jun-Aug	15-30	
	Charo	40	25	15	1.0-1.5	Steel wire	Jun-Aug	15-30	
	Ghuni	25-40	9-20	9-20	0.2-0.5	Steel wire	Jun-Aug	20-30	

Table 12. Hook and lines operated in *baors*.

Type of gear	Name of gear	Number of hooks per line of lift	Bait used/Not	Fishing period	Fisherman (FM) and boat needed
Hook and line	Dhawn borshi	Several hundred	Bait used	June-Sep	FM 1 and Donga
	Nol/Dhap borshi	Several hundred	Bait used	June-Jan	FM 1 and Donga
	Sip borshi	1 hook	Bait used	All seasons except rainy day and winter	FM 1 and with or without Donga

Table 13. Wounding gears used in *baors*.

Type of gear	Name of gear	Number of spear	Fishing period	Fisherman (FM) and boat needed	Comments
Spear/Harpoon (Wounding gear)	Koach	1	Jul-Jan	FM 1-2 and 1 boat	Poacher's generally use it
	Juti	1	Jul-Jan	FM 1-2 and 1 boat	
	Jhupi	1	Jul-Jan	FM 1 and 1 boat	
	Fulkuchi	1	Jul-Jan	FM 1 and 1 boat	

Development of Breeding and Culture Technique of Needle Fish and river Catfish

Researchers
Mohammad Ashraful Haque, SSO
Md. Shariful Islam, SSO
Anik Talukdar, SO
Md. Moshiur Rahman, SO

Objectives

- To develop breeding and larval rearing technique of Kakila fish in captive condition
- To domesticate and brood development of Bacha fish in captive condition
- To develop breeding and larval rearing technique of Bacha fish in captive condition
- To develop culture technique of Kakila and Bacha fish in captive condition
- To study the histology of the gonadal development during breeding season of Kakila and Bacha fish

Achievements

Experiment. 1. Development of the breeding and larval rearing technique of Kakila fish in captive condition

The length-weight data were collected monthly basis for the Kakila (*Xenentodon cancila*) to observe their growth performances. Randomly five Kakila fish were collected monthly by netting from the pond of the FSS, Jesshore and dissected them to estimate gonado-somatic index (GSI) (Table-1). The highest gonado somatic index (GSI) were found at the months of August 2021. Breeding trial of Kakila were attempted several times with different hormone (both PG and synthetic) doses. Finally, ovulation and induced breeding were done successfully for the first time in Bangladesh in the month of August/21 with PG hormone (Table 2). Water quality data were also collected fortnightly in every month to observe water quality parameters of the pond (Table 3). All the water quality parameters were found in suitable ranges for fish culture.

Table 1. Mean length-weight data of Kakila fish and gonad of pond at FSS, Jashore.

Trial No.	No. of pairs	Dose no.	Hormone Dose carp PG (mg/kg)		Hormone dose Ovatide/ one time (ml/kg)		Observations
			Female	Male	Female	Male	
01 (August 2021)	05	1	6.0	2.0	-	-	Ovulation and hatching success occurred
02 (August 2021)	05	1	6.0	2.0	-	-	

Table 2. Induced breeding trial of Kakila fish in FSS, Jashore.

Months	Average length (cm)	Average body weight (g)	Average gonad weight (g)	Gonado somatic index (GSI) (%)
July/2021	22.04±2.90	23.28±6.21	1.10±0.55	4.72
August/2021	22.9±1.87	26.0±2.12	1.31±0.37	5.03
September/2021	22.42±2.04	25.84±1.72	1.16±0.20	4.49
Octobe r/2021	23.28±1.62	27.24±1.68	0.94±0.10	3.40
November/2021	23.14±1.50	27.16±1.55	0.80±0.06	2.94
December/2021	18.42±1.06	15.64±1.51	0.44±0.05	2.81
January/2022	18.35± 1.31	14.61± 3.50	0.24± .08	1.62
February/2022	19.03 ±1.26	15.86 ±3.25	0.28± 0.07	1.79
March/2022	18.38±1.15	13.91±2.51	0.40±0.06	2.89
April/2022	18.44±1.13	15.57±1.66	0.46±0.05	2.95
May/2022	19.87± 2.05	18.05±3.97	0.54±0.14	2.99
June/2022	21.28±2.57	21.11±5.48	0.69+0.12	3.27

After hatching, brood were transferred to another tank immediately. First feeding was started 72 hours of hatching while yolk sac became absorbed. Meshed chicken egg yolk was supplied as first feed at 40% of body weight. Zooplankton collected from research ponds were supplied to hatchling with meshed chicken egg yolk simultaneously up to 7 days. Brine shrimp flakes were provided as supplementary feed with zooplankton. At the age of 6/7 days, fry was graded according to their size and shifted to another tank for avoiding cannibalism. After 15 days, those graded fingerlings were shifted to different hapas in the earthen pond. The eggs were about 3.5 mm (0.14 in) in diameter and were attached to plant roots, steams, leaves and hapas wall. The eggs took 4-5 days to hatch while needle fish larvae were 12-15 mm.

Figure 1. Chronologiocal photographs of induced breeding technolgy of Kakila fish (Clock wise. upper left to theright).

Table 3. Monthly average water quality parameters of Kakila fish pond water of FSS, Jashore.

Parameters	July/ 21	Aug/ 21	Sept/ 21	Oct/ 21	Nov/ 21	Dec/ 21	Jan/ 22	Feb/ 22	Mar/ 22	Apr/ 22	May/ 22	June/ 22
Temperature (˚c)	30.1± 0.14	30.7± 0.14	30.75± 0.35	31.35± 0.21	25.75± 0.35	20.4± 1.97	19.6± 0.56	21.3± 0.14	28± 1.41	31.5± 2.12	31± 1.41	32.05± 0.21
DO (mg/l)	5.0± 0.28	5.0± 0.14	5.0± 0.28	5.0± 0.14	4.45± 0.07	4.5± 0.14	5±0.14	5.7± 0.14	4.65± 0.07	4.85± 0.21	4.79± 0.07	4.95± 0.21
pH	8.0±0	8.0±0	8.0±0	8.0±0	7.75±0	7.5±0	7.5±0	7.5±0	8.0±0	8.0±0	7.95± 0.07	7.75± 0.07
Alkalinity (mg/l)	169± 1.41	165.5 ±0.7	167±1. 41	164.5± 0.70	163.5± 0.70	159.5 ±0.7	160.5± 0.70	161.5 ±0.70	164.5± 0.70	166± 0	163.5± 0.70	165± 1.41
Hardness (mg/l)	175± 1.41	172.5 ±0.7	174.5± 0.70	173.5± 0.70	171.5± 0.70	167± 1.41	164.5± 0.70	165.5 ±0.70	173± 1.41	174.5 ±0.70	172.5± 0.70	172.5± 3.54
NH_3 (mg/l)	00	00	00	00	00	00	00	00	00	00	00	00

Experiment. 2. Domestication and brood development of Bacha fish in captive condition

The length-weight data of Bacha (*Eutropiichthys vacha*) were recorded monthly to observe their growth (Table 4) and water quality parameters (temperature, DO, pH, alkalinity, ammonia, transparency) were monitored fortnightly (Table 5). All the water quality parameters were found in suitable ranges of fish culture.

Table 4. Mean length-weight data of Bacha fish of pond at FSS, Jashore.

Months	Length (cm)	Weight (g)
January/2022 (1st stock)	16.82±0.98	37.14±0.81
February/2022 (1st stock)	18.05±1.70	47.25±9.10
March/2022 (1st stock)	18.26±1.90	49.2±7.69
April/2022 (1st stock)	18.86±1.27	50.6±6.91
May/2022 (2nd stock)	10.60±0.94	9.95± 1.31
June/2022 (2nd stock)	11.0± 1.18	10.57± 2.14

Table 5. Monthly average water quality parameters of Bacha fish pond water of FSS, Jashore.

Parameters	July/ 2021	Aug/ 2021	Sept/ 2021	Oct/ 2021	Nov/ 2021	Dec/ 2021	Jan/ 2022	Feb/ 2022	Mar/ 2022	Apr/ 2022	May/ 2022	June/ 2022
Temperature (°c)	30.3± 0.21	30.65± 0.21	30.8± 0.28	31.4± 0.14	25.5± 0.42	20.55± 1.34	19.15± 0.07	21.3± 0.14	29± 1.41	33.5± 0.70	31± 1.41	32.1± 0.57
DO (mg/l)	4.95± 0.21	5.1± 0.14	5.0± 0.14	5.1± 0.14	4.5± 0.14	4.5± 0.00	4.85± 0.07	5.6± 0.14	4.7 ± 0.14	4.95± 0.07	4.75± 0.07	4.8± 0.14
pH	8.0	8.0	8.0	8.0	7.75	7.5	7.5	7.5	8.0	8.0	7.77	7.85
Alkalinity (mg/l)	168.5± 0.7	165.5± 0.70	165.5± 0.70	164± 1.41	163.5± 2.12	161± 1.41	160.5± 0.70	161.5± 0.7	165.5± 0.70	166.5± 0.70	163.5± 0.70	164± 1.41
Hardness (mg/l)	177± 2.82	173± 1.41	173.5± 0.70	172± 1.41	171± 1.41	167.5± 0.70	165± 1.41	165.5±. 70	172.5± 2.12	175.5± 0.70	171± 1.41	171.5± 0.70
NH_3 (mg/l)	00	00	00	00	00	00	00	00	00	00	00	00

Experiment. 3. Histology of the gonadal development during breeding season of Kakila and Bacha fish

Fish were collected from both wild and captive source. Gonado Somatic Index (GSI) of Kakila and Bacha fish were estimated monthly. Collected Gonad were preserved and send to BFRI Shrimp Research Station, Bagerhat for histological study.

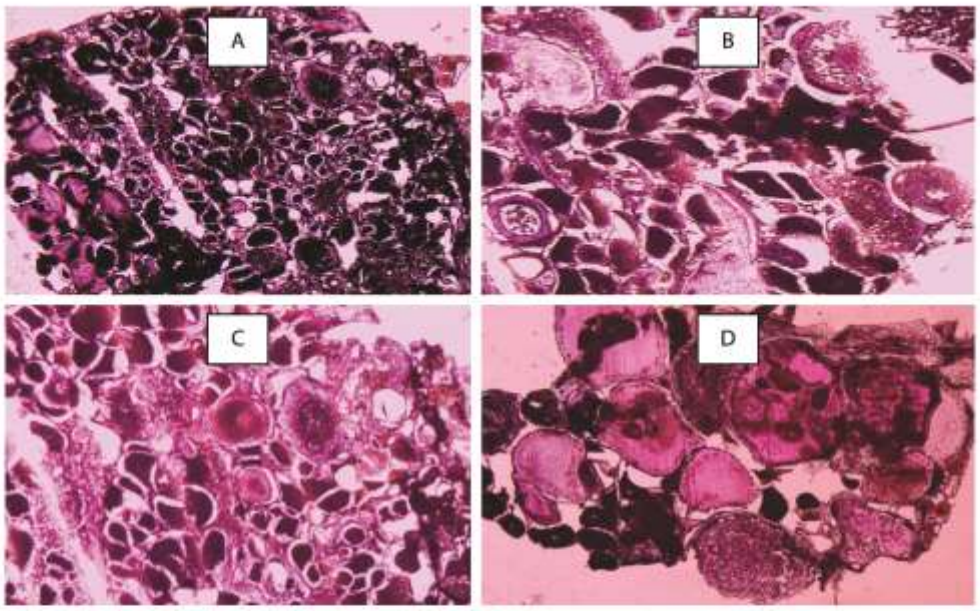


Figure 1. Transverse sections through ovaries of *E. vacha* illustrating oogenesis. A) oogonia and chromatin nucleolar stage oocytes in April; (B) early perinucleolar stage in early May; (C) late perinucleolar stage oocytes in late May; (D) yolk vesicle stage oocyte in June.

Assessment of Effectiveness of Existing Hilsa Sanctuaries for Sustainable Production in Bangladesh

Researchers

Tayfa Ahmed, SSO
Kazi Belal Uddin, SSO
Rumana Yasmin, SSO
Mezbabul Alam, SO

Objectives

- Reassess the previously identified spawning and nursery grounds and
- Identification of new spawning and nursery grounds of Hilsa

Study-1. Reassess the previously identified spawning and nursery grounds

Research was conducted on the following spawning and nursery grounds.
Lower Meghna estuary (From Char Alexander to Shahbazpur Channel, 90 km)

Physico-chemical parameters

Mean values and ranges of physico-chemical parameters over the study period has been presented in Table 1. Physico-chemical parameters such as air and water temperature (°C), transparency (cm), DO (mg/l), CO_2 (mg/l), pH, total hardness (mg/l), and total alkalinity (mg/l) were determined. The air and water temperature of the study areas were found to vary from 19 to 31°C and 17 to 30.8°C, respectively. Dissolved oxygen and free CO_2 ranged between 4.5 and 9.28 mg/l and 7.2 and 15 mg/l, respectively. The study areas pH, transparency and salinity varied from 7.5 to 8.4, 8.1 to 91cm and 1.32 to 8.49 ppt, respectively. Salinity intrusion was seen in Shahbazpur Channel from December to March within the average ranges of 0.3 to 1.62 ppt. Alkalinity and hardness ranged from 56 to 132 mg/l and 63 to 230 mg/l during the study period. The results of the physico-chemical parameters indicated that the parameters were within the suitable ranges for fish in study areas.

Table 1. Physico-chemical parameters of Shahbazpur Channel, Meghna river.

Parameters	Shahbazpur Channel	Standard value
Air temperature (°C)	27.8±0.8 (19-31)	20-30 (EQS,1997)
Water temperature (°C)	26.1±0.7 (17-30.8)	20-30 (EQS,1997)
DO (mg/l)	5.969±0.05 (4.5-9.28)	4-6 (EQS,1997)
CO_2 (mg/l)	9.94±0.5 (7.2-15)	6 ppm or less (EQS,1997)
pH	7.75±0.01 (7.5-8.4)	6.5-8.5 (EQS,1997)
Transparency (cm)	12.61±0.39 (8.1-91)	40 or less (Rahman,1992)
Salinity (ppt)	0.96±0.66 (1.32-8.49)	0-10 (Rahman,1992)
Alkalinity (mg/l)	71.2±0.6 (56-132)	>100 (Rahman,1992)
Hardness (mg/l)	96.2±7.2 (63-230)	40-400 ppm (Boyd,1998)

Plankton identification

Following the drop count method, qualitative and quantitative analysis of planktons were done (APHA 1995). Plankton identification was made following Ward and Whipple (1959) and Prescott (1962). Ten plankton groups were identified in the qualitative study of plankton, among them seven were phytoplankton and three were zooplankton groups (Table 2). Among the seven phytoplankton groups, 22 genera were identified. Bacillariophyceae, Zygnematophyceae and Chlorophyceae were the most dominant groups. But in the case of three zooplankton groups, almost six different genera were observed, including the same proportion.

Table 2. Identified Plankton groups of Shahbazpur Channel, Meghna river.

Group	Genus
Chlorophyceae	*Pediastrum, Volvox, Scenedesmus, Acanthocystis*
Ulvophyceae	*Ulothrix*
Zygnematophyceae	*Spirogyra, Nitzschia, Netrium, Staurastrum(end), Gonatozygon*
Bacillariophyceae	*Navicula, Gomphonema, Asterionella, Diatoma, Frustulia, Stephanodiscus, Cyclotella*
Cyanophyceae	*Spirulina, Rivularia, Oscillatoria*
Dinophyceae	*Ceratium*
Euglenophyceae	*Euglena*
Copepoda	*Cyclops, Nauplius*
Rotifera	*Brachionus, Keratella*
Cladocera	*Daphnia, Bosmina*

The quantitative study of phytoplankton observed a higher amount on the lower side of the Meghna river than on the upper side (Table 3).

Table 3. Quantitative assessment of plankton in Shahbazpur Channel, Meghna river.

Sampling sites	Total Plankton (cells/l)	Phytoplankton (cells/l)	Zooplankton (cells/l)
Tajumuddin	29×10^2	24×10^2	5×10^2
Ilisha Ghat	43×10^2	35×10^2	8×10^2
Daulotkhan	41×10^2	34×10^2	7×10^2

Spawning Success

In 2021, the spawning success recorded 56-89% before and after the banning period, and it was 85-97% during the 22 days banning period (04-25 October 2021). The average spawning success was 83.40 % across the sanctuary and breeding grounds.

Spent rate of Hilsa

Spent hilsa percentage was recorded at 51.76% in 2021, which was higher than the previous years.

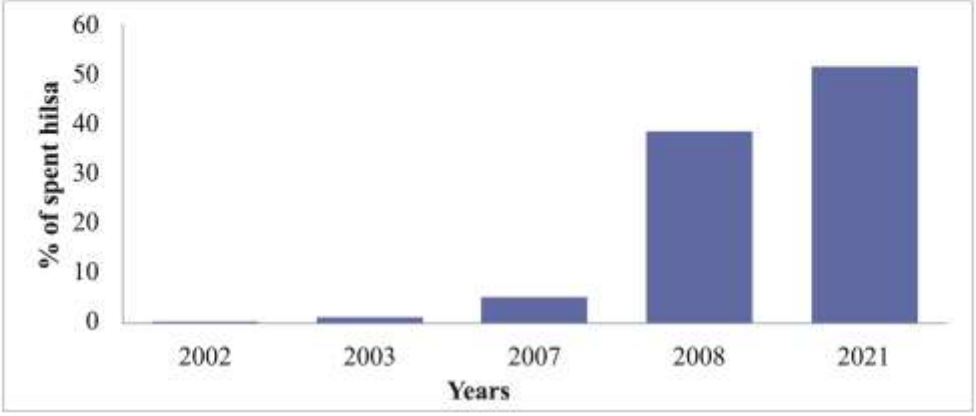


Figure 1. Spent rate (%) of Hilsa in Meghna river.

CPUE of Hilsa at Shahbazpur Channel

Catch per unit effort of Hilsa at Shahbazpur Channel of Meghna river were observed in higher amounts (5-15 kg/ 100 m net/ hour/ boat) in September and October at Shahbazpur Channel in the lower Meghna river.

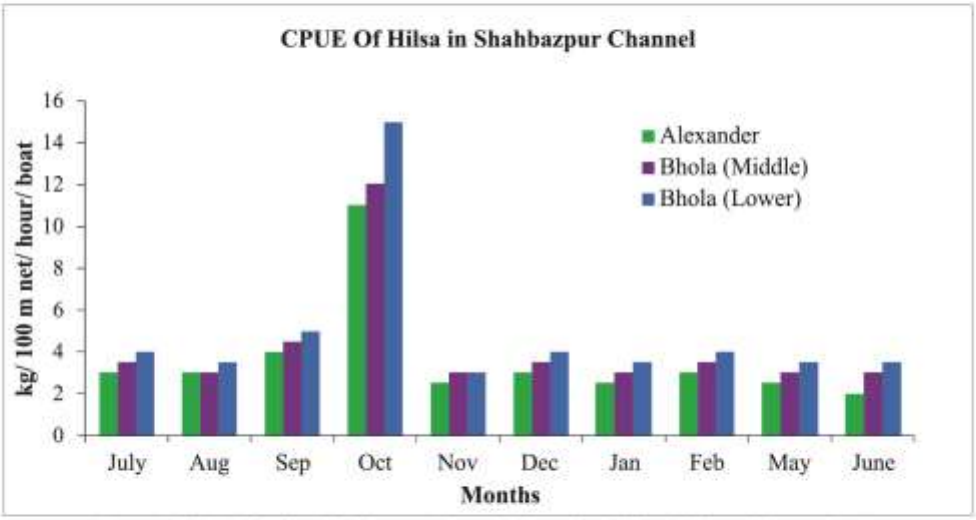


Figure 2. CPUE of Hilsa in Shahbazpur Channel (Alexander, Bhola middle and lower region).

Abundance of Jatka in the Meghna river

The average abundance of Jatka in the Meghna river was observed in a higher amount in Padma Meghna confluence (259 no. / 100 m net/ hour/ boat) and Bhola (260 no./ 100 m net/ hour/ boat) region.

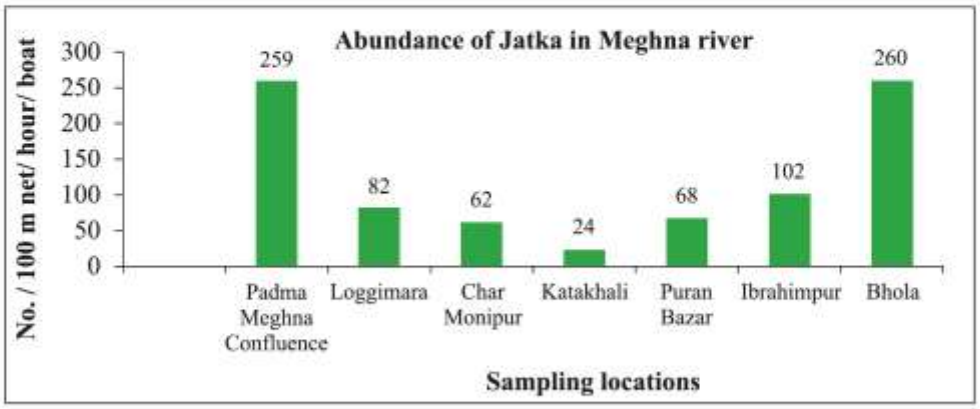


Figure 3. Abundance of Jatka in Meghna river.

Study-2. Identification of New Spawning and Nursery grounds of Hilsa

Sampling was conducted from Noria (Sureswar) to Maowa Ghat (25 km) of Padma river for the extension of 5th Hilsa sanctuary.

Physico-chemical parameters

Mean values and ranges of physico-chemical parameters over the study period has been presented in Table 4. Physico-chemical parameters such as air and water temperature (°C), transparency (cm.), DO (mg/l), CO_2 (mg/l), pH, total hardness (mg/l), and total alkalinity (mg/l) were determined. The air and water temperature (°C) of the study areas were found to vary from 19 to 32°C and 17 to 30°C, respectively. Dissolved oxygen and free CO_2 ranged between 4.6 and 6.8 mg/l and 7 and 16 mg/l, respectively. The study areas pH, transparency and salinity varied from 7.8 to 8.57, 26 to 35 cm and 0 ppt, respectively. Salinity intrusion was not observed in the Padma river during the study period. Alkalinity and hardness ranged from 48 to 58 mg/l and 60 to 92 mg/l during the study period. The analytical results of the physico-chemical parameters indicated that the parameters were within suitable ranges for fish in the study areas.

Table 4. Physico-chemical parameters of Padma river (Mawa Ghat and Sureshwar).

Parameters	Padma river	Standard value
Air temperature (^{o}C)	28.6±2.19 (19-32)	20-30 (EQS,1997)
Water temperature (^{o}C)	26.86±2.81 (17-30)	20-30 (EQS,1997)
DO (mg/l)	4.86±0.19 (4.6-6.8)	4-6 (EQS,1997)
CO_2 (mg/l)	10.40±2.60 (7-16)	6 ppm or less (EQS,1997)
pH	8.24±0.23 (7.8-8.57)	6.5-8.5 (EQS,1997)
Transparency (cm)	31.60±2.50 (26-35)	40 or less (Rahman,1992)
Alkalinity (mg/l)	50.40±2.07 (48-58)	>100 (Rahman,1992)
Hardness (mg/l)	72±4.06 (60-92)	40-400 ppm (Boyd,1998)
Salinity (ppt)	0	0-10 (Rahman,1992)

Plankton identification

Following the drop count method, qualitative and quantitative planktons were analyzed (APHA 1995). Plankton identification was made following Ward and Whipple (1959) and Prescott (1962). In the qualitative study among nine plankton groups six phytoplankton and three zooplankton groups (Table 5) were identified. Among the six phytoplankton groups, 21 genera were identified where Bacillariophyceae, Zygnematophyceae and Chlorophyceae were the most dominant groups. But in the case of zooplankton groups, four different genera were observed where Cladocera was the dominant group.

Table 5. Identified Plankton groups of Mawa Ghat, Padma river.

Group	Genus
Chlorophyceae	*Pediastrum, Volvox, Scenedesmus, Acanthocystis*
Ulvophyceae	*Ulothrix*
Zygnematophyceae	*Spirogyra, Nitzschia, Netrium, Staurastrum(end), Gonatozygon*
Bacillariophyceae	*Navicula, Gomphonema, Asterionella, Diatoma, Frustulia, Stephanodiscus, Cyclotella*
Cyanophyceae	*Spirulina, Rivularia, Oscillatoria*
Dinophyceae	*Ceratium*
Copepoda	*Nauplius*
Rotifera	*Keratella*
Cladocera	*Bosmina, Moina*

The quantitative study of phytoplankton observed a higher amount in Sureshwar (40×10^2 cells/ l) of Padma river than in Mawa ghat (32×10^2 cells/ l) (Table 6).

Table 6. Quantitative assessment of plankton in Padma river.

Sampling sites	Total Plankton (cells/l)	Phytoplankton (cells/l)	Zooplankton (cells/l)
Mawa	32×10^2	26×10^2	6×10^2
Sureshwar	40×10^2	32×10^2	8×10^2

CPUE of Jatka in Padma river.

The average CPUE of Jatka in the Padma river was recorded at 4.75 kg/100 m net/ hour/ boat, which is slightly more than the previous years.

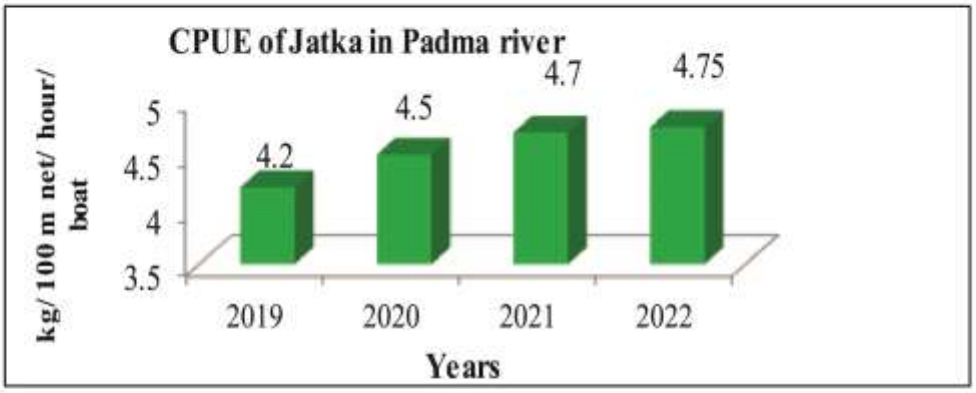


Figure 4. CPUE of Jatka in Padma river.

Ecological Assessment of Inland Open Water Fisheries Population with Bio-physicochemical Properties to Frame EBFM Approach (Comp-D)

Researchers

Md. Mozzammel Hoque, SSO
Kazi Belal Uddin, SSO
Mst. Subrina Khatun, SSO

Objectives

- To estimate population ecology and diet composition of some commercially significant inland open water fish (especially haor and beel resident fish)
- To assess bio-physicochemical properties of some selected inland water bodies (haors and beels) including seasonal variation and impact assessment of agro-chemicals level
- To assess stock and biomass of some important ecological fish groups i.e. Planktivores/Herbivores, Detrivores, Carnivores and Omnivores based on catch and CPUE data
- To formulate ecosystem-based management approach of some major inland open water bodies (especially haors and beels) with emphasizing to increase productivity, stock enhancement and conservation of the fisheries resources

Achievement

Study Areas

Haors

- Pagner Haor of Sunamgonj and
- Gongajuri haor of Hobigonj

Beels

- Padoarper beel of Chandpur

Study 1. Sampling of Bio-Physicochemical properties of inland open waters

Collection of hydrological data

Water quality parameter such as transparency, temperature, dissolved Oxygen, pH, CO^2, alkalinity, hardness, ammonia, conductivity and TDS of sampling site was recorded monthly.

Water quality of Padoarper beel.

Water quality parameters were collected monthly from Padoarper beel, Chandpur. Almost all water quality parameters were within acceptable range for fish growth. Some exception was observed in case of transparency. Highest transparency was 49 cm in the month of November. Ammonia was present in fewer amounts (0.1 mg/l) in the month of December.

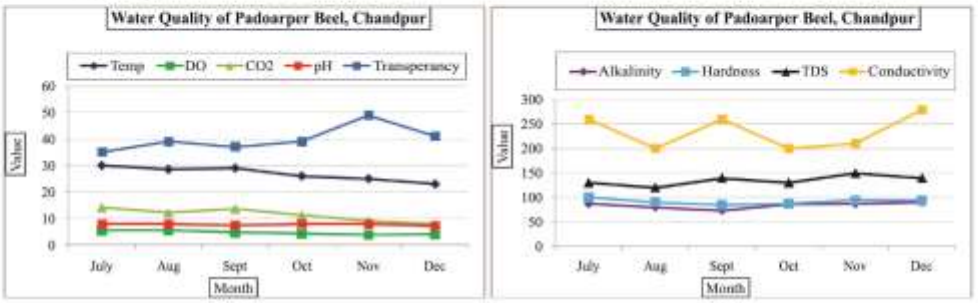


Figure 1. Water quality parameters of Padoarper beel at Chandpur.

Water quality of Pagner and Gongajuri haor

Water quality parameters were collected monthly from different sampling spot of Pagner and Gongajuri haor. Almost all water quality parameters were within acceptable range for fish growth. Some exception was observed in case of alkalinity and hardness among the sampling spot. Highest value of alkalinity was 90 mg/l and the highest value of Hardness was 94 mg/l. Lowest value of alkalinity was 14 mg/l. Lower values of alkalinity and hardness indicating beel water to be fewer nutrient enriched. Some exception was also observed in case of transparency among the sampling spot. This result may be due to the vice versa relationship among planktonic community and fish density of a waterbody. Ammonia was not found during the sampling period in Pagner haor. But in casse of Gongajuri haor, very less amount such as 0.1 mg/l was present in Ramnager *beel.*

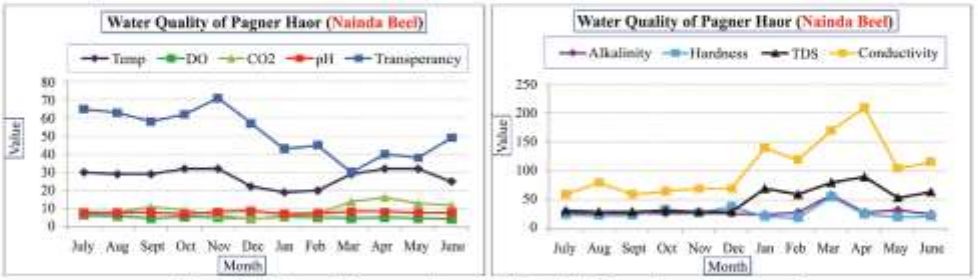


Figure 2. Water quality parameters of Nainda *beel,* Pagner *haor,* Sunamgonj.

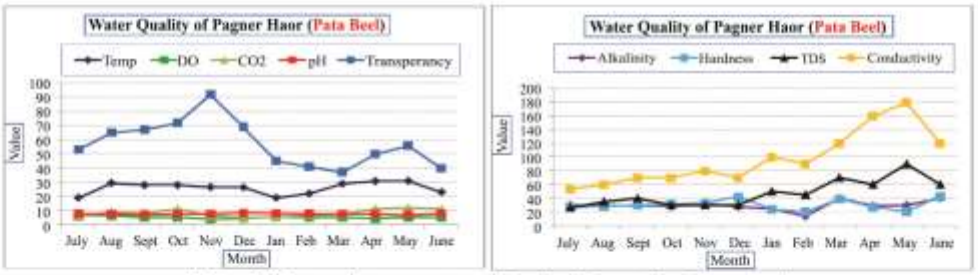


Figure 3. Water quality parameters of Pata *beel,* Pagner *haor,* Sunamgonj.

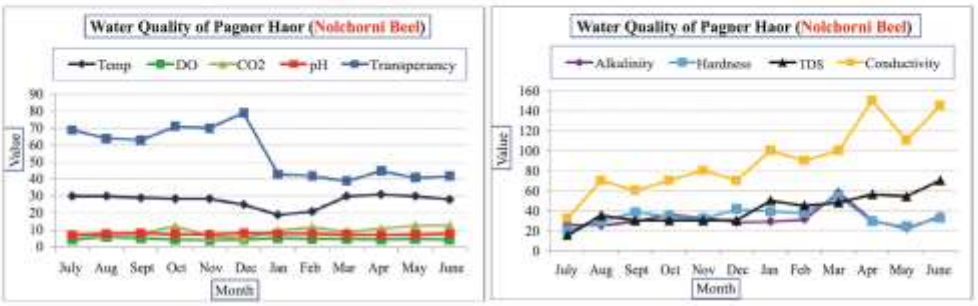


Figure 4. Water quality parameters of Nolchorni *beel,* Pagner *haor,* Sunamgonj.

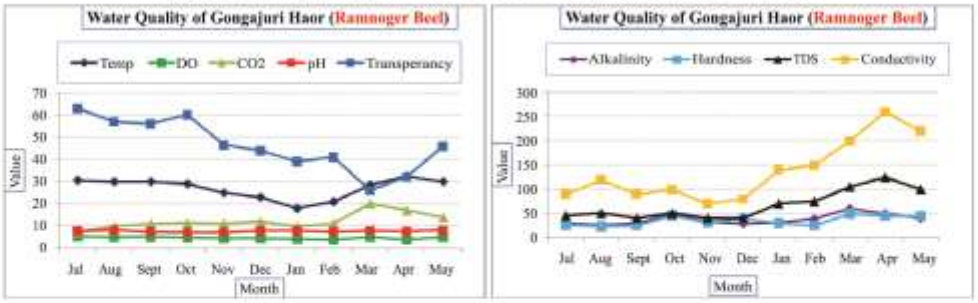


Figure 5. Water quality parameters of Ramnoger *beel,* Gongajuri *Haor,* Hobigonj.

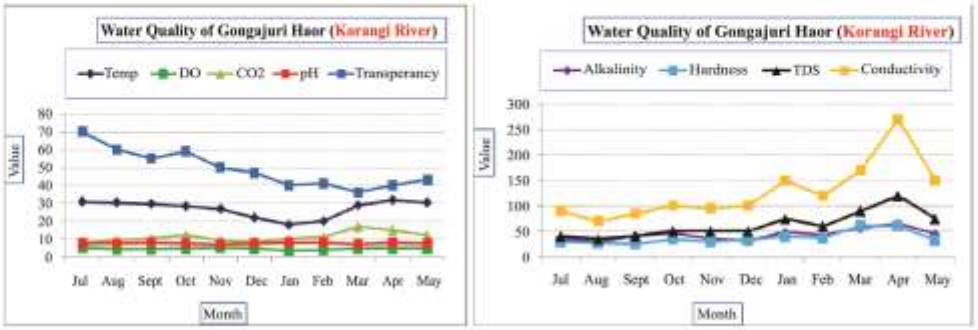


Figure 6. Water quality parameters of Korangi river, Gongajuri *haor,* Hobigonj.

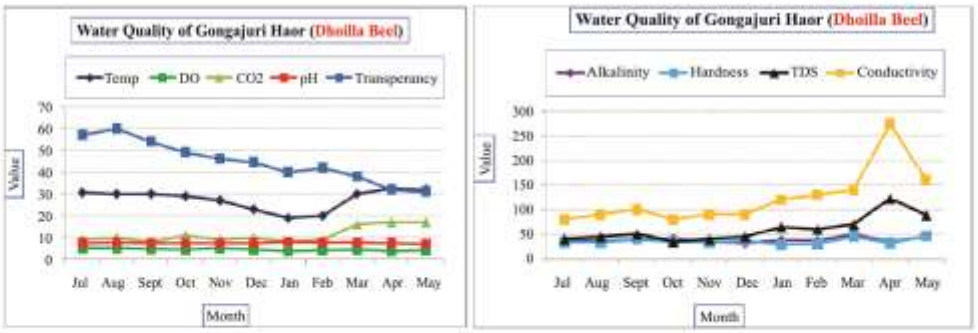


Figure 7. Water quality parameters of Dhoilla *beel,* Gongajuri *Haor,* Hobigonj.

Water quality standard

Almost all water quality parameters were within acceptable ranges for fish and other aquatic animals according to Bangladesh and other international standards (Table. 1).

Table 1. Water quality standards.

Sl. No.	Parameter	BD and other international standards	Source
1	Air Temperature (°C)	30.5	DoE, 2001
2	Water Temperature (°C)	30	EQS, 1997
3	Dissolved Oxygen (mg/l)	6.5	DoE, 2001
4	Carbondioxide (mg/l)	23	EPAUS, 1976
5	pH	8.5	EQS, 1997
6	Transperancy (cm)	45	Hossain, 2011
7	Alkalinity (mg/l)	100-200	Boyd and Tucker, 1998
8	Hardness (mg/l)	500	DoE, 1997

Plankton identification

A total of 28 genera of phytoplankton and 8 genera of zooplankton were identified of which Chlorophyceae in phytoplankton population and Crustacea in zooplankton population were dominant.

Table 2. Location wise list of different plankton of Padoarper *beel,* Chandpur.

Plankton Type	Plankton Groups	Genus
Phytoplankto n	Bacillariophyceae	*Nitzschia* sp., *Synedra* sp., *Navicula* sp., *Fragilaria* sp., *Asterionella* sp.
	Cyanophyceae	*Spirulina* sp., *Anabaena* sp., *Micricystis* sp., *Oscillatoria* sp., *Polycystis* sp.
	Chlorophyceae	*Closterium* sp., *Spirogyra* sp., *Tetraedron* sp., *Volvox* sp.
Zooplankton	Copepoda	*Cyclops* sp., *Nauplius* sp.
	Rotifera	*Keratella* sp., *Lecane* sp., *Brachionus* sp.
	Branchiopoda	*Bosmina* sp., *Daphnia* sp., *Moina* sp.

Table 3. Location wise list of different plankton of Pagner *Haor,* Sunamgonj.

Plankton Type	Plankto n Groups	Genus
Phytoplankton	Bacillariophyceae	*Asterionella* sp., *Coscinodiscus* sp., *Navicula* sp., *Nitzsehia* sp., *Synedra* sp.
	Cyanophyceae	*Anabaena* sp., *Aphanocapsa* sp., *Coelosphaerium* sp., *Lyngbia* sp., *Microcystis* sp., *Oscillatoria* sp., *Spirulina* sp.
	Euglenophyceae	*Euglena* sp. *Phacus* sp. *Trachelomonas* sp.
	Chlorophyceae	*Pediastrum* sp., *Pandorina* sp., *Closterium* sp., *Tetraedron* sp., *Scenedesmus* sp., *Volvox* sp., *Ankistrodesmus* sp.
Zooplankton	Copepoda	*Cyclops* sp., *Diaptomus* sp.
	Rotifera	*Lecane* sp., *Brachionus* sp.
	Branchiopoda	*Bosmina* sp., *Dapnia* sp., *Moina* sp.
	Protozoa	*Arcella* sp.

Table 4. Location wise list of different plankton of Gongajuri *Haor,* Hobigonj.

Plankton Type	Plankton Groups	Genus
Phytoplankton	Bacillariophyceae	*Asterionella* sp., *Cyclotella* sp., *Navicula* sp., *Synedra* sp.
	Cyanophyceae	*Spirulina* sp., *Microcystis* sp., *Nostoc* sp.
	Coscinodiscophyceae	*Coscinodiscus* sp.
	Euglenophyceae	*Euglena* sp., *Phacus* sp.
	Chlorophyceae	*Ankistrodesmus* sp., *Eudoria* sp., *Pandorina* sp., *Pediastrum* sp., *Scenedesmus* sp., *Tetradon* sp.
Zooplankton	Copepoda	*Cyclops* sp., *Nauplius* sp.
	Rotifera	*Brachionus* sp., *Lecane* sp.
	Branchiopoda	*Daphnia* sp., *Bosmina* sp., *Moina* sp.

Study 2. Field data collection for estimating population ecology of commercially significant haor and beel resident fish

Length weight

A total of 24 commercially important fish species individual's length-weight was recorded. Mean length-weight of the found fish species has been presented in Table 5.

Table 5. Mean values of 18 individuals fish length-weight (mean ± SD) recorded from different study area.

Fish	Study area					
	Padoarper *beel*		Gongajuri *haor*		Pagner *haor*	
	L	W	L	W	L	W
Punti	7.91±0.83	8.0±0.69	7.5±0.5	8.1±0.61	7.94±0.6	8.1±1.2
Mola	6.2±0.95	2.73±1.27	6.3±1	2.65±0.3	5.6±0.7	2.1±0.4
Baila	9.88±2.6	7.8±4.38	10.1±1.7	8±0.3	10.1±0.5	7.9±0.4
Boicha	4.28±0.67	1.49±0.53	4.34±0.5	1.51±0.3	4.2±0.4	3.9±0.3
Meni	8.43±2.15	9.25±5.38	17±1.5	31±2.1	14.2±1.2	21.5±1.1
Kholisha	8.7±1.02	10.3±1.8	9±0.7	11±0.5	9.1±0.2	10.2±0.23
Koi	14.09±1.7	43.3±2.7	14.1±2	45±1.4	14.2±1.5	43±0.6
Shing	16.3±4.1	26.7±2	16.4±3.1	27.3±0.4	16.5±1.6	27±0.7
Tengra	6.31±1.1	2.57±0.8	7.1±1.3	2.8±.4	8±0.4	7±0.4
Boal			70.7±5.0	1205.6±69.2	73±12	1139.5±0.4
Pabda			13.15±3.52	14.5±1.1	13±2.8	14.4±1.1
Taki	17.51±2.7	54.7±2	18±0.4	56±2	17.4±1.9	55±3.9
Shol	35.05±5.8	345.5±12.8	35.2±4.1	343±1.8	38.4±2.3	398±3.3
Mrigal			43.45±4.4	868±54.9	45.9±8.18	1050.46±26.53
Rui					42.05±15.5	1098.9±10.2
Kalbasu					30.0±8.0	450.7±10.1
Gonia			30.7±9.5	445.8±2.5	31.0±8.1	418.7±2.8
Ayre			36.5±10.6	468.1±3.6	35.64±10.7	431.2±35.8

Gear study

Various types of fishing gears/traps are used in those study area. Types of gear used depends upon fishermen's desire. Seine net, Cast net, Gill net and fish trap of different mesh size are operated daily.

Table 6. Location wise net list.

Sl. No.	Location		Net name and type
1	Padoarper *beel*	Padoarper *beel*	Fash Jal (Gill net), Current Jal (Gill net), China Doari Jal.
2	Pagner *haor*	Pata *beel*	Ber jal (Seine net), Fash jal (Gill net), Current jal (Gill net), Jhaki jal (Cast net), Charu/Chai/Unta/Bosni/Paron (Fish trap), Veshal Jal, Thela Jal.
		Nainda *beel*	
		Nolchorni *beel*	
3	Gongajuri *Haor*	Ramnogar *beel*	Ber jal (Seine net), Pata jal (Gill net) Fash jal (Gill net), Current jal (Gill net), Jhaki jal (Cast net), Charu/Chai/Unta/Bosni/Paron (Fish trap), Veshal Jal, Thela Jal.
		Bahubol Rubar Dump	
		Korangi river	

Study 3. Assessment of stock or biomass of commercially significant inland open water fish as well as water bodies.

Use of different fishing gear and traps can also serve as a rough indicator of the availability of different fish species. Some gears are species selective such as gill nets, traps, hook and lines and long line. We observed that gill net used widely during the sampling period in those study areas. CPUE of different types of fishing gear are presented in Table 7, 8 and 9.

Table 7. Monthly gear wise CPUE of Padoarper *beel* Chandpur. [kg/hour/100 m net].

Net Name	Hour	Jul	Aug	Sept	Oct
Current net	12	1.9	1.37	1.1	1.2
China Doari net	12	2.5	3	2	1.5

Table 8. Monthly gearwise CPUE of Pagner *Haor,* Sunamgong. [kg/hour/100 m net].

Net Name	Hour	Jul	Aug	Sept	Oct	Nov	Dec	Jan	Feb	Mar	April	May	June
Seine net	2	10	12	12	15	18	12	20	22	10	10	50	60
Cast net	3	2	2	1.5	2	1	2	4	6	2	2	4	6
Push net	4	1.5	1.5	2	1.2	1	1	1	1	0.5	0.5	2	4
Ricksha net	4	2	2	2	2	2	2	3	4			6	8
Fish trap	6	0.5	0.8	0.4	0.6	0.5	0.4	0.8	0.9			1.8	1.5
Veshal net	12	20	25	30	22	21						4.5	6
Gill/ Fash net	12	1	1.2	0.5	0.7	1	0.5					4	5

Table 9. Monthly gearwise CPUE of Gongajuri *Haor,* Hobigong. [kg/hour/100 m net].

Net Name	Hour	Jul	Aug	Sept	Oct	Nov	Dec	Jan	Feb	Mar	April	May
Seine net	2	9	11	10	12	14	10	18	22	0	0	30
Cast net	3	2	2	1.5	2	1	2	4	6	2	2	3
Push net	4	1	2	2	1.6	1.3	1	1	2	0.5	0.5	1
Ricksha net	4	1.6	1.5	1	1.5	2	2	3	4	0	0	4
Fish trap	6	0.5	0.8	0.4	0.6	0.5	0.4	0.5	0.8	0	0	1.8
Veshal net	12	22	23	25	20	19		0	0	0	0	3
Gill/ Fash net	12	0.5	0.8	0.6	0.5	1	0.5	0.5	0.4	0.3	1	2

Common observations

A major threat of haor and beel fisheries is complete harvesting of haor residential fish in summer season which affect their year after year recruitment. Use of current net and Moshari net (One kind of fine mesh Seine net) is an another threat for haor fisheries.

Impact of Lunar Periodicity, Saline Intrusion, Rainfall and Water Discharge on Hilsa Fisheries in a Changing Climate in Bangladesh

Researchers
Dr. Mohammad Ashraful Alam, SSO
Flura, SSO
Md. Moniruzzaman, SO
Sajida Akhter Suchana, SO
Md. Abu Kawser Didar, SO

Objectives

- To determine the effects of lunar periodicity and tidal fluctuations on hilsa breeding and production
- To assess the extent and intensity of saline intrusion on hilsa navigation route in Bangladesh and its impact on hilsa abundance and distribution
- To determine the impacts of rainfall and water discharge on hilsa production
- To estimate the impact of physico-chemical parameters on hilsa production
- To update hilsa management interventions and policy guidelines

Achievements in the past

To address the aforesaid objectives of the project, the data was collected from direct field observation, as well as some secondary data were accessed from some web sources that provide 24 hours real-time data on different meteorological factors. The data on tidal fluctuations (tidal co-efficient, tidal heights) was collected from "Tide Forecast.com" and Tides4Fishing" website. Lunar periodicity data were accessed from the "Phases of the Moon" app and classical "Time and Date.com". The data on hilsa demographics (length, weight, percentage of male, female, spent and oozing hilsa) were collected from direct field investigations from the selected sampling locations (Figure 1).
The results of the analysis are furnished below.

Average tidal co-efficient in different lunar phase

The tidal coefficients tell us the amplitude of the tide forecast (difference in height between the consecutive high tides and low tides in any given area). Average tidal co-efficient of Cox's Bazar, Ramgoti, Barishal, Borguna (Pathatghata), Potuakhali (Mohipur) and Chandpur were calculated. The tidal co-efficient showed considerable variations in different lunar phases. The highest tidal co-efficient was observed during the new moon (NM) in Cox's bazar, Barishal and Chandpur whereas the in Ramgoti the highest tidal co-efficient was found during the full moon (FM).

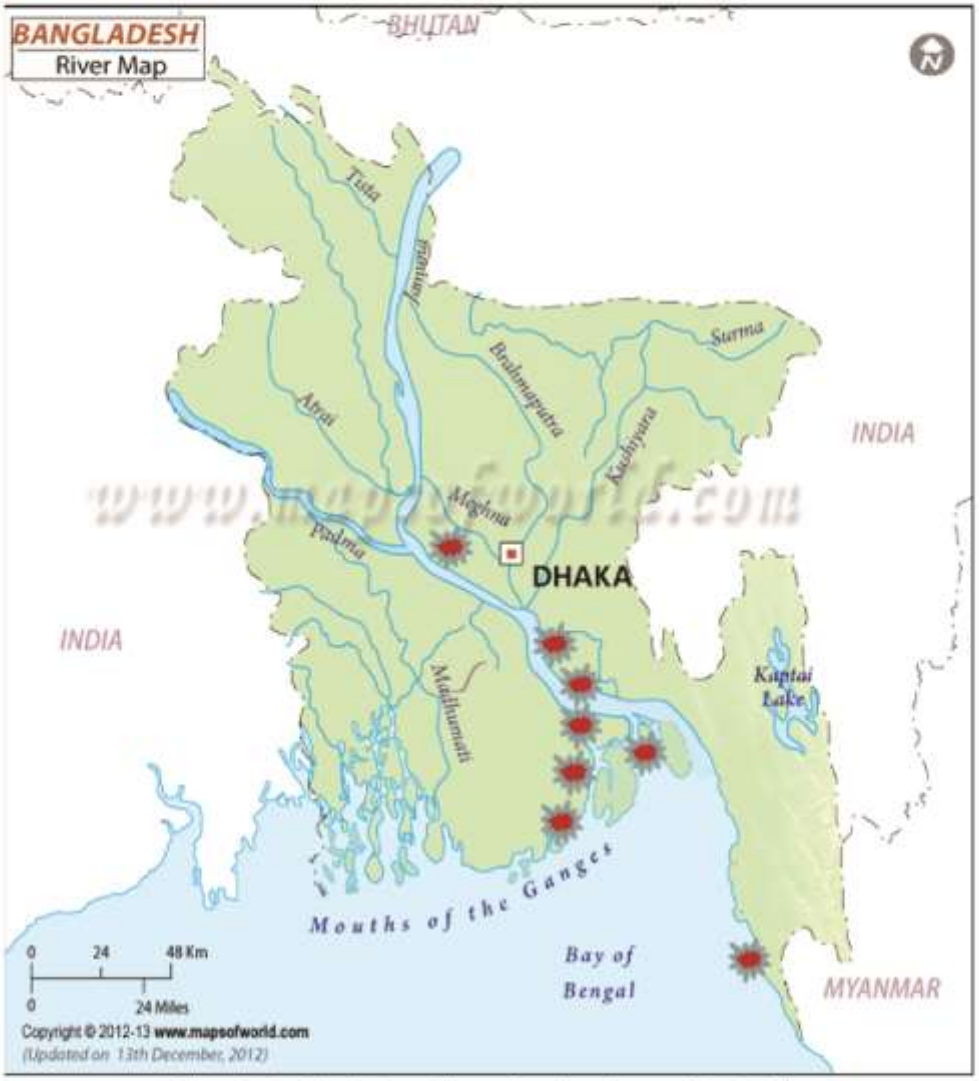


Figure 1. Sampling locations with asterisk mark on the map of Bangladesh.

On the contrary, in Potuakhali and Borguna the highest tidal co-efficient was found during the third quarter (TQ) of the moon (Figure 2). The collected data on the tidal co-efficient is still inadequate, therefore; it's difficult draw the firm line conclusion whether the tidal co-efficient is lunar phase dependent or place dependent. The relationship with average tidal co-efficient and lunar distance was also estimated based on the collected data and the results showed that the tidal co-efficient varies accordingly with the lunar distance.

Total nos. of hilsa in different lunar phases

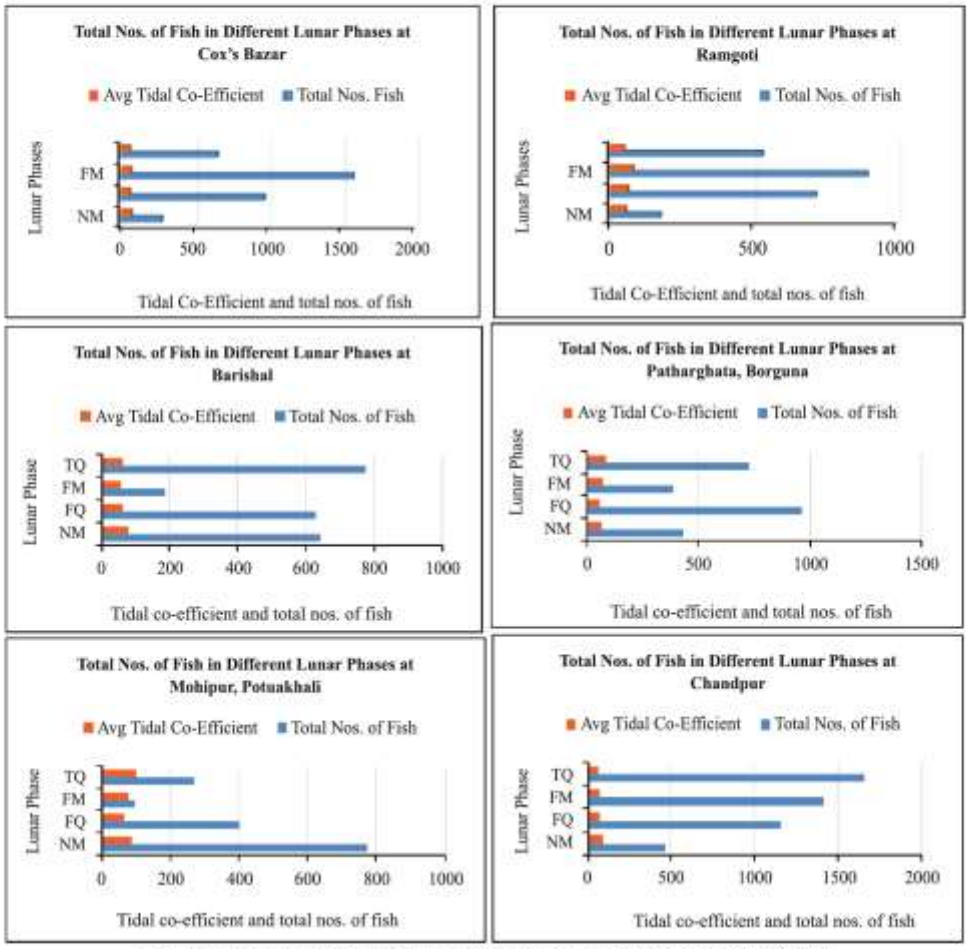


Figure 3. Total nos. of hilsa in different lunar quarters in relation to tidal co-efficient.

Total nos. of hilsa captured in different lunar phases also varied considerably. At Cox's Bazar and Ramgoti, the highest total nos. of hilsa were captured during the FM and tidal co-efficient were also higher than other phases of moon. At Barishal and Chandpur, the highest nos. of hilsa were captured during the TQ of the moon whereas at Patharghata, Borguna and Mohipur, Potuakhali, the highest nos. of hilsa were captured during the FQ and NM, respectively. The results of the present study conducted till to date indicate that marine hilsa capture (Cox's Bazar, Ramgoti and Potuakhali) is influenced by lunar distance *viz*. full and new moon (Figure 3). Albeit, tidal co-efficient is directly influenced by the lunar distance, still is difficult opine that it has specific and direct influence on the total nos. of hilsa captured, more data collection is required to reach in that conclusion.

Hilsa breeding performance in different lunar phases

The percentage of male, female, spent and oozing hilsa were calculated in the present study. The result demonstrated that highest percentage of male, female, spent and oozing hilsa were found during the NM followed by the other quarters.

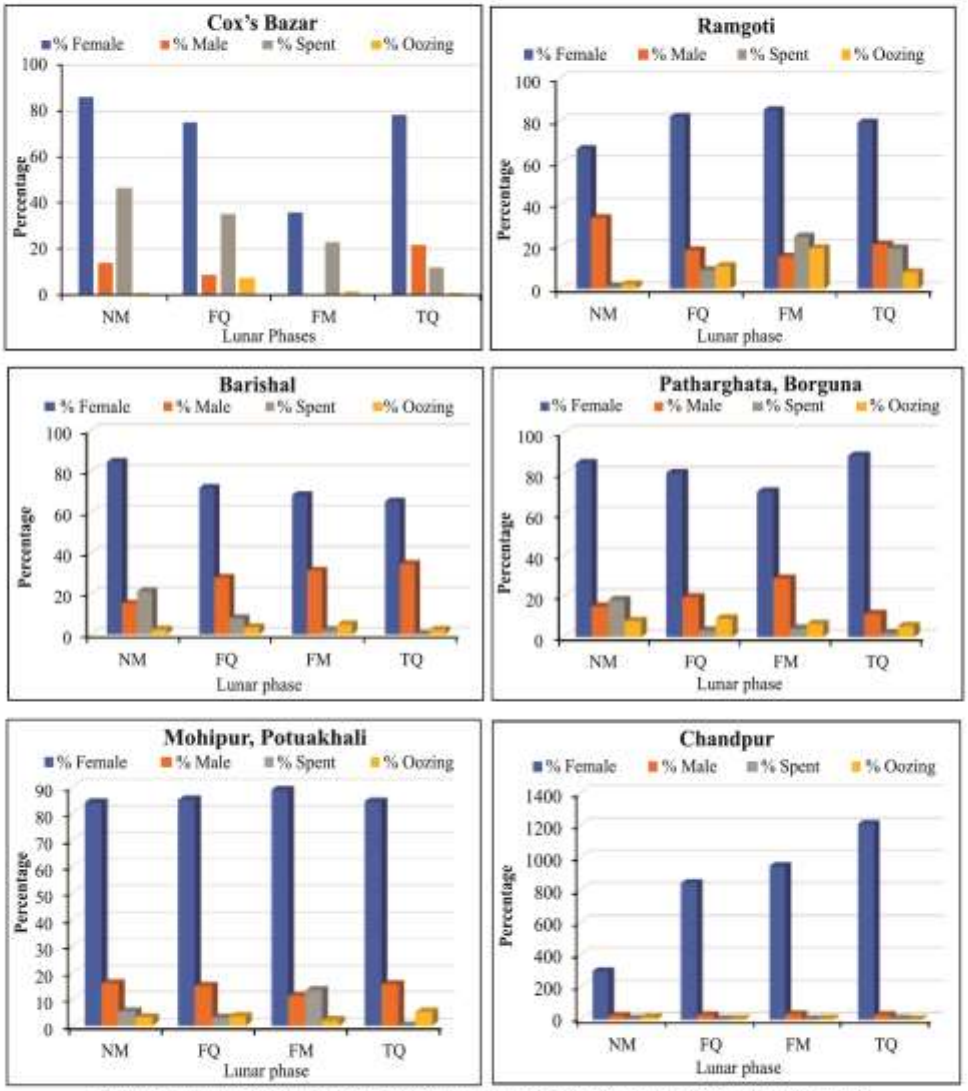


Figure 4. The percentage of male, female, spent and oozing hilsa during different lunar phases.

The gonadosomatic index (GSI) of hilsa collected from all sampling locations in different lunar phases were also calculated and the mean value of GSI exhibited considerable fluctuations. The highest GSI value was found during the NM as well, indicating that hilsa prefers to breed during the NM rather than the other quarters (Figure 4 and 5).

Annual Rainfall and Hilsa Production

Production and catchability of fish in many aquatic ecosystems varies considerably because of seasonal, annual, inter-annual and decadal variability in rainfall. The historical rainfall data (1991-2020) was collected from the meteorological department and mean annual rainfall was calculated and correlated with year wish total hilsa production in Bangladesh (Figure 6). A regression model was constructed using mean annual rainfall and total hilsa production of the country (2010-2020) and the result showed weak linear association between these two variables (Figure 7). More data is required to reach in a conclusion in this regard.

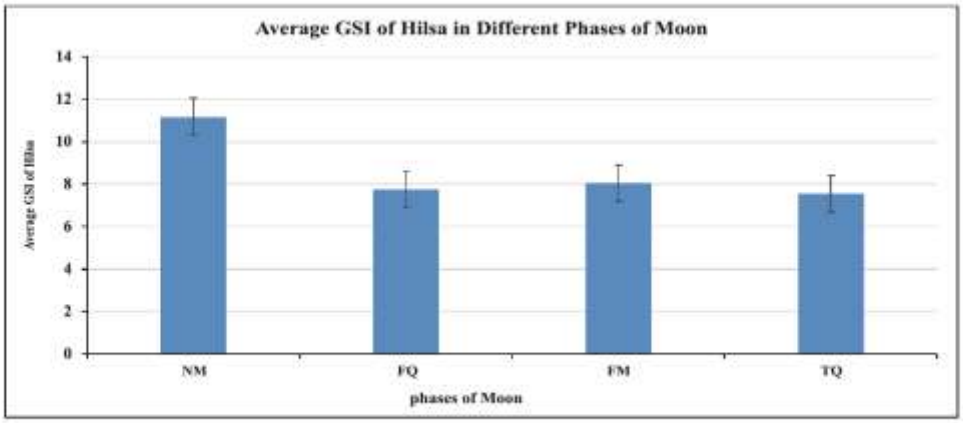


Figure 5. Gonadosomatic index of hilsa in different lunar phases.

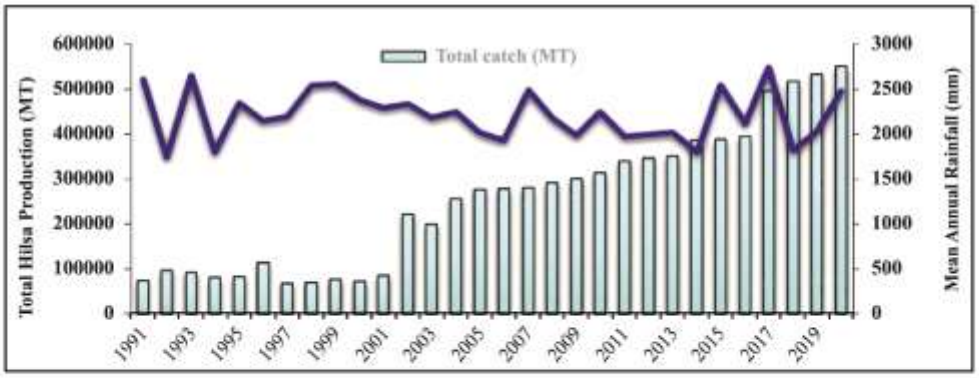


Figure 6. Mean annual rainfall and total hilsa production in Bangladesh (1991-2020).

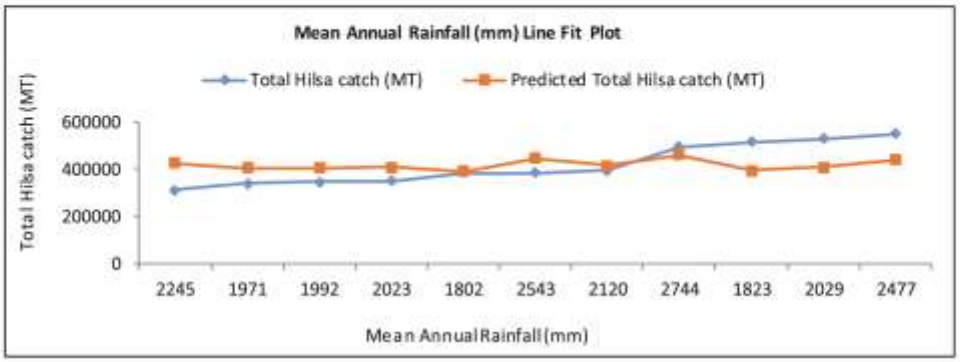


Figure 7. Linear regression model between mean annual rainfall and total hilsa production in Bangladesh (2010-2020).

Physico-chemical parameters of water in the sampling locations

Analyses of various physico-chemical factors of the water quality from different sampling points are presented in Table 1. The ranges of all studied water quality parameters were found within the acceptable limits for the growth of fish.

Parameters	Sampling points					
	Cox's Bazar (Mean±SD)	Ramgoti (Mean±SD)	Barisal (Mean±SD)	Patharghata (Mean±SD)	Mohipur (Mean±SD)	Chandpur (Mean±SD)
Air temp (°C)	30.2±2.94	29.49 ±1.22	28.25±1.71	29.73±5	29.6±4.3	28.76±2.4
Water temp (°C)	28±5.35	27.32±1.1	26.75±2.53	26.4±4.5	27.45±4.43	26.2±2.1
Transparency (cm)	41.67±13.06	19.66±11.26	33.78±9.63	31.5±5.2	60.71±21.7	29.32±9.2
Dissolve O_2 (mg/l)	6.40±0.52	7.35±0.45	6.87±0.49	6.14±0.8	7.2±1.3	6.42±0.6
Free CO_2 (mg/l)	9.9±3.0	10.73±3.30	10.62±2.38	10.8±3.2	12.8±2.8	10.0±2.0
NH_3	0	0	0	0	0.01±0.04	0
pH	7.87±0.2	7.89±0.08	8±0.26	7.9±0.3	8.18±0.3	7.8±0.3
Total alkalinity (mg/l)	97.75±6.6	62±12.83	75±12.96	90.8±21.4	93.4±28.3	68.4±12.9
Total hardness (mg/l)		78±15.76	86±23.17	81.75±33.1	80.72±23.2	77.7±12.9
Salinity	31.2±3.7	0.2±0.12	0.045±0.02	0.58±0.9	5.05±2.06	0.043±0.06
Conductivity		420.3±53.57	250.7±50.3	667.3±32.2	8095±202.3	164±10.0
Total Dissolved Solid (TDS)		249±140		373.3±270.9	6413±1636	190±52

Estimation of Nutrient Flux and Primary Productivity in the Major Nursery Grounds of Hilsa

Researchers

Flura, SSO
Dr. Mohammad Ashraful Alam, SSO
Md. Abul Bashar, SSO
Md. Moniruzzaman, SO
Sajida Akter Suchana, SO
Md. Mezbabul Alam, SO

Objectives

- To assess the primary productivity of nursery grounds of hilsa
- To study the factors affecting primary productivity of nursery grounds of hilsa
- To assess the carrying capacity of nursery grounds of hilsa

Achievements

In order to address the aforesaid objectives of the project, monthly data from six sampling locations (Shatnol, Chandpur-Alexander, Laxmipur 100 km considered as station 1, Lower Meghna and Tarabunia, Shariotpur 20 km, Lower Padma considered as station 2, Hizla, Mehindigonj, Barishal (82 km) considered as station 3, Bheduria, Bhola, Char Rustom, Potuakhali (100 km, Tetulia river) considered as station 4, Char Ilisha-Char Pial, Bhola (90 km, Shahbazpur Channel considered as station 5, Kalapara Upazilla, Patuakhali (40 km) considered as station 6 were collected and analyzed.

Primary productivity of nursery grounds of hilsa in the study areas

The primary productivity of a water body is the manifestation of its biological production. It is an ultimate outcome of photosynthesis that forms the basis of ecosystem functioning since it makes the chemical energy and organic matter available to the entire biological community. To determine the primary production, representative water samples were collected at the onset of sunrise from the mid-euphotic level with the help of a water sampler. Thereafter, both light and dark bottles were hanged in duplicate in the water at the same depth and incubated for half the period of the day length. The data were analyzed using standard protocols. The result showed that the average NPP was found higher at (S1) (0.68 ± 0.01), (S2) (0.43 ± 0.01) and (S5) (0.35 ± 0.02) (Figure 1) respectively compared to the other sampling sites. The average GPP was found higher at (S1) (0.98 ± 0.28), (S2) (0.65 ± 0.27) and (S5) (0.6 ± 0.26) (Figure 1) respectively compared to the other sampling sites. The average respiration was found higher at (S1) (0.30 ± 0.11), (S5) (0.29 ± 0.11) respectively compared to the other sampling sites (Figure 1). The average NPP and GPP was found higher at Station 1 (0.68 ± 0.01; 0.98 ± 0.28) and the average respiration was found higher at station 1(0.30 ± 0.11) compared to other sampling stations (Figure 1). This result also establishes coherence between the higher abundance of jatka (CPUE) and average NPP and GPP at Confluence (Figure 2) compared to other sampling sites.

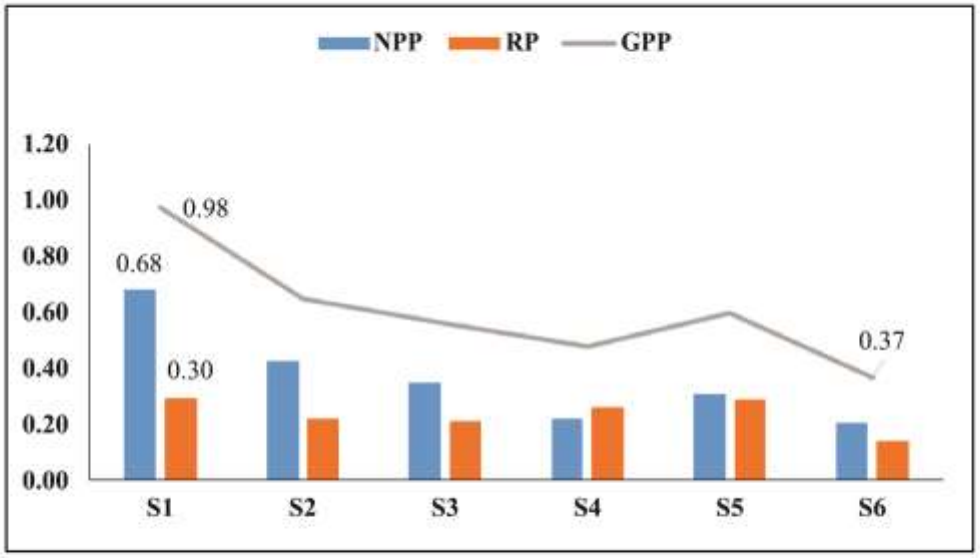


Figure 1. Net primary, Respiration and Gross primary productivity (gC/m^3/day) of sampling stations.

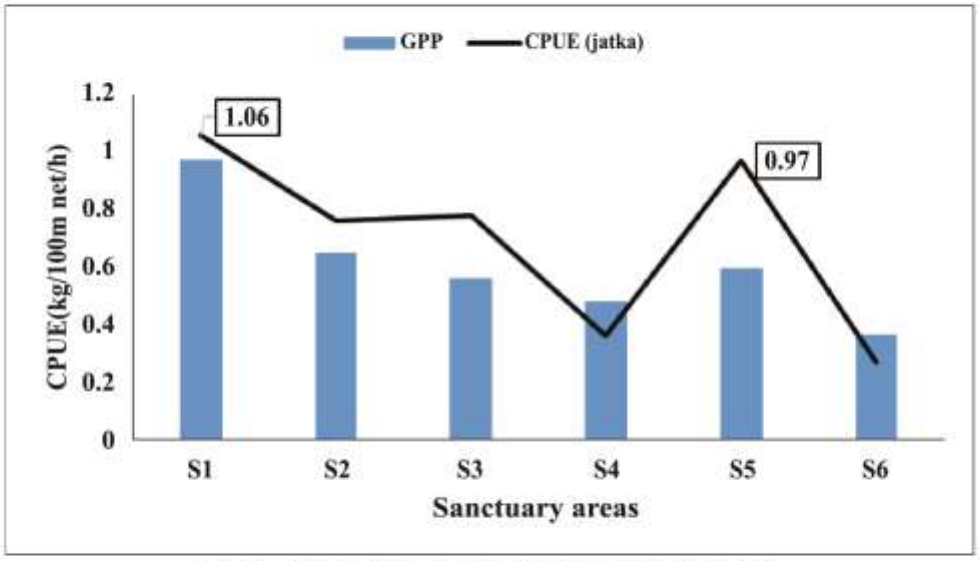


Figure 2. CPUE of jatka (kg/100 m net/h) at selected sampling stations.

Fish larvae were sorted out from other zooplanktonic organisms using a dissecting microscope. Larvae of T. ilisha were differentiate from other zooplanktonic organisms collected based on common distinguish characterisrics of clupiforms. For hilsa larvae collection, Bongonet was mainly used. Bongonet was set into the selected sampling locations for 30 minutes and all larvae were collected in plastic bottle and immediately preserved in ethanol (10%). Thereafter, the larvae of different fish species alongside with hilsa were identified with microscopic observation. The percentage of hilsa larvae was found higher at (S5) 71 % and (S1) 70% compared to other station whereas the percentage of other larvae was (S6) 82 % and (S3) 65% respectively (Figure 3).

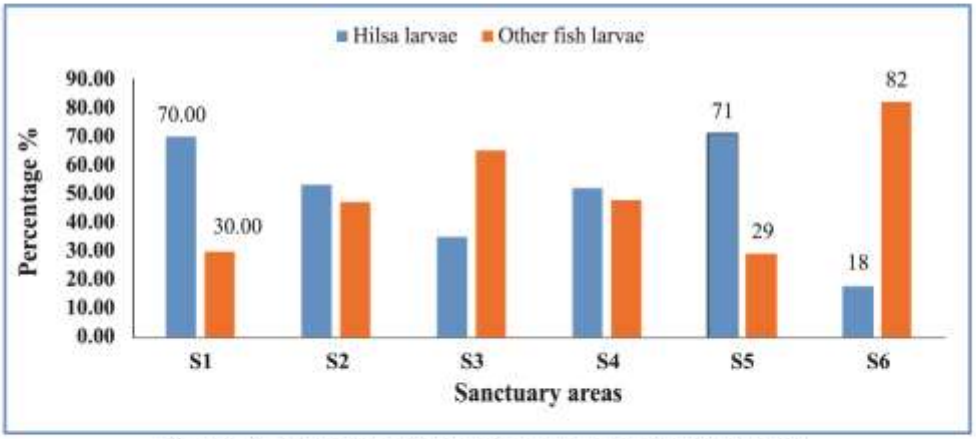
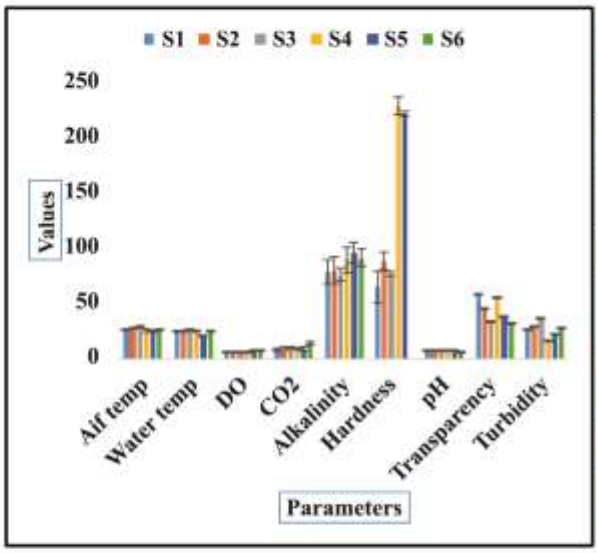


Figure 3. Abundance (%) of hilsa and other larvae at selectedsampling Stations.

Factors affecting the primary productivity

Physico-chemical parameters of water in the study areas

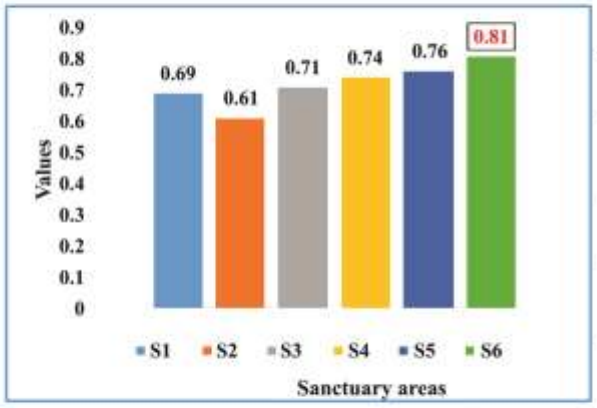
Water quality parameters of six stations exhibited considerable fluctuations. The average highest air temperature was found 27±0.70 ℃, 2757±1.9 ℃, 29 ± 0.5 ℃, at (S1, S2 and S3) respectively. The average water temperature was found 26 ±2.82 ℃, 26.23±1.72℃, 25.75 ± 0.25 ℃ at (S2, S3 and S6) respectively average highest dissolved oxygen (mg/l) was found 7.2±0.08, 7.55±0.47, and 6.57±0.42 respectively at (S2, S5 and S6). Average CO_2 (mg/l) were in acceptable limits respectively. The maximum alkalinity was found 89±1.14 mg/l at S6 and hardness 221± 7.63 mg/l at S5 (Figure 4). The average pH was found just slightly above the neutral value in the studied sampling sites. In stations 1, 2 and 3 the ranges of all studied water quality parameters were found within the acceptable limits for the growth of fish (Figure 4). In stations 4, 5 and 6 studied water quality parameters were slightly different but in acceptable limit (Figure 4).



Parameters	Standard
Aif temp	20-30 (EQS,1997)
Water temp	20-30 (EQS,1997)
DO	4-6 (EQS,1997)
CO_2	6 ppm (EQS,1997)
Alkalinity	>100 (Rahman, 1992)
Hardness	200-500 (DOE, 2003)
pH	6.5-8.5(EQS,1997)
Transparency	40 or less (Rahman, 1992)

Figure 4. Water quality parameter of sampling Stations.

Water quality index (WQI) is a dimensionless number that combines multiple water quality parameters into a single number by normalizing values to subjective score (Miller et al., 1986). Conventionally it has been used for evaluating the quality of water for water resources such as rivers, streams and lakes.



WQI Value	Category
<0.5	Excellent water
0.5-0.75	Good water
0.75-1	Moderately polluted water
>1	Highly polluted water

Figure 5. WQI at different Stations.

Parameter incorporated in WQI varies depending upon the designated water uses of the water body and local preferences. In the present study highest WQI values were found in S6 indicated moderately polluted water.

Cluster Analysis (CA) was carried out, using Bray Curtis Similarity, to show the similarity among the parameters. From the output of the cluster analysis, four clusters were found during different stations (Figure 6).

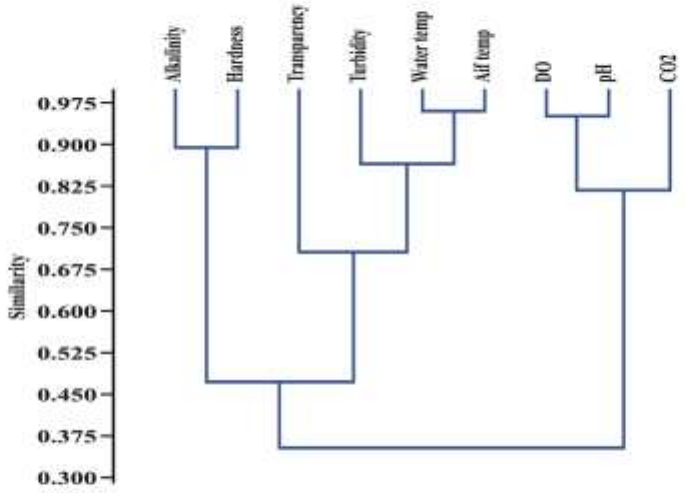


Figure 6. Dendogram showing the percentage of similarity among water quality parameters.

Nitrogen (N) and phosphorus (P) are primary nutrients and vital for life processes such as protein synthesis, cellular growth and reproduction. However, in excessive quantities, the two elements are also a major source of stream and river impairment. Large inputs of these limiting nutrients can cause deleterious algal growth with a myriad of negative ecosystem responses including eutrophication.

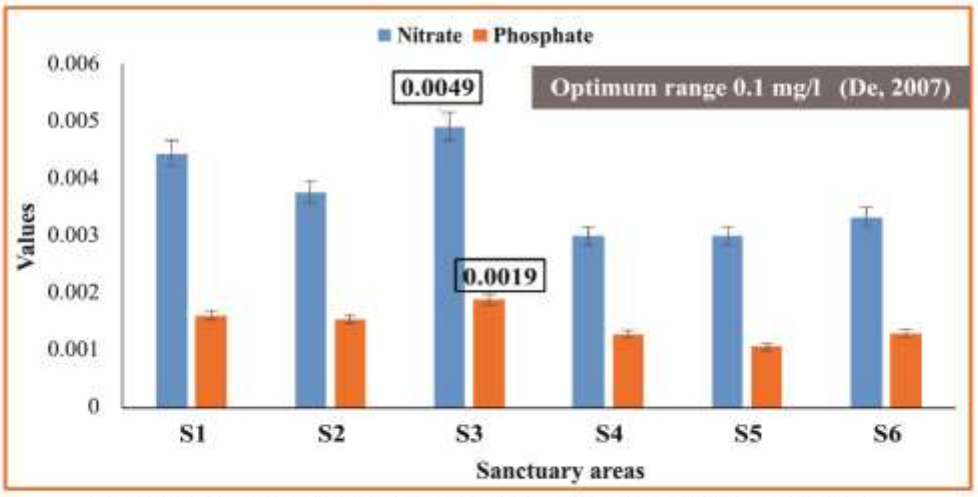


Figure 5. Concentrations of Nitrate (mg/l) and Phosphate (mg/l) in the selected sampling spots.

The highest average concentration of nitrate was found at S3 (0.0049 ± 0.002 mg/l) and S1 (0.0042±0.002 mg/l) whereas Phosphate were found at S1 (0.00161±0.005 mg/l, and S2 0.00151±0.007 mg/l respectively and the lowest at S3 (0.0013 ± 0.005 and S4 (0.00129 ±0.003 mg/l), respectively (Figure 5).

According to Bhatnagar et al. (2004), concentration of nitrate 0.02-1.0 ppm is lethal to many fish species, > 1.0 ppm is lethal for many warm water fish and < 0.02 ppm is acceptable (OATA, 2008). According to Stone and Thomforde (2004), the phosphate level of 0.06 mg/l -1 is desirable for fish culture. Bhatnagar et al. (2004), suggested 0.05-0.07 ppm is optimum and productive; 1.0 ppm is good for plankton and shrimp production. Thus, the nitrate and phosphate concentration in the present study was within the acceptable limit. The higher amount of contamination from fertilizers, municipal wastewaters, feedlots, septic systems in water increase the concentration of Nitrate, it refers that the higher (NO2 and NO3) the deviation the lower the quality of water for fish and other aquatic life and for common uses. The amount of nitrate could also be influenced by the growth of plankton.

The concentration of Chlorophyll a can act as an indicator of phytoplankton abundance in an aquatic ecosystem. One of the major objectives in analyzing photosynthetic pigments (Chlorophyll-a) in limnology is the estimation of phytoplankton biomass and its photosynthetic capacity. It is natural for levels of chlorophyll a to fluctuate over different seasons. The highest average concentration of Chlorophyll were found at S4 (7.14 ± 0.43 mg/l) and lowest at S6 (5.12 ± 0.09mg/l), respectively (Figure 6). Chlorophyll-a value is an indicator of productivity in the water body, which shows an inverse relationship with water transparency.

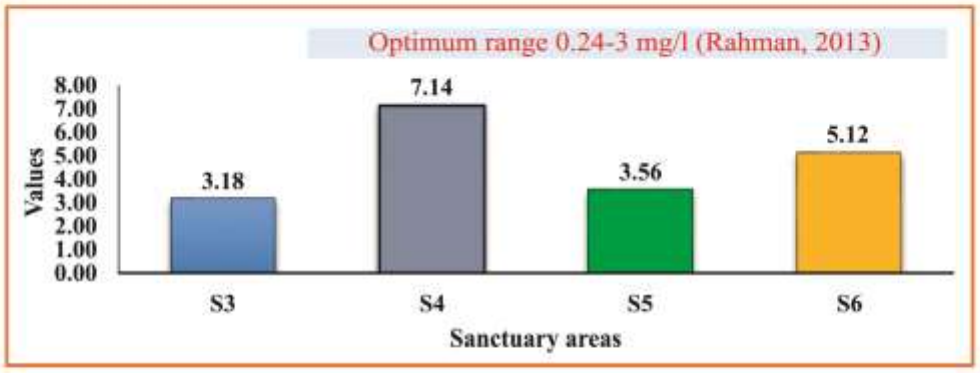


Figure 6. Concentrations of Chlorophyll-a (mg/l) in the selected sampling spots.

Plankton abundance in the study areas

In Station 1, 20 taxa were identified, of them 17 were phytoplankton and 3 were zooplankton. Among the pytoplankton, the dominant group was Zygnematophyceae in all the sites in Station 1. But in case of zooplankton the dominant group was Nymphalidae (Figure 7).

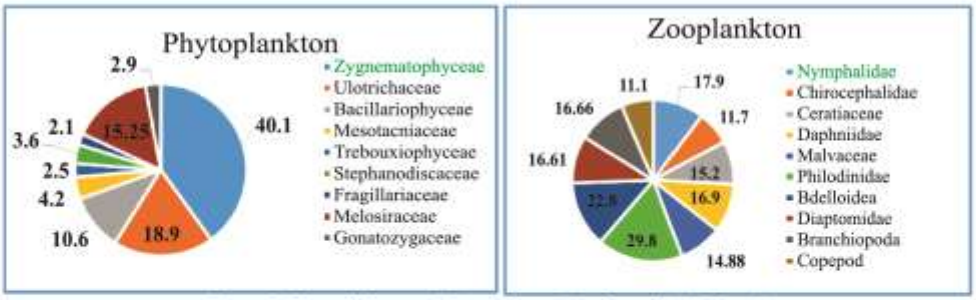


Figure 7. Phytoplankton and Zooplankton (%) of sampling Station 1.

In Station 2, 14 taxa were identified, of them 9 were phytoplankton and 5 were zooplankton. Among the phytoplankton, the dominant group was Zygnematophyceae but in case of zooplankton the dominant group was Hexanauplia (Figure 8).

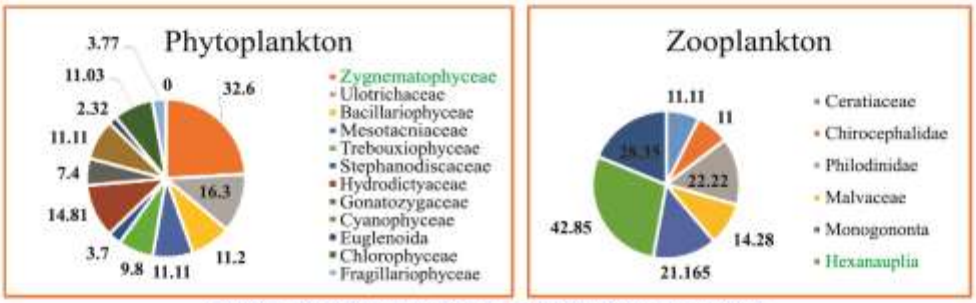


Figure 8. Phytoplankton and Zooplankton (%) of sampling Station 2.

In Station 3, 12 taxa were identified, of them 7 were phytoplankton and 5 were zooplankton. Among the phytoplankton, the dominant group was Cholorophyceae but in case of zooplankton the dominant group was Branchiopoda (Figure 9).

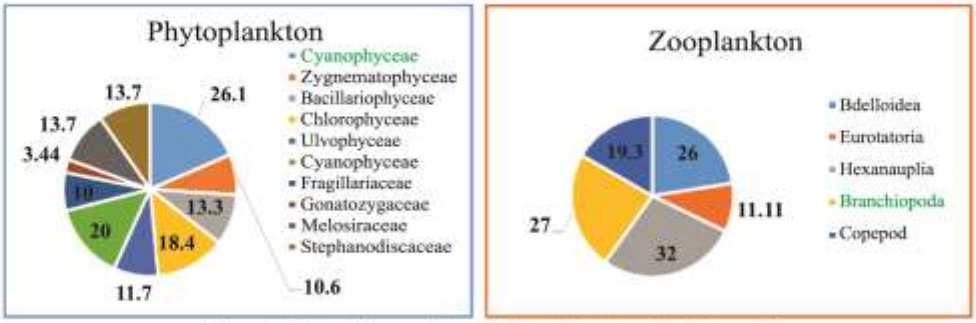


Figure 9. Phytoplankton and Zooplankton (%) of sampling Station 3.

In Station 4, 9 taxa were identified, of them 6 were phytoplankton and 3 were zooplankton. Among the phytoplankton the dominant group was Chlorophyceae but in case of zooplankton the dominant group was *Branchiopoda* (Figure 10).

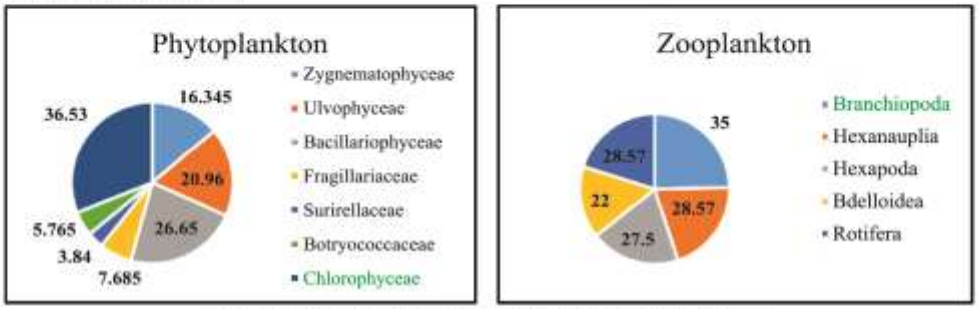


Figure 11. Phytoplankton and Zooplankton (%) of sampling Station 4.

In Station 5, 15 taxa were identified among which 9 were phytoplankton and 6 were zooplankton. Among the phytoplankton, the dominant groups were Zygnematophyceae, Bacillariophyceae and Chlorophyceae but in case of zooplankton the dominant groups were Branchiopoda and Monogota (Figure 12).

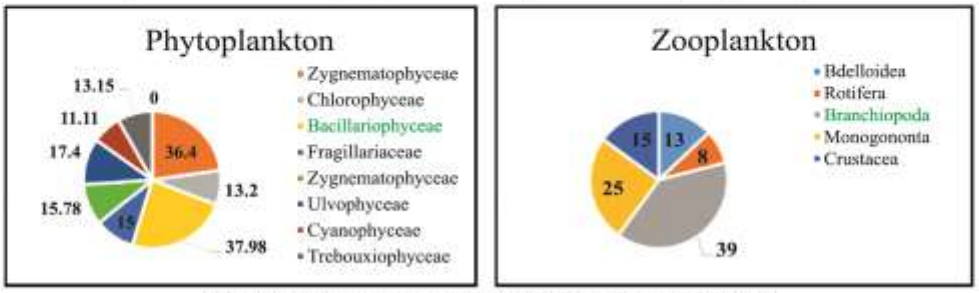


Figure 12. Phytoplankton and Zooplankton (%) of sampling Station 5.

In Station 6, 17 taxa were identified of them 10 were phytoplankton and 7 were zooplankton. Among the phytoplankton, the dominant groups were Zygnematophyceae, Bacillariophyceae and Chlorophyceae but in case of zooplankton the dominant groups were Monogononta and Branchiopoda (Figure 13).In Station 6, 17 taxa were identified of them 10 were phytoplankton and 7 were zooplankton. Among the phytoplankton, the dominant groups were Zygnematophyceae, Bacillariophyceae and Chlorophyceae but in case of zooplankton the dominant groups were Monogononta and *Branchiopoda* (Figure 13).

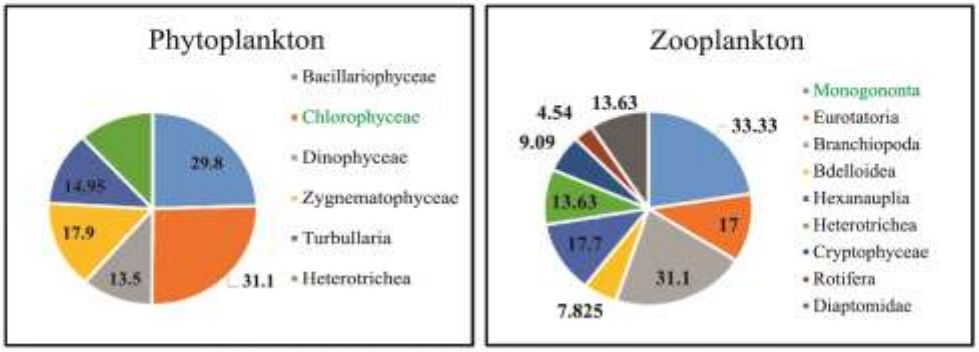


Figure 14. Phytoplankton and Zooplankton (%) of sampling Station 6.

Table 1. Abundance of plankton at sampling Stations.

Sanctuary	Total Plankton	Phytoplankton	Zo oplankton
Sanctuary 1	42×10^2	37×10^2	5×10^2
Sanctuary 2	39×10^2	33×10^2	6×10^2
Sanctuary 3	41×10^2	32×10^2	9×10^2
Sanctuary 4	37×10^2	33×10^2	4×10^2
Sanctuary 5	46×10^2	37×10^2	9×10^2
Sanctuary 6	31×10^2	24×10^2	7×10^2

Table 2. Class and Genus of Phytoplankton at sampling Stations.

Class	Genus
Chlorophyceae	*Pediastrum, Volvox, Scenedesmus, Acanthocystis*
Ulvophyceae	*Ulothrix*
Zygnematophyceae	*Spirogyra, Nitzschia, Netrium, Staurastrum, Gonatozygon*
Bacillariophyceae	*Navicula, Gomphonema, Asterionella, Diatoma, Frustulia, Stephanodiscus, Cyclotella*
Fragillariophyceae	*Tabellaria, Synedra*
Cyanophyceae	*Spirulina, Rivularia, Oscillatoria*
Trebouxiophyceae	*Protococcus, Botryococcus*
Dinophyceae	*Ceratium*
Euglenoida	*Euglena*

Class	Genus
Branchiopoda	*Daphnia, Ceriodaphnia, Sida, Bosmina, Diaphanosoma, Leptodora, Eubranchipus*
Hexanauplia	*Cyclops*
Heterotrichea	*Spirostomum*
Diaptomidae	*Diaptomus*
Monogononta	*Filinia, Brachionus*
Bdelloida	*Nauplius, Rotaria*

In the study period, this result also establishes coherence between the higher abundance of jatka and plankton density at S1 and S5 (Figure 15) compared to other sampling Stations.

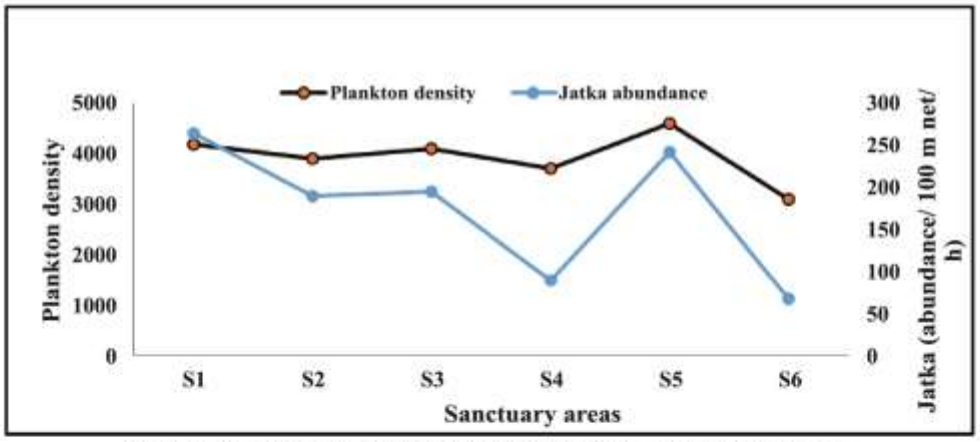


Figure 15. Relation between jatka abundance and plankton density at sampling Stations.

Diversity of Adaptive Gear and Their Impact on Kaptai Lake Fisheries

Researchers
B.M. Shahinur Rahman, SSO
Dr. Azhar Ali, PSO
Rabina Akhter Lima, SO
Md. Khaled Rahman, SO
Md. Lipon Mia, SO

Objectives

- To identify the gears used in the Kaptai Lake
- To determine the CPUE and catch composition
- Cataloging fish and gears of Kaptai lake
- To analyze cumulative length frequency

Achievement

Monthly sampling was done from different sampling sites of Kaptai Lake where 10 types of fishing gears, 06 types fishing trap and many Brush shelters were found. Among these one brush shelter catch data were collected during the study period. Length, width, personnel, fishing efficiency and others information of the fishing gears and traps as well as Catch Per Unit Effort (CPUE), length and weight data of fish were taken. During the research period cataloging or specification of Fish and Gears were done in Field and laboratory.

Identified Fishing Gears

- Kechki net/seine net
- Dharmo jal (Square Shaped Ber net)
- Current net/ gill net
- Cotton net (larger mesh size)/Gill net
- Cotton net (Small mesh size)/Gill net
- Nylon Cotton net (larger mesh size)/Gill net
- Tengra/ kajoli net or Gill net
- Dak jal (Ber net)/Seine net
- Mosquito Pelling net
- Khap jal (Cast net)

Identified Fishing Trap

- Single Borshi
- Borshi or hook line
- Long line Borshi
- Chingri Trap
- Chingri Chai
- China Duary

Brush Shelters

Brush shelters are locally known as Jak in the channel/river of the Kaptai Lake. These are prepared by different types of tree branches under water and covered by water hyacinth and different types of lure or bait are used for fish attraction. Many brush shelters or Jak found in Kaptai Lake and 01 brush shelter's species compositions were studied during the research period of 2021-2022.

Fish and Gear Cataloging or Specification

A total of 81 fish species, 10 fishing gears; 06 fishing traps and many brush shelters were found to in Kaptai Lake. Fish and gear's names are given below.

Table 1. Native species found in Kaptai Lake.

SI	Species name	Scientific name	SI	Species name	Scientific name
1	Ayre	*Sperata aor*	23	Guchi baim	*Mastacembelus pancalus*
2	Guii Ayre	*Sperata seenghala*	24	Jat Puti	*Puntius shophore*
3	Bata	*Labeo bata*	25	Koi	*Anabus testudineus*
4	Bele	*Glossogobius giuris*	26	Shol	*Channa striata*
5	Boirali	*Gonialosa manmina*	27	Batashi/tengra	*Batasio batasio*
6	Mola	*Amblypharyngodon mola*	28	Kalibaush	*Labeo calbasu*
7	Darkina	*Rasbora daniconius*	29	Kajuli	*Ailia coila*
8	Dhela	*Osteobrama cotio*	30	Round Chanda	*Parambasis ranga*
9	Ek thuta	*Hyporhamphus limbatus*	31	Kakila	*Xenentodon cancila*
10	Foli	*Notopterus notopterus*	32	Lal chanda	*Pseudambassis lala*
11	Ful chela	*Salmostoma phulo*	33	Long Chanda	*Chanda nama*
12	Vacha	*Eutropiichthys vacha*	34	Poa/Poma	*Johnius coitor*
13	Batashi	*Pseudeutropius atherinoides*	35	Rui	*Labeo rohita*
14	Chapila	*Gudusia chapra*	36	Shal baim	*Mastacembelus armatus*
15	Kechki	*Corica soborna*	37	Shing	*Heteropneustes fossilis*
16	Boal	*Wallago attu*	38	Vita tengra	*Mystus vittatus*
17	Chitol	*Notopterus chitala*	39	Tit puti	*Puntius ticto*
18	Foli (white)	*Notopterus spp.*	40	Shota shinghi	*Amblyceps mangois*
19	Kani pabda	*Ompok bimaculatus*	41	Cheng	*Channa orientalis*
20	Catla	*Catla catla*	42	Magur	*Clarias batrachus*
21	Napit koi	*Badis badis*	43	Kanchan puti	*Puntius conchonius*
22	Taki	*Channa punctata*	44	Gulsha tengra	*Mystus bleekeri*
45	Gulsha	*Mystus cavasius*	59	Khorsula	*Rhinomugil corsula*
46	Mrigal	*Cirrhinus cirrhosus*	60	Teri puti	*Puntius terio*
47	Kholisha	*Colisa fasciatus*	61	Chola puti	*Puntius chola*
48	Kata chanda	*Pseudambassis baculis*	62	Mola Punti	*Puntius guganio*
49	Kala bata	*Crossocheilus latius*	63	Cuchia	*Monopterous cuchia*
50	Gora chela	*Securicula gora*	64	Chital	*Chitala spp.*
51	Balichata	*Acanthocobitis zonalternans*	65	Red kholisha	*Colisa lalia*
52	Darkina	*Esomus danricus*	66	Gozar	*Channa marulius*
53	Bamosh	*Anguilia bangalensis*	67	Gutum	*Lepidocephalus thermalis*
54	Buno Khalisha	*Trichopsis vitatta*	68	Chingri	*Macrobrachium lamarrei*
55	Sada gonia	*Labeo gonius*	69	Kanpona/Blue Panchax	*Aplochilus panchax*
56	Couwa	*Gagata cenia*	70	Tin kata pushil/kosi hara	*Erethistes pussilus*
57	Gili puti	*Puntius gelius*	71	Ghaura	*Clupisoma garua*
58	White Chingri	*Macrobrachium spp.*	72	Black Chingri	*Macrobrachium spp.*

Table 2. Exotic species found in Kaptai Lake.

SI	Species name	Scientific name	SI	Species name	Scientific name
1	Grass crap	*Ctenopharyngodon idella*	5	Thai pangus	*Pangasianodon hypophthalmus*
2	Silver carp	*Hypophthalmichthys molitrix*	6	Sucker mouth catfish	*Hypostomus plecostomus*
3	Bighead carp	*Hypophthalmichthys nobilis*	7	Tilapia nilotica	*Oreochromis niloticus*
4	Common carp	*Cyprinus carpio*	8	Tilapia Mossambica	*Oreochromis mossambicus*
09	Thai Sarputi	*Barbonemus gonionotus*			

Gear Cataloging

Gear cataloging data were collected from the site during the research work. Among the collected data, the specification of 02 gears are given below; rest of gear cataloging data will be submitted in final report after compilation.

Kechki Net

Net Type : Kechki Net (Seine Net)
Mesh size : 02 mm
Length : 219.51-676.83 m
Wide : 12.8-52.5 m
CPUE : 1.7-160 kg
Haul time : 02-4.0 hour
Operation water depth : 9.05-22.61 m and

Species found: 50 species

Table 3. Different fish species found in Ketchki net.

Sl.	Species name	Sl.	Species name
1	Kechki (*Corica soborna*)	26	Round chanda (*Parambassis ranga*)
2	Chapila (*Gudusia chapra*)	27	Boirali (*Gonialosa manmina*)
3	Guchi Baim (*Macrognathus pancalus*)	28	Kakila (*Xenentodon cancilla*)
4	White chingri (*Macrobrachium lamarrei*)	29	Bele (*Glossogobius giuris*)
5	Ek thuta (*hyporhamphus limbatus*)	30	Darkina (*Esomus danricus*)
6	Long Chanda (*Nama chanda*)	31	Vacha (*Eutropiichthys vacha*)
7	Chela (*Chela cachius*)	32	Lal Kholisa (*Colisa fasciatus*)
8	Tit puti (*Puntius ticto*)	33	Kajoli (*Ailia coila*)
9	Mola (*Amblypharyngodon mola*)	34	Dhela (*Osteobrama cotio*)
10	Lal Chanda (*Parambassis lala*)	35	Tara baim (*Macrognathus aculeatus*)
11	Bata (*Labeo bata*)	36	Taki (*Channa punctata*)
12	Air (*Sperata aor*)	37	Kala bata (*Crossocheilus latius*)
13	Tit Puti (*Puntius ticto*)	38	Gutum (*Lepidocephalichthys guntea*)
14	Kanpona (*Aplocheilus panchax*)	39	Poma/Koitor poa (*Johnius coitor*)
15	Shalbaim (*Mastacembelus armatus*)	40	Mola (*Amblypharyngodon Mola*)
16	Foli (*Notopterus notopterus*)	41	Kalibaush (*Labeo calbasu*)
17	Mossambique Tilapia (*Oreochromis mossambicus*)	42	Mrigel (*Cirrhinus cirrhosus*)
18	White chingri (*Macrobrachium lamarrei*)	43	Kanpona/Blue panchax/Techokha/Tinchokha (*Aplocheilus panchax*)
19	Black Chingri (*Macrobrachium species*)	44	Pabda (*Ompok pabda*)
20	Dhela (*Osteobrama cotio*)	45	Couwa (*Gagatia cenia*)
21	Ghaura (*Clupisoma garua*)	46	Tilapia nilotica (*Oreochromis nilotica*)
22	Rui (*Labeo rohita*)	47	Kakila (*Xenentodon cancilla*)
23	Narikeli chela (*Salmostoma bacaila*)	48	Gulsha Tengra (*Mystus cavasius*)
24	Tengra (*Mystus vittatus*)	49	Nap[it koi (*Badis badis*)
25	Shing (*Heteropneustes fossilis*)	50	Jat puti (*Puntius shophore*)

Figure 1. Kechki net and it's catch.

Current net

Current net is also known as chapila net to the fisherman of Kaptai Lake. The specification this net is given below:

Mesh size : 1.5-3.5 cm
Length : 68.6-297.26 m
Wide : 1.83-5.4 m
CPUE : 0.22-4.0 kg
Haul time : 1.0-20 hour
Operation water depth : 4.83-32.93 m and
Species found : 23 species

Table 4. Species found in Current net in Kaptai Lake.

Sl.	Species name	Sl.	Species name
01	Chapila (*Gudusia chapra*)	13	Air (*Sperata aor*)
02	Boirali/ bori chapila (*Gonialosa manmina*)	14	Ayre (*S. seenghala*)
03	Long Chanda (*Nama chanda*)	15	Poma/Koitor poa (*Johnius coitor*)
04	Round Chanda (*Chanda baculis*)	16	Tengra (*Mystus bleekeri*)
05	Vacha (*Eutropiichthys vacha*)	17	Dhela (*Osteobrama cotio*)
06	Bele (*Glossogobius giuris*)	18	Jat puti (*Puntius sophore*)
07	Foli (*Notopterus notopterus*)	19	Bata (*Labeo bata*)
08	Shalbaim (*Mastacembelus armatus*)	20	Kalo Pabda (*Ompok pabo*)
09	Rui (*Labeo rohita*)	21	Kalibaush (*Labeo calbasu*)
10	Ek thuta (*Hyporhamphus limbatus*)	22	Kajoli (*Ailia coila*)
11	Shing (*Heteropneustes fossilis*)	23	Kechki (*Corica soborna*)
12	Gulsa Tengra (*Mystus cavasius*)		

Figure 2. Current net and it's catch.

Ecological Assessment of Inland Open Water Fisheries Population with Bio-physicochemical Properties to Frame EBFM Approach (Comp-E)

Researchers

Rabina Akther Lima, SO
Dr. Md. Robiul Awal Hossain, PSO
Dr. Azhar Ali, PSO (Additional Charge)
B.M. Shahinur Rahman, SSO
Md. Khaled Rahman, SO
Md. Lipon Mia, SO

Objectives

The main goal of this project is to improve biological management of Kaptai Lake that will restrict the declination of resources and ensuring its ability to naturally recruit and to conserve its biodiversity. Explicitly the objectives of the project can be outlined as follows:

- To assess bio-physicochemical properties of the Kaptai Lake including seasonal variation
- To assess stock of some important ecological fish groupwise. Herbivores, Detrivores, Carnivores and Omnivores based on catch and CPUE data
- To estimate population ecology and diet composition of some commercially significant Kaptai Lake fish
- To assist for framing ecosystem-based management approach for Kaptai Lake with emphasizing to increase productivity and conservation of the fisheries resources

Achievements

Experiment 01. Assessment of bio-physicochemical properties with seasonal variation in the Kaptai Lake

Seasonal variation of physicochemical properties

The study was conducted in four areas of the Kaptai Lake, they are i) Rangamati sadar ii) Longodu, iii) Kaptai and iv) Barkal. The sampling of respective areas water body has been done monthly for recording seasonal variation. Seasonal and yearly variation, range, mean values (x) and standard error (±SE) of different physico-chemical factors and relationship among the factors has been presented in Tables (1 and 2).

Seasonal variation of air temperature ranged from 27.35-30.42°C (Mean±SE 28.85±0.88). The highest air temperature (30.42±1.38°C) was recorded in monsoon season and lowest (27.35±1.73°C) in post-monsoon season. The fluctuation of water temperature varied from 26.17-28.39°C (Mean±SE 27.25±0.64°C). The maximum water temperature (28.39±1.98°C) was recorded in monsoon and minimum (26.17±1.50) in post-monsoon. The water temperature values showed close relationship with the air temperature.

Table 1. Seasonal and yearly variation of physico-chemical factors with range, mean values (x) and standard error (±SE).

Parameters (Unit)	Pre-monsoon	Monsoon	Post-monsoon	Mean±SE	Range
Air tem. (°C)	28.78±1.25	30.42±1.38	27.35±1.73	28.85±0.88	27.35-30.42
Water tem. (°C)	27.19±1.22	28.39±1.98	26.17±1.50	27.25±0.64	26.17-28.39
DO (mg/l)	6.23±0.39	5.62±0.55	6.84±0.24	6.23±0.35	5.62-6.84
DO saturation	75.38±4.83	68.13±10.06	60.67±4.32	68.06±4.24	60.67-75.38
pH	7.98±0.12	7.68±0.33	7.48±0.21	7.71±0.14	7.48-7.98
Conductivity (µS/cm)	103.92±5.30	93.17±7.72	107.40±16.27	101.49±4.28	93.17-107.40
ORP	118.35±13.75	110.06±13.54	116.79±14.36	115.07±2.54	110.06-118.35
Resistivity (KΩcm)	9.68±0.49	10.89±0.87	10.61±1.02	10.40±0.36	9.68-10.89
Alkalinity (mg/l)	87.61±1.30	66±4.72	81.47±6.77	78.36±6.42	66-87.61
Free CO_2 (mg/l)	13.89±1.63	11±2.08	13.43±1.74	12.77±0.89	11-13.89
Salinity (psu)	0.05±0.003	0.04±0.002	0.05±0.01	0.05±0.001	0.04-0.05
Transparency (m)	1.15±0.21	0.89±0.16	1.63±0.32	1.22±0.21	0.89-1.63
TDS (mg/l)	52.03±2.71	57.83±4.32	51.97±6.57	53.94±1.94	51.97-57.83
Depth (m)	3.84±1.34	10.11±6.34	12.07±6.71	8.68±2.48	3.84-12.07
Hardness (mg/l)	63.53±0.59	75.50±	80±9.22	73.01±4.91	63.53-80
Ammonia (mg/l)	0.17±0.05	0.02±.02	0.10±0.01	0.10±0.04	0.02-0.17

*NR=Not Recorded, ORP= Oxidation reduction Potential, TDS= Total dissolved solids.

Water temperature showed increasing and decreasing trend with air temperature. It also showed significant strong positive correlation with the air temperature ($r=0.914$, $p<0.01$) (Table 2).

The pH of water always found to be alkaline in nature and it varied between 7.48-7.98 (Mean±SE. 7.71±0.14). The pH showed strong negative correlation with ORP ($r=-0.694$, $p<0.01$)

Dissolved Oxygen ranged from 5.62-6.84 mg/l (Mean±SE. 6.23±0.35). The highest value (6.84±0.24 mg/l) was recorded in the post-monsoon and lowest (5.62±0.55) in monsoon season. Dissolved oxygen saturation was found to be ranged from 60.67-75.38 (Mean±SE 68.06±4.24). The highest value (75.38±4.83) was recorded in pre-monsoon and the lowest (60.67±4.32) in post-monsoon season. The oxidation reduction potential value was found to fluctuate from the range of 110.06-118.35 (Mean±SE 115.07±2.54). The highest value (118.35±13.75) was recorded in pre-monsoon and the lowest (110.06±13.54) in monsoon season.

Conductivity value was found to range from 93.17-107.40 µS/cm (Mean±SE 101.49±4.28). The highest value (107.40±16.27) was recorded in post-monsoon and the lowest (93.17±7.72 µS/cm) in monsoon season. Conductivity showed strong negative relation with resistivity (r=-917, p<0.01) and strong positive relation with salinity (r=0.974, p<0.05), hardness (r=0.82, p<0.01) and total dissolved solids (r=0.821, p<0.01). Resistivity value was found to range from 9.68-10.89 KΩ cm (Mean±SE 10.40±0.36). The highest value (10.89±0.87 KΩcm) was recorded in monsoon and the lowest (9.68±0.49 KΩcm) in pre-monsoon season. Salinity varied from 0.04-0.05 mg/l (Mean±SE 0.05±0.001). The salinity value was almost similar all the year round. Total dissolved solids (TDS) varied from 51.97-57.38 mg/l (Mean±SE 53.94±1.94). The highest value (57.83±4.32 mg/l) was recorded in monsoon and the lowest (51.97±6.57 mg/l) in post-monsoon season.

The value of total alkalinity was found to fluctuate from 66.00-87.61 mg/l (Mean±SE 78.36±6.42). The highest value (87.61±1.30 mg/l) was recorded in pre-monsoon and the lowest (66.00±4.72 mg/l) in monsoon season. The water transparency value varied from 0.89-1.63 m (Mean±SE 1.22±0.21). The highest value (1.63±0.32 m) was recorded in post-monsoon and the lowest (0.89±0.16 m) in monsoon season. Hardness varied from 63.53-80 mg/l (Mean±SE 73.01±4.91). The highest value (80±9.22 mg/l) was recorded in post-monsoon and lowest (63.53±0.59 mgl-1) in pre-monsoon season. Hardness showed moderate positive correlation with alkalinity (r=-0.687, p<0.05) and strong positive correlation with depth (r=0.875, p<0.05).

Phytoplankton and zooplankton showed no significant correlation with any physico-chemical parameters.

Table 2. Correlation among physico-chemical parameters and plankton of Kaptai Lake (Values are shown as r = coefficient correlation).

Parameters	AT	WT	DO	pH	Con	ORP	Res	Alk	CO_2	Sal	SD	TDS	Depth	Har	Amm	Phyt	Zoopl
AT	1																
WT	**.914****	1															
DO	-.431	-.568	1														
pH	-.148	-.225	.202	1													
Con	**-.638***	**-.615***	.330	.408	1												
ORP	.288	.247	-.094	**-.695***	-.470	1											
Res	.562	**.638***	-.192	-.394	**-.917****	.301	1										
Alk	**-.682***	-.565	.538	.469	.829	-.358	-.776	1									
CO_2	-.251	-.326	**.617***	.659	.480	-.212	-.398	.640	1								
Sal	**-.632***	**-.607***	.235	.428	**.974****	-.532	-.903	**.767****	.363	1							
SD	.256	.327	.473	-.029	-.203	-.008	.473	.084	.407	-.286	1						
TDS	-.434	-.430	-.135	.243	**.821****	-.406	**-.832****	.446	.159	**.810****	-.356	1					
Depth	-.022	.020	.327	.546	.452	**-.699***	-.143	.328	.413	.415	.471	.276	1				
Har	-.526	-.560	**.724***	.425	**.820****	-.694	-.582	.595	.622	**.725***	.184	**.740***	**.875****	1			
Amm	-.425	-.396	.591	.395	.219	-.088	-.254	**.687***	.552	.123	.235	-.177	-.038	-.175	1		
Phyto	.010	-.179	.437	.292	-.055	.403	-.162	.371	.289	-.083	.122	-.457	-.308	-.680	.055	1	
Zoopl	-.137	-.159	.097	-.079	-.017	.485	-.043	-.028	-.410	-.054	-.311	-.047	-.165	-.176	.027	.324	1

** correlation is significant at the 0.01 level. * Correlation is significant at the 0.05 level.

AT=air temperature, WT= water temperature, DO=dissolved oxygen Con=Conductivity, ORP=Oxidation reduction potentials, Res=resistivity, Alk=Alkalinity, sal=salinity, SD=Secchi disc, TDS=total dissolved solids, Har=hardness, Amn=ammonia, Phyto=Phytoplankton, Zoopl=zooplankton. The studied parameters were in standard limit and very close with previous studies in the Kaptai Lake (Table-03).

Table 3. Comparative study of parameters and plankton of Kaptai Lake with different studies.

Parameters	Present study	Ahmed *et al.* (1990, 91)	Karmakar *et al.* (2011)	Rahman *et al.* (2014)	Bashar *et al.* (2015)	Standard
Water temp. (^{o}C	27.25±0.64	32.10±2.1	-	27.09±5.89	27.09±5.89	25 to 30 ℃ (Rahaman *et al.*, 2013)
DO (mg/l)	6.23±0.35	7.42±0.90	5.90-6.90	6.4 ± 1.51	5.74±0.68	7.5 to 8.5 (Hoq *et al.*, 2007)
pH	7.71±0.14	7-8.4±0.40	5.36-6.93	7.6±0.52	7.49±0.37	6.5 to 8.5 (DoE, 2011)
Conductivity (µS/cm)	101.49±4.28	91.90-106.4	72-281	-	-	30-5000 µS/cm (Boyd, 1998)
Alkalinity (mg/l)	78.36±6.42	47.6-71.0	-	59.45 ± 6.71	59.45±6.71	50-150 mg/l (Boyd, 1998)
Transparency (m)	1.22±0.21	2.40±0.73	-	1.94 ± 0.57	1.94±0.57	1.6-6.09 m (DoE, 2011)
Free CO_2 (mg/l)	12.77±0.89	5-6	-	2.92 ± 0.60	2.93±0.60	6–10 (Boyd, 1998)
Salinity (ppt)	0.05±0.001	-	-	-	-	<0.5 ppt (Montagna *et al.*, 2013)
Hardness (mg/l)	73.01±4.91	47.6-65±	-	43.08 ± 5.51	43.08±5.51	40-400 mg/l (Boyd, 1998)
Ammonia (mg/l)	0.10±0.04	0.4±0.01	-	-	-	0.5 mg/l (WHO, 1993)
TDS (mg/l)	53.94±1.94	-	-	-	-	50-350 mg/l (Boyd, 1998)

Primary productivity of the Kaptai Lake

The monthly variations in gross primary productivity (GPP), net primary productivity (NPP) and community respiration (CR) values fluctuated from 0.17 gC/m^3/hr, 0.06 gC/m^3/hr and 0.05 gC/m^3/hr to 0.35 gC/m^3/hr, 0.29 gC/m^3/hr and 0.17 gC/m^3/hr, respectively. It was noted that GPP and NPP showed increasing trend from October to January and March to May (Figure 1).

In October to January lake water remains clear and facilitates sun light exposure, which increase the GPP and NPP.

Besides, high temperature and sun light during March to May facilitates GPP and NPP. Community respiration (CR) recorded highest increasing trend in February to April, indicates higher biological activity due to abundant phytoplankton and warm temperature.

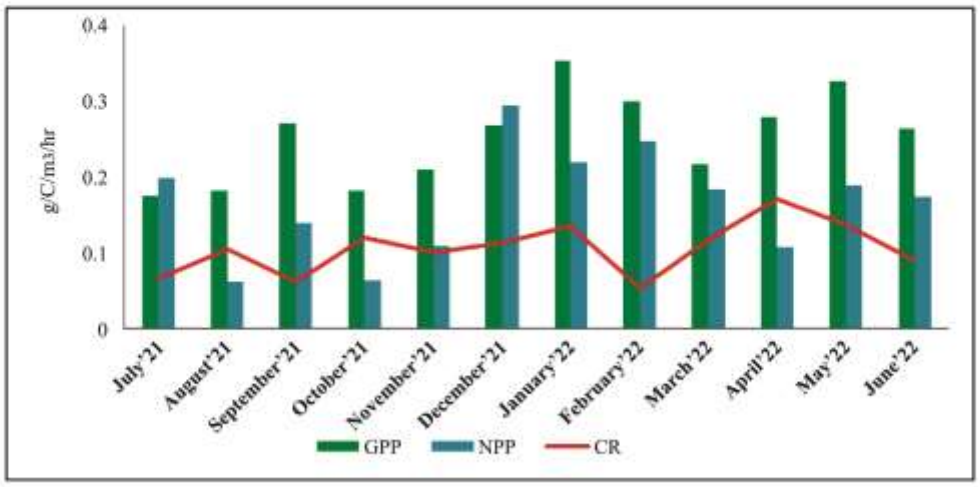


Figure 01. Monthly variation of primary productivity in Kaptai Lake.

Seasonal variation of plankton in the Kaptai lake

A plankton net of 90 µ mesh size was used for filtering water and collecting plankton samples for qualitative (preferably up to species level) and quantitative analysis. Abundance of plankton in three different seasons showed a wide range of variations (Figure 2 and 3). More than 52 genera of phytoplankton were identified under ten classes. Among them Cyanophyceae (33%) was dominating followed by Chlorophyceae (22%). Zooplankton genera was about 39 under five groups. Among them rotifera (43%) was the most dominant zooplankton.

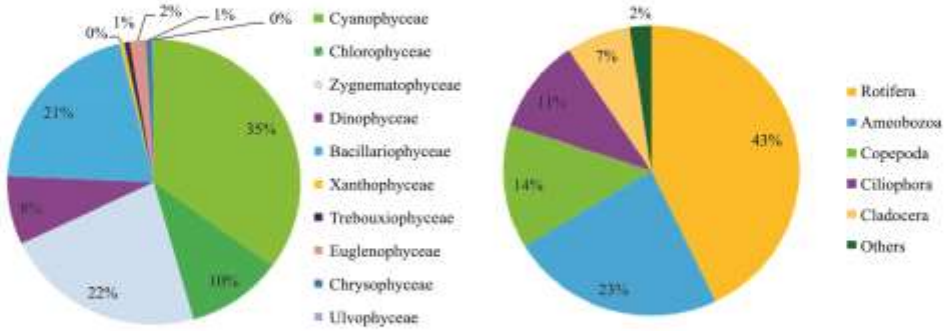


Figure 2. Species diversity of plankton (A. Phytoplankton and B. Zooplankton).

Phytoplankton abundance was highest (2.31×10^6 cells/l) was recorded in the pre-monsoon season. Whereas, phytoplankton abundance was recorded ($0.72\text{-}2.31\times10^6$ cells/1) in the post-monsoon and the monsoon season. Zooplankton abundance was 1.52×105 nos./1 was recorded highest in the pre-monsoon season and very low zooplankton abundance was found 0.61 ×105 nosL-1 in the monsoon season.

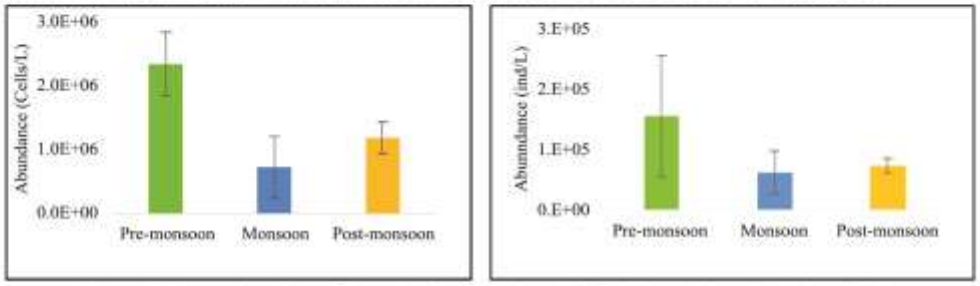


Figure 3. Temporal variation of phytoplankton abundance (left) and zooplankton abundance (right).

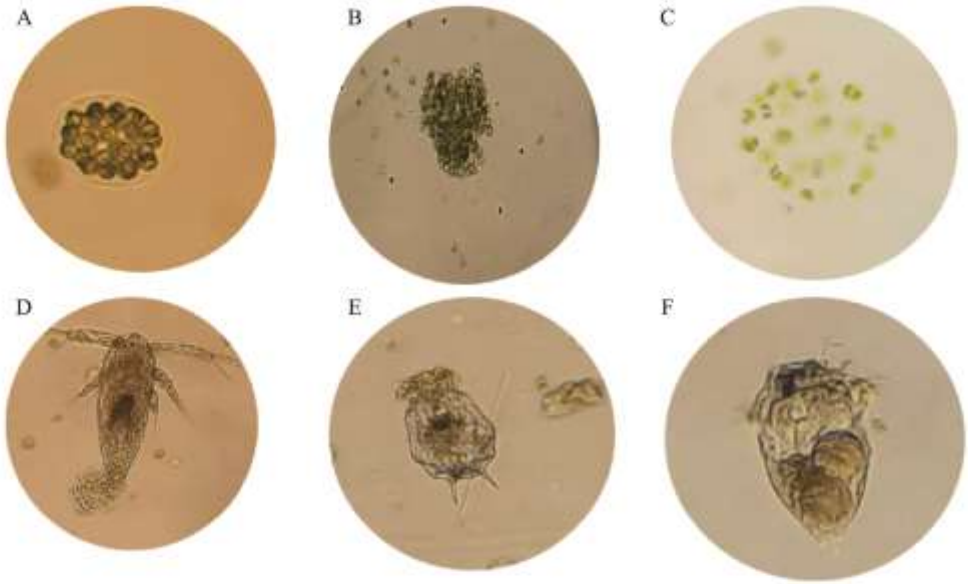


Figure 4. Phytoplankton and zooplankton observed under microscope (A. *Eudorina* sp., B. *Aphanothece* sp., C. *Dictyosphaerium* sp., D. *Cyclops* sp., E. *Lecane* sp., F. *Euchlanis* sp.) *in different* sampling stations of Kaptai Lake.

Plankton diversity indices

The diversity indices popularly used, including Shannon diversity index (H'), Margalef richness index (R), evenness index (J'), and Simpson diversity index (δ) were considered as explanatory variables of eutrophication levels. Shannon diversity index (H') ranged from 3.16 to 5.59, richness index (R) were 1.98-9.03, evenness index (J') started from 0.08-0.98 and Simpson diversity index (δ) varied from 0.18-9.03 respectively (Figure 5). All those indices represented diversified, integrated and semi -balanced phytoplankton community in the Lake water.

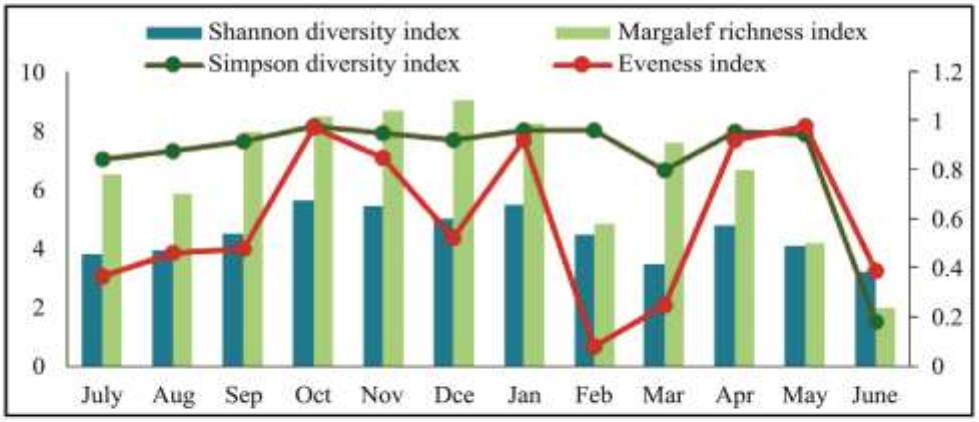


Figure 5. Monthly variation of phytoplankton diversity indices in the Kaptai Lake.

Exp 02. Assessment of some important ecological fish stock in groupwise based on catch and CPUE data

Feeding ecology of commercial fish

This study reported feeding habit of some commercial fish based on their relative gut of length and preference (Table 4).

Table 4. Feeding habit of some commercial fish in Kaptai Lake.

Fish Species	Relative length of gut	Food prefrence	Feeding habit
Puntius ticto	1.8-4.5	Phytoplankton (43%)	Omnivore
Mystus vittatus	1.1-3.8	Zoobenthos (22%)	Omnivore
Hyporhamphus limbatus	0.3-1.6	Zooplankton (28%)	Carnivore
Salmostoma phulo	1.2-2.8	Rotifera (31%)	Omnivore
Gudusia chapra	1.8-3.5	Chlorophyceae (67%)	Planktivore
Xenentodon cancila	0.3-1.4	Fish (38.9%)	Carnivore
Glossogobius giuris	0.4-0.9	Fish (46.7%)	Carnivore

CPUE data of some commercial fish in Kaptai Lake

Kechki net exhibited the highest CPUE (140 kg/haul) and cotton net of two part with lowest CPUE (0.0 kg/haul) (Table 05).

Table 5. CPUE of some commercial fish caught by different gear in the Kaptai Lake.

Gear Name	CPUE (kg/haul)	Haul time (hr)	Number of species caught
Kechki net	0.9-140	01-4.5	34
Current net	0.21-3.4	0.52-17	21
Cotton net	0.0-3.6	1.0-17	03
Tengra/Kajoli net	0.245-6.0	0.5-8	13
Mosquito pelling net (Chingri net)	1.0	0.9	08
Square shaped lift net (Dhormo jal)	0.35-2.2	0.5-1.0	15
Chingri trap	0.54-2.5	72-288	25
Seine net (Ber jal. One part)	0.67-5.0	0.5-3.0	15
Seine net (Ber jal. Two part)	0.46-1.2	0.5-1.0	3
Single hook (Borshi)	0.7-1.5	0.5-3.0	04
Longline hook (Borshi)	0.6-4.0	4.0-15	03
Chingri chai	0.45-5.6	15-20	10
Tengra chai	0.23-2.6	12-18	08

Ecological assessment of Stock

Fish catch data was analyzed and sorted to four feeding groups. We found ecosystem ratio, Planktivore:Detrivore:Carnivore:Omnivore = 43:1:15:41 (Figure 6).

Whereas, the standard ratio is, Herbivore/Planktivore:Detrivore:Carnivore:and Omnivore = 40:30. 20:10

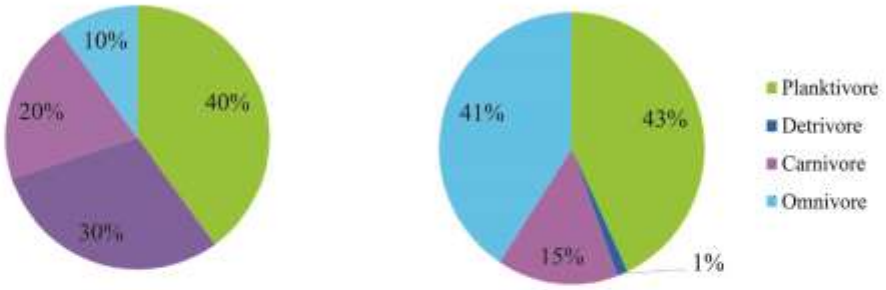


Figure 6. Ecosystem status of Kaptai Lake (Left. Ideal ecosystem; Right. Kaptai Lake ecosystem 2021-22.

In the present study, planktivore and omnivore fish were higher due to high percentage of Chapila (*G. chapra*) and Kechki (*C. soborna*) fish, detrivore fish were drastically lower and carnivore fish were lower than standard ratio (Figure 6). Therefore, Carnivorous and Detrivorous fish stocking and their proper management is time demanding for this ecosystem conservation.

Refinement and Validation of Culture Technology of Cuchia in Hill Tract Districts

Researchers

Md. Khaled Rahman, SO
Dr. Md. Robiul Awal Hossain, PSO
Dr. Azhar Ali, PSO
B. M. Shahinur Rahman, SSO
Rabina Akther Lima, SO
Md. Lipon Mia, SO

Objectives

The overall objectives of this proposed project is to understand the Cuchia fry rearing, enhancement, and ranching of this endangered species and to introduce this exportable fish into the aquaculture of Bangladesh for enhancement of income by the fish farmers. The specific objectives are:

- To develop culture technique of *M. cuchia*
- To disseminate *M. cuchia* culture in Chittagong hill tract districts
- To popularize Cuchia culture in Hill tract area

Achievement

Site selection

Six culture ponds were selected in five Upazilas of three hill districts of Chittagong division (Rangamati, Khagrachari and Bandarban). To fulfill the objectives of the experiment, the following design was followed.

Table 1. Design of the experiment.

Study area	Feed type	Stocking density
Rangamati	SIS (1.5% of BW) 3 days interval and Vermi Compost (1.5% of BW) Every day	$10/m^2$
Bandarban		
Khagrachori		

Figure 1. Pictorial view of site selection.

Pond Preparation

The experiment was conducted in 6 ponds with water area 40 m^2 for each. Ponds were dug with (30×15×3.5) ft^3. The pond bottom was covered by Polythene, knotless nylon net and triple then fill-up with 08-12 inch clay mud. The ponds were protected by fencing with a nylon net. The ponds were prepared by treating the soil with quick lime at the rate of 2 kg per decimal. Ponds were filled up with 0.6-0.8 m water and then dolomite at the rate of 15 ppm to strengthen the water's buffer capacity. After three days, the pond water was fertilized with Urea, TSP and MoP at the rate of 2.5 ppm, 3.0 ppm and 1.0 ppm, respectively to accelerate primary productivity. Water hyacinth and PVC pipe was used as shelter for Cuchia.

Figure 2. Pictorial view of pond preparation.

Stocking cuchia fingerlings

After sufficient plankton production, Cuchia fingerlings were stocked at a density of 10 individual/m2. Feeding and sampling is in progress.

Figure 3. Stocking of Cuchia in sampling sites.

Feeding

Fingerlings and vermi compost were fed according to design. Feed was supplied at night up to satiation level. Feed was supplied by a feeding tray to check the waste of feed.

Figure 4. Vermi compost.

Water quality parameters

Water quality parameters of all the sampling sites were monitored monthly by using a multiparameter. All the water quality parameters were in a suitable range for Cuchia culture (Table 2).

Table 2. Water quality parameters in culture ponds.

Water quality parameters	Rangamati		Bandarban		Khagrachari	
	P_1	P_2	P_1	P_2	P_1	P_2
Water temp. (^{0}C)	27.84±1.09	26.6±1.2	27.2±1.6	27.75±0.9	26.75±1.7	27.05±0.8
DO (mg/l)	7.06±0.5	5.59±0.9	8.28±0.39	6.29±0.6	6.28±0.8	6.63±0.5
pH	6.67±0.6	7.0±0.47	8.2±0.7	6.97±0.7	6.68±0.52	6.56±0.71
TDS (ppm, mg/l)	25.5±3.65	35.5±2.76	36±3.59	70.75±4.9	31.4±4.3	34.9±3.6
Alkalinity (mg/l)	58.3±3.4	70±5.67	74±4.89	110±9.45	55.3±3.5	55±4.77
Ammonia (mg/l)	0.1±0.01	0.2±0.01	0.2±0.01	0.1±0.02	0.2±0.01	0.2±0.01
Hardness (mg/1)	43±2.01	51±4.66	53±2.31	58±5.12	65±3.45	62±2.22

Figure 5. Water quality monitoring.

Growth performance of Cuchia

Cuchia growth of all the sampling sites was recorded monthly and sampling is going on. On average weight gain in one month was 25-30 g (Table 3).

Table 3. Growth performance of Cuchia in culture ponds.

Location	Pond	Initial		After 60 days	
		Length (cm)	Weight (g)	Length (cm)	Weight (g)
Rangamati	P_1	32.41±3.23	51.05±5.02	48.5±2.42	94.8±6.6
	P_2	32.26±2.79	50.26±5.74	48.45±2.52	95.67±5.56
	Mean±SD	**32.34±3.01**	**50.66±5.38**	**48.45±2.52**	**95.67±5.56**
Bandarban	P_1	33.71±2.78	51.53±3.94	52.45±2.34	103.5±7.45
	P_2	34.26±2.64	50.85±4.43	53.91±3.02	105.9±9.11
	Mean±SD	**33.99±2.71**	**51.19±4.19**	**53.18±2.39**	**104.7±6.7**
Khagrachori	P_1	34.11±2.81	51.94±3.99	51.03±2.41	101.77±6.2
	P_2	33.98±2.59	51.06±4.53	50.22±3.5	99.41±5.56
	Mean±SD	**34.04±2.7**	**51.5±4.26**	**50.63±2.79**	**100.59±7.2**

Figure 6. Growth monitoring of *Monopterus cuchia*.

Harvesting and Data Analysis.

After 60 days of the culture period, the highest mean weight was recorded in 104.7±6.7 g in Bandarban district and the lowest 95.67±5.56 g in Rangamati district.

Figure 07. Monopterus cuchia after two month rearing.

Breeding and Culture Potential of Marine Oyster and Green Mussel in the Bay of Bengal Bangladesh Coast (Comp-B)

Researchers

Abu Bakker Siddique Khan, SO
Md. Amirul Islam, PSO
Aovijite Bosu, SSO
Md. Rahamat Ullah, SO
Mousumi Akhter, SO

Objectives

- To develop culture techniques of oyster and green mussel in the Kuakata coastline
- To develop breeding techniques of oysters in captivity
- To develop larval rearing techniques of oyster

Achievements

Experiment-1. Development of culture techniques of oyster (Saccostrea cucullata)

Methodology

Site Selection

Gangamoti estuary of the Kuakata coast was selected for oyster culture in nature.

Seed Collection

Spats of oyster (*Saccostrea cucullata*) were collected from the Moheshkhali channel in Cox's Bazar and carried to RSS, Khepupara in the icebox with proper aeration. Oxygen tablets were supplied every four (4) hours to the icebox and the water quality was strictly maintained by changing seawater every six (6) hours.

Figure 1. Oyster, *Saccostrea cucullata.*

Raft culture method

Oysters were suspended from a raft using three (3) plastic baskets. Water depth was maintained at 1.0-1.5 m. The raft was eight (8) meter long and three (3) meter width. Thirty spats were stocked per basket.

Rack-string method

Oysters were suspended from the raft using a velon screen bag. Ten spats were suspended on racks in a velon screen bag.

Figure 2. Pictorial view of oyster research activities.

Water quality parameters

Water temperature, salinity, transparency, pH and dissolved oxygen (DO) were recorded every 07 days interval.

Calculation of specific growth rate (SGR)

The specific growth rate was calculated by following the formula of Bal and Jones (1960).

$$SGR = \frac{Log_e L_2 - Log_e L_1}{T_2 - T_1} \times 100$$

Where, L2 and L1 are the shell length at times T2 and T1. The growth was expressed in percentage per month.

Results

Water quality parameters in the culture site are presented in the Table 1. Water transparency and salinity were lower than the optimum level in the culture site (Table 1).

Table 1. Water quality parameters (mean ± SD) of Oyster and Mussel culture site, Kuakata.

Parameters	Gangamati estuary	Optimum range
Temperature (^{o}C)	28.7±0.3	22-32 ^{o}C (FAO, 2021)
Water transparency (cm)	69.3±0.5	110-140 cm (Haque *et al*., 2021)
Salinity (ppt)	19.6±0.6	27-35 ppt (FAO, 2021)
DO (mg/l)	5.02±0.2	>5.7 ppm (FAO, 20 21)
pH	7.56±0.2	8.1-9.3 (FAO, 2021)

Initial shell length and weight were 8.8±1.62 cm and 153.4±13.23 g, respectively. The final shell length and weight were 9.2±1.18 cm and 163.7±11.84 g, respectively. The specific growth rate was very low which is near to zero in the culture site (Table 2).

Table 2. Growth performance of *oyster, S. cucullata*

Species	Date	Shell length (cm)	Weight (g)	SGR (%)
Saccostrea cucullata	15.12.2021	8.8±1.62	153.4±13.23	-
	02.01.2022	8.9±1.32	156.7±11.27	0.03±0.01
	15.01.2022	9.1±1.49	161.4±10.93	0.07±0.02
	02.02.2022	9.2±1.04	163.2±12.51	0.03±0.01
	15.02.2022	9.2±1.18	163.7±11.84	0
	01.03. 2022	Not survived		

The survival rate of oyster, *S. cucullata* decreases throughout the experimental period. After 75 days rearing all the oysters died (Figure 3).

Figure 3. The survival (%) of oyster, *S. cucullata* in the Kuakata coast during 75 days of rearing.

Experiment-2. Development of culture techniques of green mussel (Perna viridis)

Methodology

Site Selection

Gangamoti estuary of the Kuakata coast was selected for green mussel culture in nature.

Seed Collection

Green mussel (*Perna viridis*) spats were collected from Moheshkhali channel, Cox's Bazar. Green mussels were transported with optimum aeration in the icebox. The icebox was supplied with oxygen tablets every four (4) hours, and the water quality was strictly maintained by changing seawater every six (6) hours.

Figure 4. Spats of green mussel (*Perna viridis*).

Raft rope culture method

Green mussels were suspended from a raft using plastic baskets. Water depth was maintained at 1.0-1.5 m. The raft was eight (8) m long and three (3) m width. Fifty spats were stocked per basket. The growth performance of green mussels was assessed fortnightly.

Figure 5. Pictorial view of research activities of green mussel.

Rack-string method

Green mussels were suspended from the raft using a velon screen bag. Ten green mussels were kept inside the velon screen bag and suspended from racks. Water quality parameters were checked every seven (7) days.

Results

After 18 days of stocking all green mussels died and no growth were observed in the culture site.

Table 3. Growth performance of green mussel (*Perna viridis*).

Species	Date	Shell length (cm)	Weight (g)
Perna viridis	15. 12.2021	8.8±1.62	153.4±13.23
	02.01.2022	Not Survived*	

*Reasons behind not survival of oyster and green mussel

Water quality parameters in the Gangamoti estuary were not suitable for the culture of oyster and green mussels (Table 1) though suitable water quality is a prerequisite for oyster and green mussel culture. Water transparency of 110-140 cm is needed for oyster and green mussel culture but in the culture site, it was 69.3±0.5 cm. Moreover, the salinity of 27-35 ppt is much required for the culture of oyster and green mussel, but in the culture site, it was 19.6±0.6 ppt, which was very low from the optimum level.

Identification, Culture and Bio-activity Analysis of Some Commercially Important Seaweed in Mid-Southern Coast of Bangladesh

Researchers

Md. Amirul Islam, PSO
Aovijite Bosu, SSO
Md. Monjurul Hasan, SSO
Abu Bakker Siddique Khan, SO
Md. Rahamat Ullah, SO
Farhana Yasmin, SO

Objectives

- To identify seaweed species grown at Kuakata coastline and the Sundarban areas
- To find out the potential area for seaweeds in this region
- To demonstrate the culture technique of seaweeds in the Kuakata coastline
- To investigate bioactive compounds and their activity of certain seaweed

Achievements

Study-1. Detailed survey and identification of natural growing seaweed species at the Kuakata coastline and the Sundarban areas

Methodology

A detailed survey was conducted to identify and collect the mangrove seaweeds. Samples were collected from Fatrar char, Patuakhali; Fakirhat, Barguna; Pasur and Sela river, Mongla, Bagerhat and Bhola river, Sarankhola, Bagerhat district. Field surveys in the research area were used to collect primary data. Key informants such as Upazila Fisheries Officers, seaweed researchers from various universities, and relevant NGO workers were interviewed for cross-checking. Secondary Data on seaweed in Bangladesh were collected from published and unpublished documents, relevant Government and Non-Government organizations, research institutes, and universities.

Results

Samples were collected from Fatrar char, Patuakhali, and Fakirhat, Barguna districts as well as from the Pasur and the Sela river of Mongla upazilla and Bhola river of Sarankhola, Bagerhat district. Six species were found in this survey i.e., *Catanella* sp., *Rhizogonium* sp., *Calaglossa* sp., *Bostisia* sp., *Enteromorpha* sp., and *Colpomenia* sp. (Figure 1).

Figure 1. Identified seaweed species at the Kuakata coastline and the Sundarban areas.

Experiment-2. Feasibility of seaweed Gracilaria tenuistipitata culture in the Kuakata coastline, Bangladesh

Methodology

Site selection

Three sites were selected at the Midpoint of Gangamati, Gangamati Estuary, and 33 Kani in the Kuakata coastline.

Seaweed culture method

Young, growing fragments of *Gracilaria tenuistipitata* were collected from Bakkhali estuary; Cox's Bazar and culture were started at the Kuakata coast on 15 December 2021 and continued for 90 days. Three horizontal nets were used with 22 cm of mesh size. The area of each net was 16 m2. Initial seaweed weight was 1 ± 0.05 kg fw/m^2 and initial length was 20±0.54 cm.

Water quality parameters monitoring

Water temperature, salinity, transparency, pH, and dissolved oxygen (DO) were recorded every 7 days interval at the cultivation sites using a multiparameter water test kit (HANNA, HI98194).

Daily growth rate

Every 15 days of culture, the daily growth rate (DGR) %/day was computed using Hung et al., (2009) formula as follows.

$$DGR = [(Wt/W0)1/t - 1] \times 100\ \%/day$$

where, W0 is the initial fresh weight, Wt is the final fresh weight, and t is the days of culture.

Biomass yield

Harvesting was done by clipping the algae hanging on the rope leaving the base on the rope to grow further. Seaweed biomass is expressed as the fresh weight of seaweed per unit culture area (kg/m^2) and computed with the following modified formula of Doty (1986).

$$Y = (Wt - W0)/A$$

where. Y = biomass production; Wt= fresh weight at day t; W0 = initial fresh weight; A = area of net.

Statistical analysis

Statistical Package for Social Sciences (SPSS) software version v25.0 and Microsoft Office Excel 2019 were used to analyze the data. For comparisons, a one-way ANOVA was used with Tukey's HSD post-hoc analysis. In this investigation, the level of significant was set at $P<0.05$.

Results

Water quality parameters

Seaweed cultivation requires the use of appropriate water quality parameters. Table 1 shows the results of water quality parameters in the culture sites of the Kuakata coast, Bangladesh. All the water quality parameters were in the optimum range but the salinity and water transparency level were slightly lower than the optimum level (Table 1).

Table 1. Water quality parameters (mean ± SD) of seaweed culture sites, Kuakata.

Parameters	Mi dpoint of Gangamati	Gangamati Estuary	33 Kani
Water Transparency (cm)	69.3±0.5	55.65±0.5	63.70±1.2
Salinity (ppt)	19.60±0.6	19.80±0.8	19.60±0.3
Water temperature (^{o}C)	28.70±0.3	28.20±0.3	28.60±0.1
pH	7.56±0.2	7.63±0.2	7.51±0.1
DO (mg/l)	5.02±0.2	4.90± 0.2	5.50±0.1

Daily growth rate (DGR)

The maximum daily growth rate was found on the 60th day in Gangamati Estuary (2.49±0.15 %/day), while the minimum daily growth rate was found on the 15th day in the Midpoint of Gangamati (0.85±0.05%/day) (Table 2). The DGR of three sites was attributed to significant changes ($P<0.05$).

Table 2. Daily growth rate (%/day) of seaweed *Gracilaria tenuistipitata* at three culture sites of Kuakata coast for 90 days of culture period.

Culture Duration (Day)	Midpoint of Gangamati (Mean ± SD)	Gangamati Estuary (Mean ± SD)	33 Kani (Mean ± SD)
15	0.85 ± 0.05^{b}	0.97 ± 0.06^{a}	0.92 ± 0.03^{a}
30	1.45 ± 0.06^{c}	1.88 ± 0.07^{a}	1.70 ± 0.06^{b}
45	1.72 ± 0.08^{c}	2.21 ± 0.12^{a}	1.89 ± 0.07^{b}
60	1.86 ± 0.11^{c}	2.49 ± 0.15^{a}	2.14 ± 0.10^{b}
75	1.66 ± 0.09^{b}	2.14 ± 0.10^{a}	1.86 ± 0.09^{ab}
90	1.42 ± 0.12^{b}	1.95 ± 0.08^{a}	1.67 ± 0.06^{b}

Mean values in the same row having different letters are significantly different ($P<0.05$).

Production

The biomass production was significantly higher in the Gangamati Estuary (6.21 ± 0.97 kg/m^2) and lowest in the Midpoint of Gangamati (4.58 ± 0.67 kg/m^2) (Table 3).

Table 3. Production of *Gracilaria tenuistipitata* at three culture sites, Kuakata.

Production	Midpoint of Gangamati (Mean ± SD)	Gangamati Estuary (Mean ± SD)	33 Kani (Mean ± SD)
K g/m ^{2}	4.58 ± 0.67^{b}	6.21 ± 0.97^{a}	5.43 ± 0.78^{ab}

Mean values in the same row having different letters are significantly different ($P<0.05$).

Experiment-3. Effect of stocking densities and water depth on the growth and production performances of red seaweed, Gracilaria tenuistipitata in the Kuakata coast, Bangladesh

Methodology

Study area

Gracilaria tenuistipitata trial farming was carried out in the Mid Southern Kuakata coast of Bangladesh (21˚48'21.3" N and 90˚08'44.5" E) from December 2021 to March 2022.

Effect of stocking densities

To determine the optimum stocking density for growth and biomass yield in the culture system, four separate stocking densities, 1, 2, 3, and 4 kg/m^2, were trialed in square rafts (1 m × 1 m) at the water surface. In the square rafts method, seaweeds were cultivated for 90 days before being collected.

Effect of water depths

Gracilaria tenuistipitata was cultured to find the best water depth for growing in the square raft (1 m x 1 m)

using three separate depths as treatments in three replications; surface, 0.5 m, and 1.0 m below the water surface. In each raft, the preliminary seaweed stocking density was 1 kg/m^2.

Growth rate and biomass yield

To measure seaweed's daily growth rate (DGR), tuft weights were taken every 15 days interval and Hung et al.'s (2009) formula were used to determine the daily growth rate as follow:|

$$DGR = [(Wt/W0)1/t - 1] \times 100\ \%/day$$

Where, W0 is the initial fresh weight, Wt is the final fresh weight, and t is days of culture.

Biomass (Y) was calculated as mean (kg/m^2) using a modified Doty (1986) equation that incorporated the propagules' starting weight as follows:

$$Y = (Wf\text{-}Wi)/m^2$$

Where, Wf is the final fresh weight and W0 is the initial fresh weight. This was used for the Y determination of the square raft method.

Statistical analysis

All data were provided as means with standard error (SE) from three replications. Statistical analyses were performed using GraphPad Prism 9 and the Statistical Package for the Social Sciences (SPSS) version 25.0 (SPSS Inc., Chicago, IL, USA). The significance of each variable across different treatments was determined using a one-way ANOVA. The ANOVA was followed by Tukey's test if a mean effect was significant. The 95 % confidence level was used to determine the significance.

Results

Effect of stocking densities

Seaweed *G. tenuistipitata* was affixed to a square raft and allowed to grow out at various stocking densities in this experiment. The results showed that stocking density had a significant impact on daily growth rate ($P<0.001$), although there was no significant difference ($P>0.05$) between 1 kg/m^2 and 2 kg/m^2 stocking densities (Figure 2). The maximum daily growth rate was 2.27% per day for the 2 kg/m^2 stocking density at 60 days, and it dropped to only 0.59% per day for the 4 kg/m2 density at 90 days as stocking density increased.

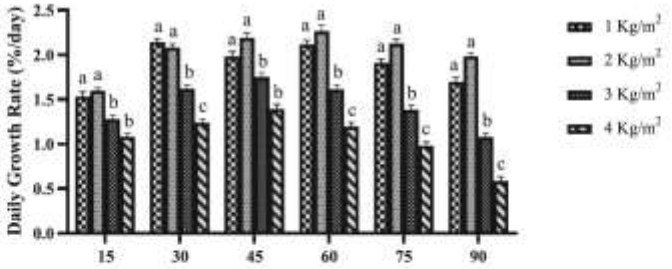


Figure 2. Daily growth rates (DGR) of G. tenuistipitata for 90 days at four stocking densities in the Mid Southern Kuakata coast of Bangladesh. Error bars are standard errors. Different small letters over bars denote significant differences at $P< 0.05$.

Since the growing performance of seaweed stocking densities of 1 and 2 kg/m^2 was comparatively better, periodical significance was examined separately for 1 and 2 kg/m^2. In the initial phase, the DGR of *G. tenuistipitata* increased significantly ($P<0.05$) as time passed for both stocking densities, but it dropped significantly ($P< 0.05$) after the cultivation of 60 days (Figure 3).

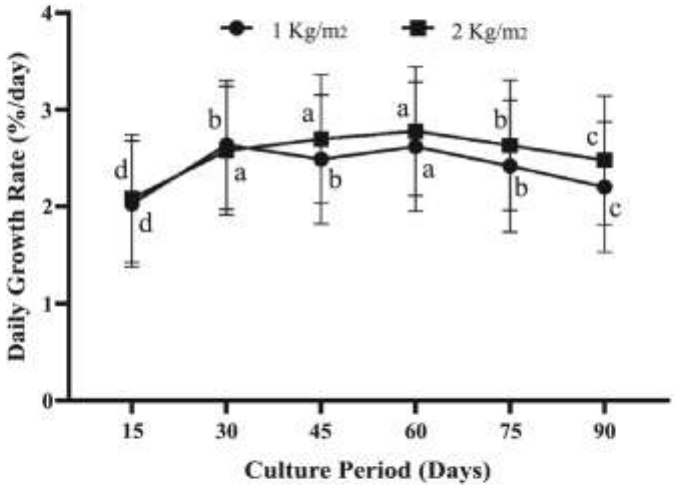


Figure 3. Daily Growth Rates (DGR) of *G. tenuistipitata* for 90 days at two stocking densities on the Mid Southern coast of Bangladesh. Error bars are standard errors. Different small letters denote significant differences at $P< 0.05$.

In comparison to DGR, biomass production per raft was highest (13.52±2.04 kg/m^2) for the 4 kg/m^2 stocking density and lowest (6.21±0.97 kg/m^2) for the 1 kg/m^2 stocking density (Table 4), even though there was no significant difference between 2, 3, and 4 kg/m^2 stocking densities.

Table 4. Biomass yield of *G. tenuistipitata* cultivated in Mid Southern Kuakata coast of Bangladesh at different stocking densities (from 1 kg/m^2 to 4 kg/m^2).

Stocking density (kg/m^2)	Biomass yield (kg/m^2)
1	6.21 ± 0.97^{b}
2	11.63 ± 1.16^{a}
3	12.89 ± 1.24^{a}
4	13.52 ± 2.04^{a}

Effect of water depths

The figure 4 shows the DGR (percent) statistics at three different water depths. Seaweed *G. tenuistipitata* grew at different rates (P< 0.05) depending on the water depths. The maximum DGR (2.39% per day) was observed at 0.5 m water depth at 60 days on the Kuakata coast, and the lowest DGR (1.38% per day) was reported at 1 m water depth at 15 days.

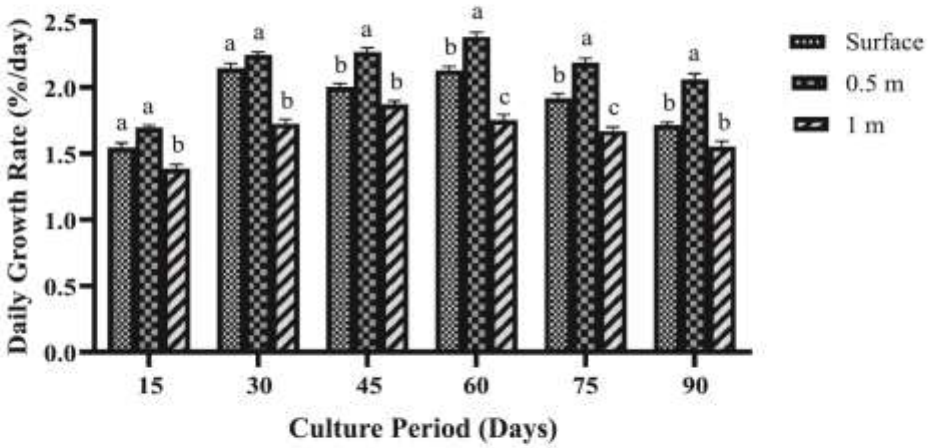


Figure 4. Daily Growth Rates (DGR) of *G. tenuistipitata* for 90 days at three water depths on the Mid Southern Kuakata coast of Bangladesh. Error bars are standard errors. Different small letters over bars denote significant differences at P< 0.05.

The biomass production of *G. tenuistipitata* at various water depths during the course of 90 days is shown in table 5. Biomass yield (8.53±1.06 kg/m^2) was significantly greater (P<0.05) at 0.5 m water depth, but there was no significant variation in biomass production between surface and 1.0 m depth. The results stated that the biomass yields of *G. tenuistipitata* were influenced significantly (P< 0.05) by water depths.

Table 5. Biomass yield of *G. tenuistipitata* cultivated in the Mid Southern Kuakata coast of Bangladesh at different water depths (from surface to 1.0 m). Values are means ± SE. Means with the same superscripts are not significantly different (P = 0.05).

Water depth (m)	Biomass yield (kg/m^2)
Surface	6.21 ± 0.97^{b}
0.5	8.53 ± 1.06^{a}
1	5.84 ± 0.86^{b}

Experiment-4. Determination of bioactive compounds and analysis of their activity from certain seaweeds

Methodology

The sample of *Gracilaria tenuistipitata* seaweed was collected and sent to Shrimp Research Station (SRS), Bagerhat for antioxidant activity analysis.

Antioxidant activity of seaweed *Gracilaria tenuistipitata*

2, 2-Diphenyl-1-picrylhydrazyl (DPPH) free-radical scavenging assay

2,2-Diphenyl-1-picrylhydrazyl free-radical scavenging assay was used to determine the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging capacity of the extract. In brief, 3.9 ml of ethanolic DPPH (60 mM) was first mixed with 0.1 mL of undiluted extract or ethanol (as control) and stored in a dark environment at room temperature for 30 minutes. Subsequently, the absorbance of extract and control was measured against ethanol (as blank) at 517 nm using a UV spectrophotometer. The absorbance measurements of extract and control were carried out in triplicate. The percentage of DPPH free radical scavenging capacity was calculated using the following formula.

DPPH free-radical scavenging activity (%) = [1 - (As/Ac)] x100%

where, As is the absorbance of crude extract at 517 nm and Ac is the absorbance of control at 517 nm. The samples of blank and control (0.01 mg/ml standard Trolox) were analyzed.

ABTS free-radical scavenging assay

ABTS radical scavenging capacity assay was performed according to the procedures described by Cai et al., (2015). The ABTS radical solution was first prepared by mixing 10 mL of 7 mM ABTS solution with 10 mL of 2.45 mM potassium persulfate solution in an amber bottle. Subsequently, the ABTS radical solution was allowed to stand in a dark environment at room temperature for 12 to16 hours to give a dark blue solution. The ABTS radical solution was then diluted with denatured ethanol until its absorbance was equilibrated to 0.7 ± 0.02 at 734 nm before usage. Approximately 3.9 mL of ABTS radical solution was first mixed with 0.1 mL of undiluted extract or ethanol (as control) and the mixture was stored in a dark environment at room temperature for 6 minutes. Subsequently, the absorbance of oil and control was measured against ethanol (as blank) at 734 nm using a UV spectrophotometer. The absorbance measurements of crude and control were carried out in triplicate. The percentage of ABTS free-radical scavenging activity was calculated using the following formula.

ABTS free-radical scavenging activity (%) = [1 - (As/Ac)] x 100%

Where, As is the absorbance of oil at 734 nm and Ac is the absorbance of control at 734 nm. The samples of blank and control (1.0 mg/ mL standard Trolox) were analyzed.

Determination of gallic acid from *Gracilaria tenuistipitata*

Collected seaweeds were prepared for extraction. After extraction, the bioactive compound was determined.

Solvent extraction

Methanol, ethanol, and water were used as solvents for solvent extraction. About 30g of sun-dried crashed samples and 300 ml of solvent were placed in beakers and stirred overnight at 40°C at 250 rpm. After extraction, the solvent was evaporated by a rotary vacuum evaporator at 50°C. The extract was collected in a vial and stored at -60°C until further analysis.

Determination of gallic acid using HPLC

Chromatography analysis for the quantification of chemical marker gallic acid in *Gracilaria tenuistipitata* was done using HPLC, ran with clarity software, equipped with symmetry C18 (4.6 mm x 250 mm x 5 µm) column and 3250 UV vis detection of Sykam Company, Germany. The mobile phase was composed of acetonitrile (A) and 0.1% phosphoric acid (B) in gradient elution mode. Gradient elution was performed as follows. 0-2 min, 92 % B, 2 min 90% B, 4 min 86% B. The flow rate of mobile phase was kept at 1.0 ml per min using 261 nm as the preferred wavelength. The temperature of the column was kept at 35°C and the sample injection volume was 500 µL.

Results

Antioxidant activities

Higher antioxidant activity (DPPH and ABTS) of *G. tenuistipitata* was found in ethanol extract than hexane extract (Table 6).

Table 6. Antioxidant activity of seaweed *G. tenuistipitata* with DPPH and ABTS reagent.

Extract using different solvents	DPPH activity	ABTS activity
Ethanol Extract	78.26±2%	86±0.3%
Hexane Extract	76.37±2%	84±0.2%

Determination of gallic acid

Gallic acid content of *G. tenuistipitata* was higher in methanol extract than in ethanol extract (Table 7).

Table 7. Amount of gallic acid of *G. tenuistipitata* in different solvents.

Extract using different solvent	Gallic acid (µg/ml)
Ethanol Extract	34.894±0.30
Methanol Extract	62.743±0.36

Development of Mariculture Practice of Seabass (*Lates calcarifer*) in the South-West Coast of Bangladesh (Component-C)

Researchers
Aovijite Bosu, SSO
Md. Amirul Islam, PSO
Md. Monjurul Hasan, SSO
Abu Bakker Siddique Khan, SO
Farhana Yasmin, SO
Mousumi Akhter, SO

Objectives

- To develop cage culture technique of seabass (*Lates calcarifer*) in coastal water of Bangladesh
- To study growth and survival of seabass in net cage
- To develop brood stock of seabass in coastal environment

Achievements

Experiment 1. Culture of Seabass (Lates calcarifer) using net cages in tidal river system

Methodology

Study area

Experiment was carried out in protected area of the Andharmanik river, near to the riverine Sub-Station, Khepupara, Patuakhali. The site preferably located in an area, where influence of tidal fluctuation was not pronounced with salinity ranging from 5-15 ppt and suitable for seabass culture. The site was far from the area, where biofoulers abound and far from the sources of domestic, industrial and agricultural pollution and other environmental hazards.

Rearing of *L. calcarifer* fry in ponds.

Before going to culture in net cages, seabass fry were reared in ponds.

Pond preparation

Two ponds situated on the bank of the Andharmanik river (each of 40 decimal) of RSS, Khepupara were selected. Ponds were prepared following standard method before stocking of seabass fry. To get rid of unwanted fish species, the pond's bottom was dried out. Drying ensured that hazardous compounds were oxidized and organic matter was mineralized. Floods can carry away the pond's fish during the rainy season, which is a common problem for most ponds. Despite the pond's proximity to a river or stream, it required a taller embankment or dike. Two earthen ponds at BFRI were 2 to 3 feet higher than the pond's highest water level. While excavating or de-mudding, this was achieved quickly and effortlessly. For

the pond system to work properly, it needed an effective inlet and outlet system. The pond's entry system shape of a pipe through which water enters and departs the system was kept slightly higher than the output system to achieve maximum water flow. The removal of cannibalistic and undesired fish was very important in the pond preparation process. Cannibalistic fish such as shol, gozar, boal, taki were eradicated from the pond by drying it out. Lime was applied at 500g/decimal in two experimental ponds during pond preparation. Fertilizers were also applied in those experimental ponds, urea and TSP at 150 and 75 g/decimal, respectively.

Figure 1. Pictorial view of pond liming.

Collection and stocking of Seabass fry

Seabass fries were collected from the Kholpetua river of Ashasuni upazilla under Satkhira district. After acclimation, fry of the same size (25.6±7.21 g) was stocked into the ponds on the August 25, 2021, at a stocking density of 14 fry/decimal.

Feed and feeding of seabass fry

Live feed (Tilapia fry, silver carp fry, shrimp etc.) was applied at 10% of total biomass of the seabass fry. Feeding was done twice daily in the morning at 08.00 hours and afternoon at 17.00 hours.

Figure 2. Pictorial view of feeding the seabass fry.

Water quality parameters monitoring.

Water quality parameters *viz*., pH, dissolve oxygen, salinity, ammonia of pond water were measured fortnightly using a multiparameter water test kit (HANNA, HI98194) recorded.

Figure 3. Pictorial view of water quality parameter monitoring.

Growth parameters monitoring

Growth of Seabass was monitored monthly. Length of fish was measured by measuring scale and weighed by digital weighing machine.

Figure 4. Pictorial view of length-weight measurement of Seabass.

Results

Water quality parameters

All the water quality parameters of seabass rearing ponds were within the optimum range of seabass rearing in the ponds, except salinity of pond water (Table 1). Lower salinity of pond water might be due to the variation of salinity of the Andharmanik river.

Table 1. Water quality parameters of seabass rearing ponds.

Month	Water Temperature (^{o}C)	pH	Ammonia (ppm)	Salinity (ppt)	DO (ppm)
August	30.7	7.10	0.00	0.25	5.93
September	32.2	7.83	0.01	0.16	5.82
October	30.4	7.33	0.01	0.89	5.38
November	28.7	7.55	0.02	3.00	4.37
December	23.2	7.43	0.03	5.56	4.69
January	21.4	7.2	0.02	2.05	5.88
February	22.7	7.3	0.03	1.99	4.53
March	27.8	7.6	0.02	4.04	4.64
Mean±SD	27.14± 4.14	7.2 ±0.49	0.02 ±0.008	2.24 ±1.89	4.95 ±0.58

Growth performances of seabass fry in the ponds

Mean initial weight of seabass fry was 23.6±7.21 g. After rearing of 228 days, mean weight of pond 1 and pond 2 were 320.69± 221.84 and 333±115.78 g, respectively. The higher survival (55%) was also found in pond 2 than pond 1 (Table 2).

Table 2. Growth performances of seabass fry in ponds for 228 days of rearing.

Parameters	Pond 1	Pond 2
Stocking density	14/deci.	14/deci.
Total nos. of fingerlings	550	550
Initial mean length (cm)	8.25±2.14	10 .15±1.95
Initial mean weight (g)	23.6±7.21	23.6±7.21
Final mean weight (g)	320.69± 221.84	333±115.78
Survival rate (%)	52	55

Culture of Seabass (*Lates calcarifer*) in net cages

Methodology

Design and construction of net cages

The net cages were attached to wooden, GI pipe and bamboo frames. Cages were kept afloat by floating material such as metal, plastic, foam drum. The shape of the cage was maintained with the use of concrete weights attached to the corners of the cage bottom. Volume of a floating cage was 64 m^3. Six (6) Cages were constructed by 1 cm mesh size knotless polythene nylon net. Cages were set in the Andhermanik river.

Stocking of seabass juveniles in cages

Prior to stocking seabass juveniles in the cages, fish was acclimatized to the ambient temperature and salinity prevailing in the cages. Stocking was done following the experimental design denoted 64 juveniles/cage as T_1, 96 juveniles/cage as T_2 and 128 juveniles/cage as T_3. After establishment of cages, seabass juveniles were stocked in the cages on the 13th April, 2022.

Figure 5. Cage setting and establishment in the Andhermanik river.

Feed and feeding

Live feed (Tilapia fry, silver carp fry, shrimp etc.) was supplied at 10% of total biomass. Feeding was done twice daily in the morning at 08.00 hours and afternoon at 17.00 hours. After two months of culture 30% live feed was replaced by formulated feed.

Figure 6. Pictorial view of feeding and sampling of the Seabass.

Results

Water quality parameters of cage site of the Andhermanik river

Water quality parameters of cage site of the Andhermanik river are shown Table 3. All the water quality parameters found to be congenial for seabass growth. The lowest salinity of water was found in the month of August 2021 and it increased gradually to the highest (13.79 ppt) in the month of April 2022, when the seabass juveniles were stocked in the cages.

Table 3. Water quality parameters of Andharmanik river (cage site).

Month	Water Temperature (^{0}C)	pH	Ammonia (ppm)	Salinity (ppt)	DO (ppm)
15 Aug 21	30.7	7.5	0.00	2.25	6.14
15 Sep 21	32.9	7.54	0.0	2.48	5.31
15 Oct 21	30.7	7.26	0.19	4.74	5.36
15 Nov 21	26.7	6.75	0.2	6.65	4.32
15 Dec 21	23.8	6.43	0.01	8.82	6.21
15 Jan 22	21.5	7.27	0.02	11.54	5.47
15 Feb 22	22.84	7.8	0.03	13.65	4.53
15 Mar 22	29.72	7.9	0.01	10.12	5.98
15 April 22	30.73	7.8	0.01	13.79	6.27
15 May 22	30.83	7.5	0.01	7.54	6.39
15 June 22	31.31	7.4	0.01	2.19	6.50
Mean±SD	28.34±3.93	7.59±.023	0.01±0.05	7.88±4.64	5.86±.047

Growth performances of Seabass in the net cages

After a culture period of 60 days, the growth performances of *Lates calcalifer* in terms of length and weight gain were found to be satisfactory. From the results, it was observed that stocking density had effect on the growth of seabass juveniles (Table 4). The highest mean weight (698±230.06) was obtained in T_1 than other Treatments.

Table 4. Growth performances of seabass in the net cages for a culture period of 60 days in the Andhermanik river.

Parameters	T_1	T_2	T_3
Stocking density	64	96	128
Initial Avg. length (cm) (13 April 2022)	29.61±4.88	28.12±4.58	22.12±4.44
Initial Avg. weight (g) (13 April 2022)	335±132.65	290±115.65	138±102.65
Avg. length of the last sampling (cm) (13 June 2022)	39.7 ±6.58	37.5±5.73	28.44±5.40
Avg. weight of the last sampling (g) (13 July 2022)	698±230.06	562.17± 191.46	225±126.45

Population Dynamics of Important Fish and Shell Fish in the Sundarbans Mangrove of Bangladesh

Researchers

Md. Hashmi Sakib, SSO and PI
Shawon Ahmmed, SO
Rafia Afrin, SO
Md. Reaz Morshed Ranju, SO

Objectives

- To assess the abundance and to estimate growth parameters of important fish and shell fish species,
- To calculate the mortality rate and exploitation level of selected species, and
- To identify vulnerable size groups of a fish species in the Sundarbans

Achievement

Study 1. Estimation of abundance and growth parameters of selected fish/crustacean species in the Sundarbans mangrove river

Site selection

The Sundarbans mangrove territory consist of three districts namely Bagerhat, Khulna and Shatkhira. The forest lies a little south to the Tropic of Cancer between the latitudes 21°30'N and 22°30'N, and longitudes 89°00'E and 89°55'E. A total of 13 major rivers flow through the Sundarbans and met to the Bay of Bengal. Among 13 rivers, the Shibsha, the Arpangasia (lower stream of the Kholpetua) and the Pashur river of Khulna, Satkhira and Bagerhat district, respectively were selected for sampling. Single sampling Station/spot from each river was selected as, Hodda in the Sibsha river; Nildumur and Gabura at the joint betwen the Arpangasia and the Kholpetua river, and Karamjol spot of the Pashur river (Figure 1).

Species selection

To begin with, this study has been being progressed through different published and unpublished literatures from BFRI library and open access libraries for getting updated data on the species in the selected areas (Figure 2). Meanwhile, procured data was analyzed to extract core messages by conducting a critical literature review (CLR) method. The CLR evaluates scholarly articles on a specific subject matter by analyzing in a systematic way according to the objectives for the purpose of apprehending the trends, enhancing understanding, and getting insights into relevant studies about a subject of inquiry (Saunders et al., 2003). This method also assists a researcher to ensure the quality of the content in a study by identifying the most related and important matters to include in the findings (Saunders and Rojon, 2011). As a result, some suitable species had appeared for conducting study on population dynamics in the Sundarbans fisheries. At the initial phase, five commercially important species already were taken from three groups of the fish i.e., fish, crustacean and mollusk. From the fish group, spotted scat *Scatophagus argus* (Linnaeus, 1766), tank

goby *Glossogobius giuris* (Hamilton, 1822) and grey-eel catfish *Plotosus canius* were opted. Similarly, mud crab *Scylla olivacea* (Herbst, 1896) was picked up from the crustaceans. In addition, one mollusk, blood cockle *Tegillarca granosa* was considered for an assessment to get the important population parameters.

Focus group discussion (FGD) is regularly used as a qualitative approach to gain an in-depth understanding of societal response towards a particular issue (Nyumba et al., 2018). Through this qualitative study, the salient features of stock status, catchability and particular place of species availability were explored. One FGD were arranged with 10 participants for every single river to validate quantitative results. The relevant participants from the community of fishery stakeholders including different age groups viz. young, middle age, old age fisherman; Local leader; Professional leaders; social leaders, fish traders were considered.

Sampling was done monthly basis either during full moon or during new moon period (considering lunar cycle) for a period of 07 months from Dec 2021 to June 2022, using three types of gear such as hooks, nets and traps in this area for crab harvesting fish and shell-fish. A day long fishing operation were operated for understating the catch composition of the species and abundance in rivers at the Sundarbans.

Total length (TL) in cm and total body weight (BW) in g for each individual were measured using measuring scale and an electronic balance, respectively. We were calculated Length-Weight Relationship (LWR) using the equation.

$$BW=a \times TL^{b} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (1)$$

Where, BW is the total body weight (g), and TL is the total length (cm).

Figure 1. The sampling sites in the map.

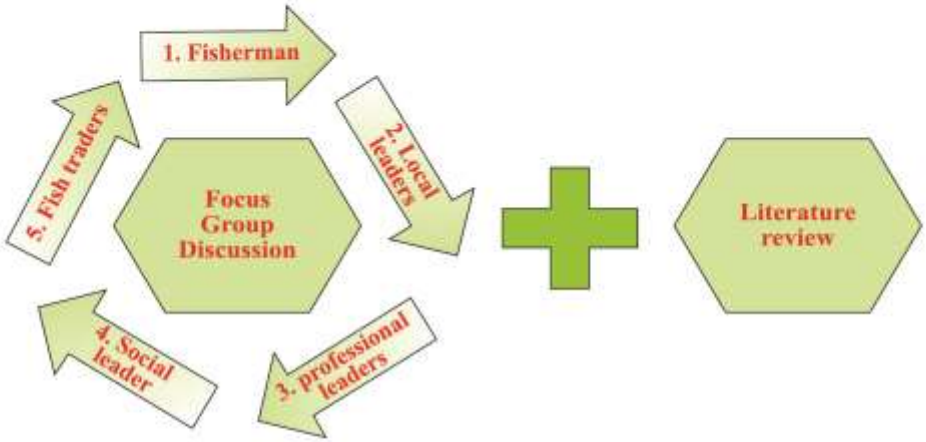


Figure 2. A chart for understanding of the species habitat, stock nature and abundance.

Sampling procedure and frequency

The estimation of parameters a and b was done by linear regression analyses, which follows equation such as ln(W) = ln(a) + bln(L). Additionally, 95% confidence interval was calculated for parameters a and b. We were also calculated the coefficient of determination (r^2). Regression analyses were performed to eliminate outliers (Froese, 2006).Statistical Product and Service Solution (SPSS) software was used to perform statistical analyses. The statistical difference from the isometric value (b = 3) for LWRs were determined by t-test. All statistical analyses were considered at 5% significance level (P < 0.05).

Fig. 1. Gears for sampling

Fig. 2. Data collecting

Generally, CPUE estimation is done by dividing annual fish landing amount by a total number of fishing trips in a year. Moreover, CPUE can also be calculated by considering fishing days and vessel numbers. CPUE is one of the important indices of species abundance (Chen and Chiu, 2009). However, it is not a firm indicator of stock abundance since it can be influenced by some factors (Harley et al., 2001). Usually, these factors affect fish harvest from the sea during fishing operation (Maunder et al., 2006).

Like other factors, vessel's capacity in gross registered tonnage (GRT) was found as a significant contributor to CPUE (Parente, 2004). In this study, a standard formula was opted to estimate the abundance of a species as; $C_t, P_t / T_t$ .....(2)

where C is the catch per unit effort (CPUE) for fish species (kg/day/effort.). Ct is the CPUE for the year t. Pt represents fish catch for a particular season t. Tt indicates number of days of fishing with a particular fishing craft in the same season t.

Incorporated ELEFAN-I (Electronic Length Frequency Analysis) in FiSAT-II program was assigned to estimate the value of asymptotic length and growth co-efficient (K) from following formula of the von Bertalanffy;

$Lt=Lx\ (1-e^{-k(t-t0)})$..... (3)

Where, t indicates the age of a fish species (yr), L is the mean total length at age t (cm), t0 is the hypothetical age when L is zero, K represents a growth coefficient (yr). From K and Lx the Growth performance index (ϕ) of species were calculated according to the formula of Pauly and Munro (1984);

$\phi = \text{Log } K+2 \text{ Log } L$..... (4)

This section was concluded with some finding such as length-weight relationship, abundance, asymptotic length, growth coefficient, and growth performance index of a stock.

This section was concluded with some finding such as length-weight relationship, abundance, asymptotic length, growth coefficient, and growth performance index of a stock.

Length-Weight Relationship

Spotted scat (*Scatophagus argus*)

The relationship between total length (TL) and body weight (BW) of *S. argus* has been displayed in Table 1. Logarithmic form of the equation ($BW = a \times TL^b$) was considered to establish TL-BW relationship. All values of total lengths were plotted against the values of respective body weights to complete the scatter diagram for getting a curvilinear relationship (Figure 3). Parabolic curves were made by plotting the calculated value of the body weight against the total length of the *S. argus*. In contrast, the values of log total TL against their log calculated BW were plotted to get a linear line.

Table 1. TL-BW association of sampled *Scatophagus argus* from the Sundarbans of Bangldesh.

Species	Size (N)	*a*	*b*	*r*	R^2	Allometry	p-Value
Scatophagus argus	865	0.0352	2.9792	0.91	0.98	Negetive	0.00

The estimated b value was calculated as 2.98. Here, *S. argus* showed nearly isometric growth (b = 3). The Pearson correlation co-efficient (r) value was estimated as 0.91 for *S. argus*. It indicates highly significant relationships ($p < 0.01$) between TL and BW of this species.

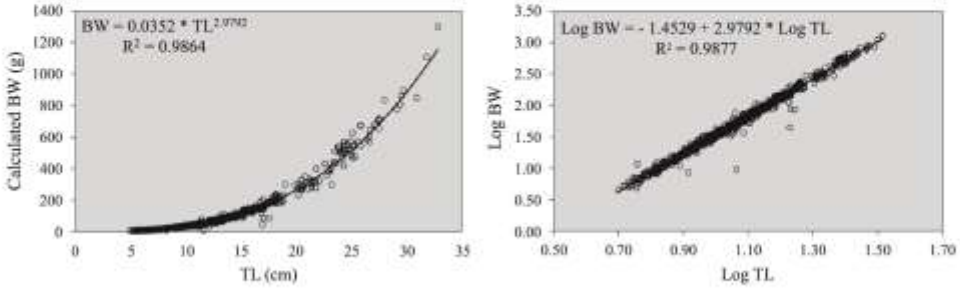


Figure 3. The relationship between Total lenth (TL) and body weight (BW) of *Scatophagus argus* in the Sundarbans mangrove forest of Bangladesh.

Tank goby (*Glossogobius giuris*)

In Table 2, the association between total length (TL) and body weight (BW) of *G. giuris* has been shown. A TW-BW relationship was established in a form of the equation, $BW = a \times TL^b$. All values of total lengths (TL) were plotted against the values of respective body weights (BW) to complete the scatter diagram for getting a curvilinear line (Figure 4). Parabolic curves were made by plotting the calculated value of the body weight against the total length of the *G. giuris*. In contrast, the values of log total TL against their log calculated BW were plotted to get linear lines.

Table 2. TL-BW association of sampled *Glossogobius giuris* from the Sundarbans of Bangldesh.

Species	Size (N)	*a*	*b*	*r*	R^2	Allometry	p-Value
Glossogobius giuris	1498	0.0092	2.97	0.92	0.95	Negative	0.00

The number of total sampled *G. giuris* was 1498. The estimated b value was calculated as 2.97. This species showed negative growth allometry. The Pearson correlation co-efficient (r) value was estimated as 0.92. It reveals highly positive and significant relationships ($p < 0.01$) between TL and BW of this species.

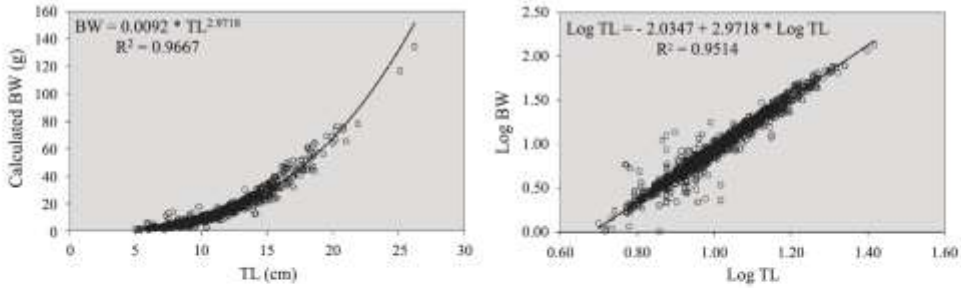


Figure 4. The relationship between Total lenth (TL) and body weight (BW) of *Glossogobius giuris* in the Sundarbans mangrove forest of Bangladesh.

Blood cockle (*Tegillarca granosa*)

The relationship between shell length (SL) and body weight (BW) of *T. granosa* has been displayed in Table 3. Logarithmic form of the equation ($BW = a \times SL^b$) was considered to establish SL-BW relationship. All values of shell length were plotted against the values of respective body weights to complete the scatter diagram for getting a curvilinear relationship (Figure 5). Parabolic curves were made by plotting the calculated value of the body weight against the shell length of the *T. granosa*. In contrast, the values of log total SL against their log calculated BW were plotted to get a linear line.

Table 3. SL-BW association of sampled *Tegillarca granosa* from the Sundarbans of Bangladesh.

Species	Size (N)	*a*	*b*	*r*	R^2	Allometry	p-Value
Tegillarca granosa	2695	1.1709	2.2818	0.91	0.88	Negative	0.00

The number of sampled *T. granosa* was 2695. The estimated b value was 2.2818 for the species. *T. granosa* showed Negative growth allometry. The Pearson correlation co-efficient (r) values were estimated as 0.91 for *T. granosa*. It showed a positive and highly significant relationships ($p < 0.01$) between SL and BW of this species.

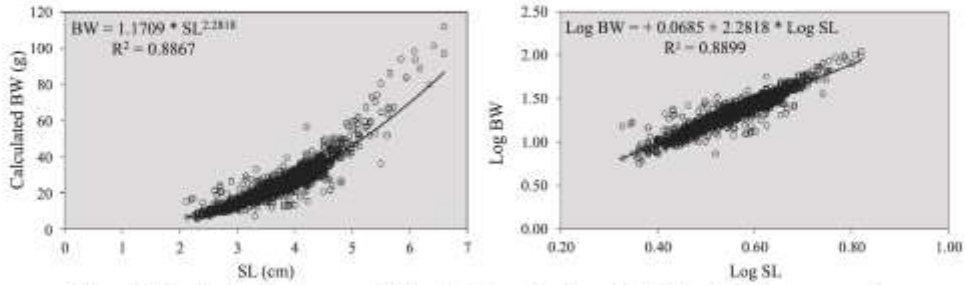


Figure 5. The relationship between shell length (SL) and body weight (BW) of *Tegillarca granosa* in the Sundarbans mangrove forest of Bangladesh.

Mud crab (*Scylla olivacea*)

The relationship between carapace width (CW) and body weight (BW) of *S. olivacea* has been displayed in Table 4. Logarithmic form of the equation ($BW = a \times CW^b$) was considered to establish CW-BW relationship. All values of carapace widths were plotted against the values of respective body weights to complete the scatter diagram for getting a curvilinear relationship (Figure 6). Parabolic curves were made by plotting the calculated value of the body weight against the carapace width of the *S. olivacea*. In contrast, the values of log total CW against their log calculated BW were plotted to get linear lines.

Table 4. CW-BW association of sampled *Scylla olivacea* from the Sundarbans of Bangldesh.

Species	Size (N)	*a*	*b*	*r*	R^2	Allometry	p-Value
Scylla olivacea	1316	0.2063	2.9462	0.90	0.94	Negative	0.00

The total number of the mud crab sample (N) was 1316. The estimated b values were 2.90 for *S. olivacea*. This shellfish showed negative growth allometry. The Pearson correlation co-efficient (r) values were estimated as 0.90 for both sexes of *S. olivacea*. It indicates highly significant relationships ($p < 0.01$) between CW and BW of this species.

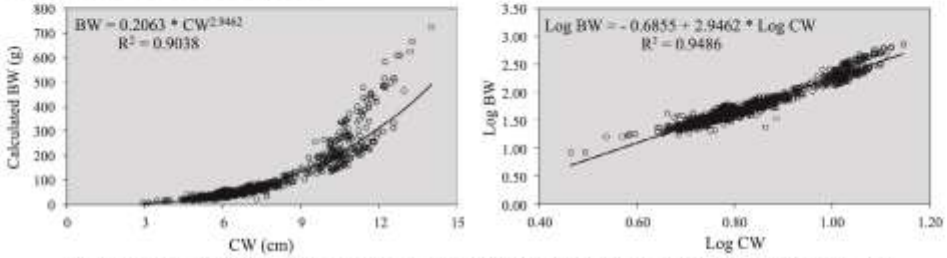


Figure 6. The relationship between carapase width (CW) and body weight (BW) of *Scylla olivacea* in the Sundarbans mangrove forest of Bangladesh.

Gray eel-catfish (*Plotosus canius*)

The relationship between total length (TL) and body weight (BW) of *P. canius* has been displayed in Table 5. Logarithmic form of the equation ($BW = a \times TL^b$) was considered to establish TL-BW relationship. All values of total lengths were plotted against the values of respective body weights to complete the scatter diagram for getting a curvilinear relationship (Figure 7). Parabolic curves were made by plotting the calculated value of the body weight against the total length of the *P. canius*. In contrast, the values of log total TL against their log calculated BW were plotted to get a linear line.

Table 5. TL-BW association of sampled *Plotosus canius* from the Sundarbans of Bangladesh.

Species	Size (N)	*a*	*b*	*r*	R^2	Allometry	p-Value
Plotosus canius	545	0.0052	3.02	0.92	0.98	Positive	0.00

The total number of the Grey eel-catfish sample (N) was 545. The estimated b value was calculated as 3.02. Therefore, *P. canius* showed positive growth allometry but close to isomerism (b = 3). The Pearson correlation co-efficient (r) values was estimated as 0.92 for *P. canius*. It indicates highly significant association ($p < 0.01$) between TL and BW of this species.

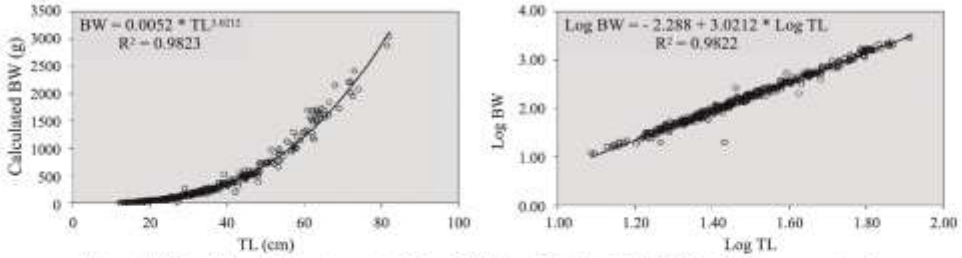


Figure 7. The relationship between total length (TL) and body weight (BW) of *Plotosus canius* in the Sundarbans mangrove forest of Bangladesh.

Species-wise abundance

From the fish group, abundance of spotted scat *Scatophagus argus* (Linnaeus, 1766), tank goby *Glossogobius giuris* (Hamilton, 1822) and grey-eel catfish *Plotosus canius* were 725 gm, 215 gm and 1.90 kg per person in one day. Similarly, abundance of mud crab *Scylla olivacea* and blood cockle *Tegillarca granosa* were 2.5 kg and 20 kg per person on daily basis (Table 6).

Table 6. Species-wise abundance in the Sundarbans.

Species	CPUE (weight/person/day)
Spotted scat (*Scatophagus argus*)	725 g/person/day
Tank goby (*Glossogobius giuris*)	215 g/person/day
Blood cockle (*Tegillarca granosa*)	20 kg/person/day
Mud crab (*Scylla olivacea*)	2.5 kg/person/day
Grey eel-catfish (*Plotosus canius*)	1.90 kg/person/day

Growth parameters

The von Bertalenffy asymptotic lengths were 34.65 cm, 28.35 cm, 7.35 cm, 14.70 cm and 86.10 cm and the K were 0.38/yr, 1.20/yr 0.66/yr 0.38/yr and 0.98/yr for spotted scat, tank goby, blood cockle, mud crab, and grey eel-catfish, respectively (Table 7). The estimated growth performance index (ϕ') of spotted scat, tank goby, blood cockle, mud crab, and grey eel-catfish were observed to be 2.65, 2.98, 3.55, 1.91 and 3.86.

Table 7. Growth parameters of selected species from the Sundarbans

Species	L∝	K	Φ
Spotted scat (*Scatophagus argus*)	34.65	0.38	2.65
Tank goby (*Glossogobius giuris*)	28.35	1.20	2.98
Blood cockle (*Tegillarca granosa*)	7.35	0.66	3.55
Mud crab (*Scylla olivacea*)	14.70	0.38	1.91
Grey eel-catfish (*Plotosus canius*)	86.10	0.98	3.86

Study 2. Estimation of mortality rates and exploitable level of the selected species

Mortality is a key component to understanding the population dynamics of fish species. Total mortality is often estimated from the sequential decline observed in cohorts of fish. length converted catch curve method of Beverton and Holt (1956) was applied to determine total mortality (Z). The formula of the total mortality as follows:

$Z = F/M$ ..... (5)

Where, Z indicates total mortality of the stock, F is the fishing mortality and M is the natural mortality. Natural mortality is the removal of fish from the stock due to causes not associated with fishing. Such causes can include disease, competition, cannibalism, old age, predation, pollution or any other natural factor that causes the death of fish. In fisheries model's natural mortality is denoted by (M). Natural mortality (M) was estimated according to Pauly (1980) as follows in the formula 6;

$\log10M = -0.0066 - 0.279 \log10L\infty + 0.6543 \log10K + 0.4634 \log10T$ …… (6)

where, M indicates natural mortality of the stock, $L\infty$ is the asymptotic length of a species, K is the growth co-efficient and T is the habitat temperature. However, fishing mortality rate is the proportion of a fish stock removed by fishing (as opposed to predation or other causes of death). By following formula, we were estimated the fishing mortality;

Fishing mortality (F) = Z − M ……..(7)

Applied on a fish stock, it is the proportion of the numbers or biomass removed by fishing. A 10% exploitation rate means that 10% of the available stock is being harvested within the time frame considered (per year, per month, etc.). As a measure of fishing pressure, it is proportional to fishing mortality;

Exploitation rate (E) = F/Z ..…… ……..(8)

Mortality and exploitation

The total mortality (Z) of spotted scat, tank goby, blood cockle, mud crab, and grey eel-catfish were estimated as 2.08/yr, 6.19/yr, 4.38/yr, 2.94/yr, and 3.75/yr, respectively, by using length converted catch curve analysis (Table 8). Fishing mortalities (F) were 1.15/yr, 4.09/yr, 2.31/yr, 1.75/yr, and 2.40 for spotted scat, tank goby, blood cockle, mud crab, and grey eel-catfish, respectively. In contrast, natural mortalities (M) of spotted scat, tank goby, blood cockle, mud crab, and grey eel-catfish were calculated as 0.94/yr, 2.10/yr, 2.07/yr, 1.19/yr, and 1.34/yr, respectively. Thus, expoitation rate (E) of spotted scat, tank goby, blood cockle, mud crab, and grey eel-catfish were computed as 0.55, 0.66, 0.53, 0.60, and 0.64, respectively (Table 8). The maximum permissible limit of exploitation (Emax) value is 0.50 for all selected species. Generally, all E values of selected species were exceeded the maximum permissible limit of expoitation (Emax).

Table 8. Mortalities and explitations of selected species from the Sundarbans.

Species	Z	M	F	E
Spotted scat (*Scatophagus argus*)	2.08	0.94	1.15	0.55
Tank goby (*Glossogobius giuris*)	6.19	2.10	4.09	0.66
Blood cockle (*Tegill arca granosa*)	4.38	2.07	2.31	0.53
Mud crab (*Scylla olivacea*)	2.94	1.19	1.75	0.60
Grey eel-catfish (*Plotosus canius*)	3.75	1.34	2.40	0.64

Study 3. Identifying vulnerable size groups of a fish species

Probability of capture

Probability of capture calculated from the length-converted catch curve routine were used to estimate the final values of L25, L50 and L75 i.e., lengths at which 25%, 50% and 75% of the fish would be vulnerable to the different gears such as different nets, long lines and traps for a specific species (Pauly, 1984).

Virtual population analysis

Virtual population analysis (VPA) is a cohort modeling technique commonly used in fisheries science for reconstructing historical fish numbers at age using information on death of individuals each year. This death is usually partitioned into catch by fisheries and natural mortality. VPA is virtual in the sense that the population size is not observed or measured directly but is inferred or back-calculated to have been a certain size in the past in order to support the observed fish catches and an assumed death rate owing to non-fishery related causes.

Virtual population analysis was introduced in fish stock assessment by Gulland in 1965 based on older work. The technique of cohort reconstruction in fish populations has been attributed to several different workers including Professor Baranov from Russia in 1918 for his development of the continuous catch equation, Professor Fry from Canada in 1949 and Drs. Beverton and Holt from the UK in 1957. Because cohort reconstruction is essentially an accounting exercise it was likely independently conceived many times. The virtual population analysis (VPA) was employed to estimate the extent of mortality on various size classes of a species. The fishing pressure on a particular sized fish species was indicated against the number of anticipated populations (Figure 8).

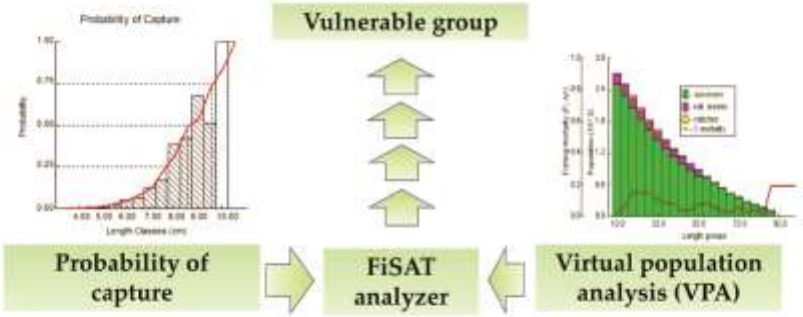


Figure 8. Strategies to calculate vulnerable size group of a species.

Vulnerable size groups

Probability of capture is one of the very useful drivers in stock assessment of fisheries science. It shows the vulnerability of different sizes of fin fish and shellfish to different grars in a given location at a given time. The probabilities of capture analysis for spotted scat (*S. argus*) found that 25% of 8.16 cm TL, 50% of

10.28 cm TL and 75% of 12.31 cm TL were vulnerable to the gears. Therefore, it can be assumed that more than half of the harvested *S. argus* remained between the total length of 8.16 cm and 10.28 cm. In addition, VPA results reveals that a maximum fishing pressure on *S. argus* population was found between total length of 29.01 cm and 33.0 cm.

Similarly, probabilities of capture analysis for *G. giuris* depicted that 25% of 6.30 cm TL, 50% of 7.80 cm TL and 75% of 9.34 cm TL were vulnerable to the gears. Therefore, it can be assumed that more than half of the harvested crabs remained between the carapace length of 6.30 cm and 7.80 cm. In addition, VPA revealed that a maximum number of *G. giuris* were caught between totla length of 7.01 cm and 27.0 cm. Similarly, the high values of F for the species occurred within some length groups, ranging from 7.01 cm to 27 cm, with vales of F between 0.65/yr and 4.84/yr.

Again, the probabilities of capture analysis for blood cockle (*T. granosa*) showed that 25% of 3.21 cm SL, 50% of 3.59 cm SL and 75% of 03.99 cm SL were vulnerable to the gears. Therefore, it can be assumed that more than half of the harvested Blood cockle remained between the shell length of 3.21 cm and 3.59 cm. In addition, VPA results indicates that a maximum number of Blood cockles were heavily expoited as a first group having shell length from 3.51 cm to 4.50 and as a second group having shell length from 6.01 cm and 7.0 cm.

Further, an analysis on the probabilities of capture for mud crab (*S. olivacea*) revealed that 25% of 4.69 cm CW, 50% of 5.45 cm CW and 75% of 6.21 cm CW were vulnerable to the gears. Thus, it can be assumed that more than half of the harvested crabs remained between the carapace length of 4.69 cm and 5.45 cm. Moreover, VPA results indicates that a maximum number of crabs were caught between the carapace length of 12.01 cm and 14.00 cm.

Furthermore, the probabilities of capture analysis for grey eel-catfish (*P. canius*) showed that 25% of 17.02 cm TL, 50% of 20.77 cm TL and 75% of 24.88 cm TL were vulnerable to the gears. Therefore, it indicates that more than half of the harvested *P. canius* remained between the total length of 17.02 cm and 20.77 cm. In addition, VPA results indicates that a maximum number of *P. canius* was caught between total length of 67.01 cm and 82.0 cm. Likewise, the high peaks of F for *P. canius* occurred within some length groups, ranging from 27.01 cm to 37 cm, and 62.01 cm to 67 with vales of F between 1.12/yr and 2.86/yr.

Discussion and conclusion

The Sundarbans mangrove forest is one of the significant features of the coastline of Bangladesh, which is recognized as the world largest mangrove forest. The forests have a great role in the country's national economy, and provides livelihood for the local people through fishing, tourism, wood and non-wood products. Artisanal fisheries are mainly predominant in the mangrove and adjacent tributaries which comprises various kinds of traditional fishing gears and crafts. These fisheries activities are most often occurred in the coastal areas to catch fish. Many fish and crustaceans are highly dependent on mangroves for completing their life cycle, and thus mangroves serve as a higher fisheries biodiversity.

Despite having several laws, policies and management plans, the forest is now showing clear signs of degradation. Recently, many new fish species were found around the Sundarbans mangrove areas.

However, stock status of these identified fish species is totally unknown to the fisheries stakeholders. Therefore, fisheries stock assessment is an important tool to estimate the resource for a better plan to use them in a sustainable manner. In Bangladesh, fisheries business is a heritage in the southern part of the country for daily livelihood. As a result, people fished directly from nature in order to meet demand for different products for local and international markets. Thus, there is a possibility of collapsing various fish population fishery soon. Thus, fish stock assessment was carried out to address the issue of determining the status of the populations in the mangrove regions.

To estimate population parameters of five species (spotted scat, tank goby, blood cockle, mud crab, and grey eel-catfish), length-weight and length-frequency data was collected from various areas of the Sundarban mangrove forest to evaluate growth parameters, mortality rates and exploitation levels. We have managed to collect seven months (Dec/2021 - June/2022) data for analyzing and estimating all parameters. In terms of growth, all species showed nearly isometric growth except blood cockle (negative allometric). There are some specific groups were identified for each species as vulnerable groups. In addition, the populations of five selected species were summarized as overexploited in the Sundarbans of Bangladesh. However, total twelve months data will explain more and estimate all population parameters accurately and might change the present assumptions on these stocks.

Potentiality of aquatic weed as alternative feed ingredients for the development of cost-effective fish feed for coastal aquaculture

Researchers
Debashis Kumar Mondal, SSO
Md. Masudur Rahman, SO
Shahanaj Parvin, SO
Md. Abu Naser, SO

Objectives

- To investigate the status of available aquatic weed in South-west region and make inventory based on morphometry and DNA barcode analysis
- To observe the nutritional status (proximate composition, macro and micro elements) of important aquatic weed
- To examine the potentials of explored weed as dietary ingredients in fish feed

Achievement

Experiment-1. Identification of potential aquatic weed species using morphometric characteristics and DNA barcodes

The study was conducted in the Bangladesh Fisheries Research Institute, Brackishwater Station, Paikgacha, Khulna in collaboration with 'Invent technology', Dhaka according to the following experimental procedures.

Collection of samples from South-western coastal area

Sample was collected according to the methodology of the project proposal and many others respective literature review. At first sample was collected from nature. Then they were softly washed by water of the respective places for removing mud, dirt and unnecessary substances. Sample was collected with very carefully for avoiding contamination of other species. During sample collection, necessary photographs were taken by DSLR camera.

On spot packing

Every zipper bag was marked by permanent marker according to sample number recorded in data sheet. Then preliminary washed samples were inserted into zipper bag.

Travel preservation

For travelling samples were kept in insulated ice box with ice. Ice box was also kept in cool area.

Data recording

With various devices data were taken while sample were collected. Dissolved oxygen and pH were measured by DO meter and Hanna pH meter, respectively. Salinity was taken by refractometer. Total alkalinity was measured by titrimetric method. GPS location was taken by android apps 'My GPS Location'. Ecological information was recorded in field note book. At the same time photograph of the ecosystem was also taken.

Weed processing

After reaching at station samples were taken out from insulated box carefully. Then the collected weeds were washed carefully with tap water as morphological characteristics remain same as they were taken from nature. After washing samples were kept in water absorbing foam for removing excessive water. After 30 minutes morphological characteristics such as color, root, branch, cell type, microscopically view of cell, leaf number, appearance etc. were carefully observed and the then noted down. At the same time photo was also taken by camera. Then the samples were kept overnight for removing remains water with a blotting paper.

Weed drying

After 8-10 hours weed was dried under sun in weed dryer made by BS technology. After 1-2 days when samples were totally dried they are taken out from dryer and preserved at zipper bag. Zipper bag was marked according to data sheet number before keeping sample.

Weed preservation

After drying the samples was tagged and kept in a plastic box and shaded area for further study and subsequent molecular work in "Invent technology", Dhaka. Regular observation was taken till they are sent to Lab for proximate analysis and barcoding.

Morphological identification

Each sample was identified morphologically by assessing morphometric characteristics. Different taxonomic book, standard monographs of Boergesen, (1913); Islam, (1976); Sen and Naskar (2003) and Encyclopeia of Flora and Fauna of Bangladesh will be used for morphological study.

DNA extraction, PCR amplification and sequencing

The process of identification of aquatic weed using DNA bar code is still on-going. DNA will be extracted from collected samples using commercial kits for good success as high-throughput work. The target rbcL gene region was amplified using universal plant DNA barcoding primers (CBOL, 2009), rbcLa-F. 5'-ATGTCACCACAAACAGAGACTAAAGC-3' and rbcLa-R. 5'- GTAAAATCAAGTCCACCRCG-3'. 16S rRNA also can be used for identification of the organisms if necessary. PCR will be performed using a reaction mixture of a total volume of 25µl; 12.5µl of Taq PCR Master Mix (Invitrogen, India), 11µl distilled water, 0.5µl forward primer (10 µM), 0.5µl reverse primer (10 µM), and 0.5µl of the DNA template (50–80 ng/µl). The PCR conditions were as follows. 1 cycle (94 °C for 3 min), 35 cycles (94 °C for 1 min, 55 °C for 1 min, and 72 °C for 1 min), and 1 cycle 72 °C for 7 min. The same negative control used during DNA extraction acted as negative control for PCR. PCR products of rbcL gene fragment will be run and visualized by 1.5% agarose gel electrophoresis (using above protocol described in 5.2.2.) for 600bps amplicon. The purified PCR products will be sequenced by sanger sequencing method in both directions. Each unique sequence will be served as a blast query to the GenBank database to identify the most similar sequence in GenBank to the queried sequence. DNA sequencing will be performed from commercial sequencing company, Macrogen Inc. (Seoul, South Korea). Received sequences will be analysed in Invent Technology, Dhaka.

Molecular identification, genetic diversity and DNA taxonomy using bioinformatics tools

Several computer software and modern bioinformatics program such as Ultra Edit, BioEdit, Clustal X, Interactive Tree Of Life (iTOL), Molecular Evolutionary Genetic Analysis (MEGA) will be used for sequence editing, phylogenetic and population genetic study, and phylogenetic analysis using DNA sequence data by high speed computer. NCBI BLAST search and BOLD database will be used for molecular identification of the weed. The sequences will be aligned in Clustal X ver. 2.0.6 (Thompson et al., 1997) and (MEGA) ver. X will be used for phylogenetic and pair-wise distance analysis (Kumar et al., 2018). The pair-wise distance was calculated as per Kimura-2 parametric distance model (Kimura, 1980). NJ tree was redrawn using Interactive Tree Of Life (iTOL) (Letunic and Bork, 2019) for better representation of tree based identification. Kimura-2 parameter distance model (Kimura, 1980) was to calculate the distances between the sequences. All three codon positions and non-codon positions were included and all the alignment positions containing gaps and missing data was eliminated from the analysis. MEGA X was also used to conduct nucleotide diversity and Tajima's neutrality (Tajima, 1989; Nei and Kumar, 2000) tests.

A total of 28 specimen of aquatic weed was collected of which 19 was green weeds, 6 was red weeds and 3 was brown weeds. Among 28 samples 9 samples were identified morphologically with the help of different taxonomic books and internet sources (Table 1).

Table1. Group-wise abundance of collected weeds.

Group of weed	Number of collected weed	Morphologically identified weed
Green weed	19	6
Red weed	6	1
Brown weed	3	2
Total	28	9

Among total collected 28 samples, 68% was green weed, 21% was red weed and 11% was brown weed (Figure-1).

Figure 1. Percentage of different groups of collected aquatic weeds.

List of identified samples

Samples were preliminary identified according to morphometric characteristics and photo combination from 'Sea Weed of Bangladesh Coast" book, journals, and also internet (Table 2).

Table 2. Morphologically identified weeds.

Group name of weed	Scientific Name
Red weed	*Kappaphysis alvarezii*
Green weed	*Najas graminea* *Cladophora laetivirens* *Ulva lactuca* *Ulva compressa* *Enteromorpha intestinalis* *Ulva conglobate*
Brown weed	*Colpomenia perigrina*
	Colpomenia sinuosa

Table 3. Details of the collected aquatic weeds.

Photo	Name	Characteristics	Water quality parameters	GPS
	Not identified	Grow in muddy region, rooted , attached with subtitle area, generally light green color	pH. 8.46 DO. 5 Salinity.9 Depth.6 FT	La- 22.27 Lo- 89.20 Al- +6m
	Not identified	Attached with tree root, looks like spider net, deep green color, unicellular, length 5-6 cm	pH. 8.46 DO. 5.1 Salinity. 9 Depth. 6 FT	La- 22.27 Lo- 89.20 Al- +6m
	Common Name. Elkhorn sea moss Scientific Name. *Kappaphysis alvarezii*	Red color, attached with tree root of sundarbans, grow in colonial form, branchial, length 2-3 cm	pH. 8.46 DO. 5.2 Salinity. 9 Depth. 6 FT	La- 22.27 Lo- 89.20 Al- +6m
	Not identified	Attached with tree root, looks like spider net, deep green color, branchial, length 5-6 cm	pH. 8.46 DO. 5.2 Salinity. 9 Depth. 1.5 FT	La- 22.27 Lo- 89.20 Al- +6m
	Not identified	Red color to brownish, attached with tree root of Sundarbans, grow in colonial form, branchial, length 2-3 cm	pH. 8.36 DO. 3.6 Salinity. 13 Depth. 3 FT	La- 22.24 Lo- 89.24 Al- -3m

	Scientific Name. *Ulva conglobata*	Grow in muddy region, , attached with subtidal area, generally deep green color, branchial and rooted, 2-6 cm length	pH. 8.31 DO. 3.6 Salinity. 10 Depth.1.5 FT	La- 22.24 Lo- 89.23 Al- -6m
	Not identified	Attached with tree , deep green color, branchial, length 5-6 cm	pH. 8.31 DO. 3.7 Salinity. 10 Depth. 1.5 FT	La- 22.24 Lo- 89.23 Al- -20m
	Not identified	Red color, attached with tree root of Sundarbans, grow in colonial form, branchial, length 2-3 cm	pH. 8.45 DO. 10 Salinity.10 Depth. 8 FT	La- 22.25 Lo- 89.23 Al- -18m
	Not identified	Attached with river side muddy region, spread in mud area, grow colonial, deep green color, branchial	pH. 8.71 DO. 3.3 Salinity. 4 Depth. 5 FT	La- 22.59 Lo- 89.32 Al- +9m
	Not identified	Attached with kewra tree, filamentous, generally deep green in moisture area, branchial, grow up to 5-10 cm	pH. 8.72 DO. 3.7 Salinity. 2 Depth. 3 FT	La- 22.58 Lo- 89.38 Al- -0m
	Not identified	Hairy like green tubular shaped. Hardy texture, attached with tree root	pH. 8.81 DO. 3.9 Salinity. 1 Depth. 7 FT	La- 22.58 Lo- 89.40 Al- +5m

	Not identified	Bright green, attached in lower root branch of tree. greenish color, grow up to 5-10 cm, branchial, colonial form	pH. 8.8 DO. 3.9 Salinity.1 Depth. 7 FT	La- 22.58 Lo- 89.40 Al- +5m
	Not identified	Found in upper surface area. bright green, grow up to 5-10 cm, branchial, colonial form	pH. 8.8 DO. 3.9 Salinity.1 Depth. 7 FT	La- 22.58 Lo- 89.40 Al- +5m
	Not identified	Attached with gewa root near surface land, Light green, grow up to 5-10 cm, branchial, colonial form	pH. 8.8 DO. 4 Salinity.1 Depth. 8 FT	La- 22.58 Lo- 89.40 Al- +6m
	Local Name. Kata shewla Scientific Name. *Najas graminea*	Generally grow in shallow coastal area specially in gher. maximum growth 5-6 ft, greenish color, branchial, rooted	pH. 9.5 DO. 4.7 Salinity. 5 Depth. 1.5 FT	La- 22.58 Lo- 89.36 Al- 5m
	Local Name. Suti shewla Scientific Name. *Cladophora laetivirens*	Attached with substrate in water surface, filamentous, generally deep green in moisture area, branchial, grow up to 3-4 cm	pH. 8.8 DO. 4.8 Salinity. 3 Depth. 4 FT	La- 22.596 Lo- 89.304 Al- 16m
	Not identified	Attached with muddy area, submerged, branchial, rooted, leafs look like flower, grow up to 10-12 ft	pH. 8.8 DO. 4.8 Salinity.3 Depth. 4 FT	La- 22.596 Lo- 89.304 Al- 16m

	Not identified	Attached with rock block, submerged in water, reddish color, branchial	pH. 8.8 DO. 4.8 Salinity.3 Depth. 4 FT	La- 22.596 Lo- 89.304 Al- -16m
	Scientific Name. *Ulva lactuca*	Grow in muddy region, , attached with subtitle area, generally deep green color, branchial and rooted, 2-6 cm length	pH. 8.45 DO. 5 Salinity.10 Depth. 3 FT	La- 22.249 Lo- 89.222 Al- -9m
	Scientific Name. *Ulva compressa*	Grow in muddy region, , attached with subtitle area, generally deep green color, branchial and rooted, filamentous ,spreaded in moisture area specially near river bank, 2-6 cm length	pH. 8.45 DO. 5 Salinity.10 Depth. 3 FT	La- 22.249 Lo- 89.222 Al- -9m
	Common Name. Stone hair Scientific Name. *Enteromorpha intestinalis*	Attached with floating substances, deep greenish color, hairy like, unicellular and tubular form, grow up to 10-12 cm	pH. 8.45 DO. 5 Salinity. 10 Depth. 24 FT	La- 22.253 Lo- 89.226 Al- -5m
	Not identified	Attached with floating substances, brownish color, branchial, grow up to 2-3 cm	pH. 8.41 DO. 6 Salinity.10 Depth. 24 FT	La- 22.25 Lo- 89.226 Al- -5m
	Not identified	Attached with floating substances, look like root, brown color, grow 2-3 cm	pH. 8.41 DO. 6 Salinity.10 Depth. 24 FT	La- 22.25 Lo- 89.226 Al- -5m

	Common Name. Sea potato Scientific Name. *Colpomenia perigrina*	Slight submerge in water, brown color, attached with muddy area, colonial, ear like structure, available during collection	pH. 8.3 DO. 4.2 Salinity.15 Depth. 3 FT	La- 22.28 Lo- 89.33 Al- +4m
	Not identified	Attached with floating substances, brownish color, branchial, grow up to 2-3 cm	pH. 8.58 DO. 6.5 Salinity.7.5 Depth.1 FT	La- 22.36 Lo- 89.61 Al-+4m
	Common Name. Puffy Brown seaweed Scientific Name. *Colpomenia sinuosa*	Grow under root area, upper tide near zone, shaded area, yellowish color	pH. 8.58 DO. 6.5 Salinity. 7.5 Depth.1 FT	La- 22.36 Lo- 89.61 Al- +4m
	Not identified	Red color, attached with tree root of Sundarbans, grow in colonial form, branchial, length 2-3 cm	pH. 7.92 DO. 8 Salinity.15 Depth. 3 FT	La- 22.43 Lo- 89.58
	Not identified	Attached with submerged gher, filamentous, generally deep green in moisture area, branchial, Grow up to 3-4 cm	pH. 7.92 DO. 8 Salinity.15 Depth. 4 FT	La- 22.43 Lo- 89.59

DNA extraction, PCR amplification and sequencing. Samples has been sent to lab for barcode analysis and the result will be available shortly.

Discussion and conclusion

A total of 28 specimen of aquatic weed was collected from the adjacent river and canal of Sundarban mangrove forest and from coastal ghers of which 19 was green weeds, 6 was red weeds and 3 was brown weeds. Among 28 samples 9 samples were identified morphologically with the help of different taxonomic books and internet sources. Among the nine morphologically identified weeds, only one species (*Kappaphysis alvarezii*) was red weed, six species (*Najas graminea*, *Cladophora laetivirens*, *Ulva lactuca*, *Ulva compressa*, *Enteromorpha intestinalis and Ulva conglobate*) were green weed and two species were brown weed (Colpomenia perigrina, Colpomenia sinuosa). Identification of all coastal aquatic weeds will be done shortly after bar code analysis. Water quality parameters of the sampling sites were within the suitable range (pH. 8.3~9.5, DO. 3.6~10 mg/l, Salinity. 1~15 ppt).

Domestication, reproductive biology, breeding and culture of indigenous brackishwater prawns under captive conditions

Researchers

Shawon Ahmmed, SO and PI
Md. Hashmi Sakib, SSO
Md. Reaz Morshed Ranju, SO
Rafia Afrin, SO

Objectives

The goal or broad objective of the project is to develop captive breeding and seed production technique of indigenous brackishwater prawns (*Macrobrachium villosimanus*, *M. lamarrei* and *M. dayanum*). In achieving the goal, experiments will be conducted with the following specific objectives:

- Identification of target prawn using taxonomic and barcoding approach
- To domesticate brackishwater prawns under captive condition for broodstock development
- To investigate the reproductive biology (fecundity, GSI, breeding time, embryonic development, etc.) of the prawns
- To develop breeding and larvae rearing protocol of the prawns

Achievement

Experiment/study 1. Identification of target prawn using taxonomic and barcoding approach

This experiment was performed at Brackishwater Station's Disease and Microbiology Laboratory. The targeted brackishwater prawn samples were collected from the natural sources of south-west coastal region. Taxonomic study was performed qualitatively and quantitatively (Figure 1). Though morphometric and meristic study were performed but it is difficult to accurately identify the targeted prawns. Therefore, for accurate identification of the targeted prawns up to the species level, the DNA extraction were performed by DNeasy Blood and Tissue Kit (Qiagen, Hilden, Germany) (Flowchart 1).

Then, the DNA amplification was performed by mtCOI PCR marker. After that, the PCR product purification was purified using QIAquickR Gel extraction kit. Sequencing was generated by bidirectional sequencing using Sanger sequencing method. Finally, the species identification process was completed by checking the generated sequences using sequence analysis software (ABI) and assured by online BLAST search program.

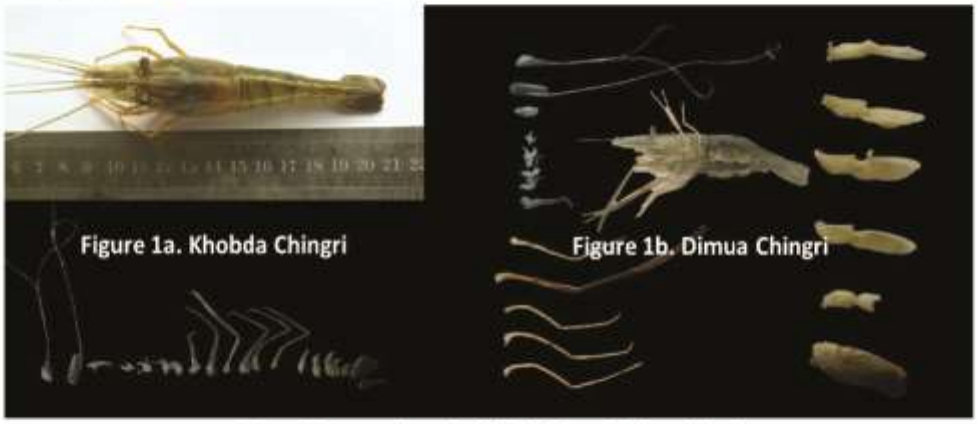


Figure 1. Taxonomic study of the Khobda and Dimua chingri.

The DNA quantification was performed with Nano Drop Spectrophotometer (Nano Drop 2000 UV-Vis Spectrophotometer, Thermo Fisher Inc., USA). The DNA concentration was shown in Table 1.

Table 1. Quantification of extracted DNA using NanoDrop™ spectrophotometer.

Sample Code	A260/280	A260/230	DNA Concentration (ng/µl)
BS@BFRI-1	1.94	1.61	183.2
BS@BFRI-2	1.97	1.99	58.9
BS@BFRI-3	1.85	0.32	205.3

Amplification of DNA samples were performed by Polymerase Chain Reaction (PCR) for further analysis. The following primer sequences were used in PCR showed in Table 2. The reaction mixture for PCR was set up by mixing the explicit volume of the components in an applicable sized given in Table 3. About 10 µl master mix containing buffers, dNTPS, $MgCl_2$, Taq polymerase (Promega, USA) without template DNA was prepared and allocated into PCR tubes. The specific template DNA was included in a marked PCR tube. The PCR tube containing master mix and template DNA was copped appropriately pursued by spinning to blend the mixure mildly and gather all the components to the base of the tube individually. The complete blend was then recollected, fixed and put in a thermal cycler and the cycling was begun quickly. PCR enhancement was done in an oil-free thermal cycler (Gene Atlas, G2, Astec, Japan).

Table 2. Primer sequences used in PCR.

Primer		5`-3` Sequence	Expected amplicon
mtCOI	LCOI490	GGTCAACAAATCATAAAGATATTGG	650 bp
	R-HCO2198	TAAACTTCAGGGTGACCAAAAAATCA	

Table 3. Components and reaction volume for the identification of prawns.

Reaction components	Volume	Total volume
Master mix	10 µl	10×5 = 50 µl
Primer Forward	1 µl	1×5 = 5 µl
Pri mer Reverse	1 µl	1×5= 5 µl
Template DNA	1 µl	1×5= 5 µl
Nuclease free water	7 µl	7×5 = 35 µl
Total Volume	20 µl	100 µl

For ensuring the denaturation of all the DNA templates, the PCR tubes containing mixtures of reaction volume were preheated at 95°C for 5 minutes in the thermal cycle. Then the PCR reaction continued according to the following Table 4. The successful amplification of the desired genes was visualized by resolving the PCR products in 1 % agarose gel depending on the size of amplicon. The process of gel electrophoresis was described in Flow chart 2.

Table 4. PCR condition for DNA barcoding.

Number of cycle	Step	Temperature	Time
1	Pre Heat	95°C	5 min
32 cycles	Denaturation	95°C	30 sec
	Annealing	48°C	30 sec
	Extension	72°C	1 min 30 sec
1	Final extension	72°C	5 min
1	Hold	4°C	Overnight

Gel electrophoresis showed positive amplification for all three samples (BS@BFRI-1, BS@BFRI-2 and BS@BFRI-3 (Figure 2). The image also makes it clear that all the samples (BS@BFRI-1, BS@BFRI-2 and BS@BFRI-3) showed positive band in gel electrophoresis. Moreover, the PCR product was purified and sequencing was done.

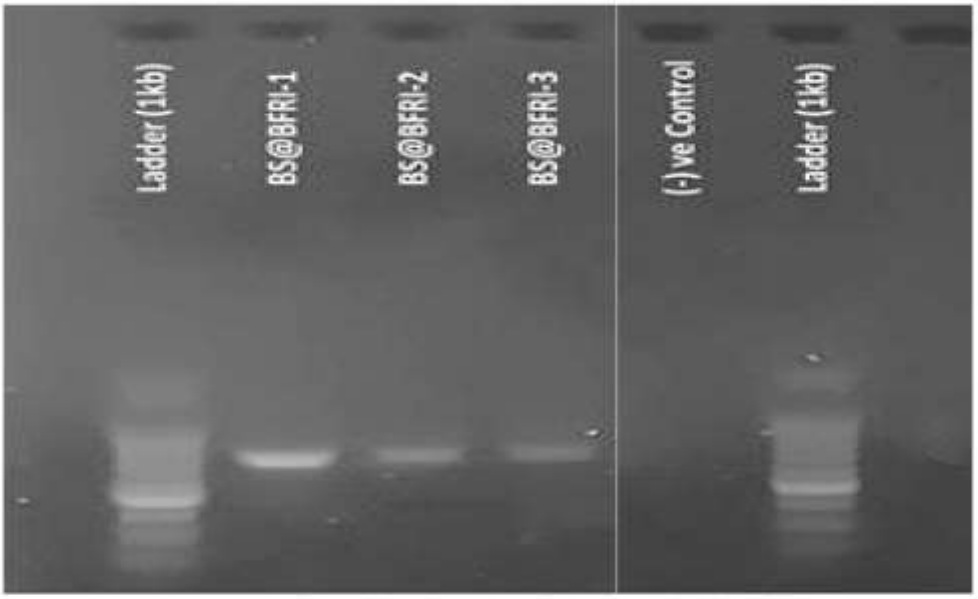


Figure 2. PCR Product profiles of three respective prawn samples with 1 kb ladder.

After sequencing of the prawn samples, the sequence chromatogram were checked (Figure 3) and aligned. Thereafter, the samples were accurately identified up to the species level through NCBI Blast search tool. The identified species mentioned in Table 5.

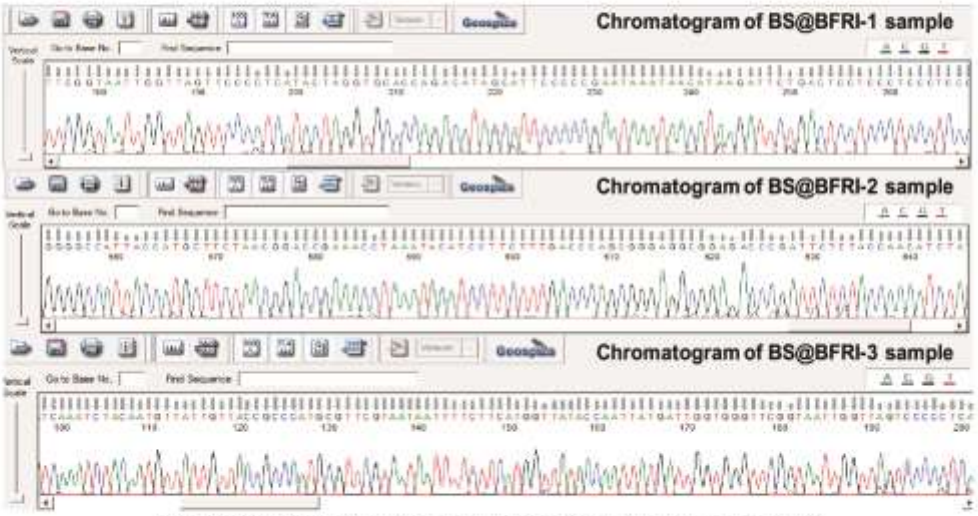


Figure 3. Sequence chromatogram of three respective prawn samples in FinchTV.

Table 5. Identification of the respective prawn samples with NCBI Blast tool.

Sample	Matched with	Max Score	Total Score	Query Cover	E value	Percentage Identity
BS@BFRI-1	*Macrobrachium villosimanus*	1029	1029	100%	0.0	99.64%
BS@BFRI-2	*Macrobrachium lamarrei*	1208	1208	100%	0.0	99.70%
BS@BFRI-3	*Macrobrachium dayanum*	1164	1164	100%	0.0	98.48%

Experiment/study 2. Domestication and culture of wild brackishwater prawns under controlled condition for brood development

This experiment was performed at Brackishwater station earthen pond and hatchery complex. The size of pond was 0.1 ha each and cistern size was 7 m^2 each. Fry and sub-adult size of 0.1 g and 10 g, respectively was collected from the wild sources and stocked at the rate of 5 individuals/m^2 for domestication in brackishwater pond conditions and habituated with commercial feeds. About 30-40% crude protein containing feed was fed at the rate of 10-5 % BW twice daily. The growth was monitored at fortnight interval. Water quality variables *viz*., temperature, salinity, pH, dissolved oxygen and ammonia were monitored in weekly basis as according Standard methods (APHA, 1998). Ranges of water quality variables of the domestication ponds were recorded as, temperature 18-30 °C, salinity 2-14 ppt, pH 7.64-8.47, dissolved oxygen 5.00-8.26 ppm and ammonia 0.00-0.25 ppm. Till reporting the domesticated prawn attained the average total length of 9.88±1.38 cm and average body weight of 13.16±4.23 g.

Study 3. Investigation of food and feeding habits brackishwater prawns

This experiment was performed at Brackishwater station nutrition laboratory. Prawn of different size, maturity and sex was collected from the natural sources. Then the stomach content was measured qualitatively and quantitatively. The food fractions were measured by the counting method. Finally, the food item was counted under microscope on a Sedgwick Rafter chamber. Food items observed in the stomach of the prawn samples has been depicted in Figure 3 and divided into six categories. aquatic plant fragments (6%), algae (10%), animal debris (35%), diatoms (7%), unidentified debris (38%), and sand grains (4%).

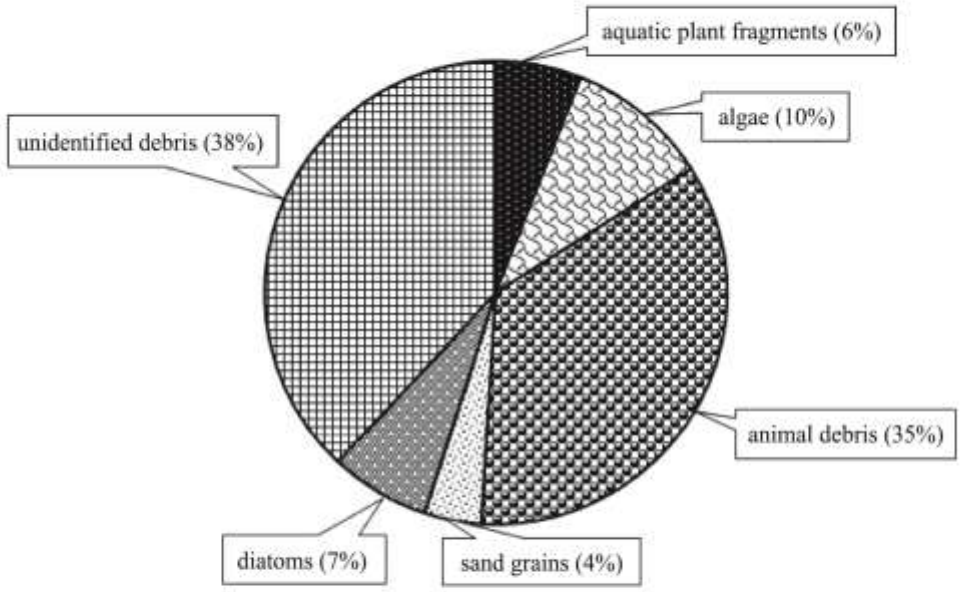


Figure 3. Relative importance of major groups of food items in targeted prawns.

Experiment/study 4. Determination of maturity and spawning season of brackishwater prawn

The study was performed at Brackishwater station disease and microbiology laboratory. Three species of targeted prawn samples were collected from local market and nearby ghers. The Length (cm) was measured with a measuring scale, weight was estimated by electric balance and sex was determined by observing the secondary sexual characteristics. Five male and five female prawns of different sizes were dissected at every 15 days interval. Then, the GSI and fecundity was estimated with the help of the following equations:

$$\text{Gonadosomatic Index (GSI). (Weight of gonad) / (Weight of Prawn/Shrimp)} \times 100$$

$$\text{Fecundity} = \text{NOV} / \text{WSs} \times \text{WOV}$$

Where, WOV = weight of the ovary; WSs= weight of the sub-samples; NOV= number of mature ova in sub-samples

Among the three species, the maturity and spawning season of Dimua chingri (*M. villosimanus*) was determined in terms of GSI and fecundity. The average length of collected Dimua chingri was 9.68±0.98 cm and mean average weight was 12.96±4.34 g. The mean GSI and fecundity of *M. villosimanus* was 13.61±5.56 and 25,259.42±14,994.30 eggs per prawn, respectively during the period of June to August.

Experiment/study 5. Breeding and seed production of brackishwater Dimua Chingri (Macrobrachium villosimanus)

This experiment was conducted in the hatchery complex of Brackishwater Station. The suitable broods were collected from the domesticated ponds and natural sources. Then, the broods were stocked in 0.01 ha brackishwater pond and fed with earthworms and protein rich pellet feeds at the rate of 5 % body weight. Potential broods were harvested from the pond and after that, unilateral and/or bilateral eyestalk was ablated to trigger the reproductive maturation. Once the prawn extruded the egg and aggregates into the abdominal flaps (turned into berried), the berried females were transferred to the incubation tank after proper disinfection with 20-50 ppm formalin solution. A little portion of egg sample was picked with sterilized forceps at 3 days intervals and examined under stereo microscope. Fertilization rate, progress of embryonic development stages and any infection onto the egg mass was noted down. Sufficient photographs were taken for documentation. As the color of egg mass turned into dark grey, the berried females were transferred to the hatching tanks filled with 10-12 ppt disinfected water. After hatching, the larvae were transferred to the prepared larvae rearing tanks and fed with live feed. Prevalence of virulence bacterial pathogen was tested at microbiology laboratory. Finally, powdered feed and artemia nauplii were provided for larval development and nutrition. Fertilization, hatching, survival rate and larval stage index was calculated following the formulae below.

$$\text{Fertilization rate (\%)} = \frac{\text{Number of fertilized eggs in sub-sample}}{\text{Total number of eggs observed in sub-sample}} \times 100$$

$$\text{Hatching rate (\%)} = \frac{\text{Number of hatchlings}}{\text{Total number of fertilized eggs}} \times 100$$

$$\text{Survival rate (\%)} = \frac{\text{Final population (Pt)}}{\text{Initial population (Po)}} \times 100$$

$$\text{Larval Stage Index (LSI)} = \{(A1 \times A2) + (B1 \times B2)\} / C$$

Where, A1= amount of previous stage larvae, A2 = previous stage, B1 = amount of highest stage larvae, B2 = highest stage, C = total amount of sample.

Till reporting, a total of 12 breeding trials were made in the hatchery complex of Brackishwater Station to produce Dimua prawn PL. The fertilization rate was 98.67±0.47 % and hatching rate was 91.33±2.49 %. The embryonic development of *Macrobrachium villosimanus* is consist of nine different stages/phages which were characterized by various notable morphological changes (Figure 5).

Stage 1. Fertilization of eggs (00-04 hours)

Fertilization of eggs continued up to four hours from beginning of fertilization and finished just before the first cell division. Fertilized eggs were almost globular shape and consisted of a granulous mass uniform dark olive color which wrapped with a lucid chorion.

Stage 2. Cleavage (04-09 hours)

Various cleavage furrows arise in the egg mass, pointing up the formation of the first embryonic cells. A translucid region was found at one pole of the egg shrinking slightly the eggs inside mass. These changes occurred at the beginning of embryonic development where eggs volume increased slightly.

Stage 3. Blastula (09-33 hours)

Translucid area of the egg expanded gradually without remarkable changes. Consequently, two parts were observed inside the eggs where the abdominal part of the developing embryo represented by a light region and cephalic area represented by a dark olive region.

Stage 4. Gastrula (33-130 hours)

After 33 hours of fertilization, the internal mass of the egg consolidated mainly in the peripheral region. The abdominal region with the presence of some abdominal segments perfectly separated from the "V" form cephalic region. This stage concluded five days after fertilization.

Stage 5. Nauplius (130-163 hours)

A spacious black spot developed in the cephalic region of the embryo which continuously shortened due to the propagation of abdominal part 140 hours after fertilization, the black spot illustrate a diagram of the embryos ocular region turned more evident. Besides some vitellin reserve vesicles developed in the peripheric part of the cephalic region.

Stage 6. Post-nauplius with a heartbeat (163-190 hours)

Previously developed optic region expanded with more pigmentation. The caudal papilla inhered with an elementary telson at the abdominal region and folded to the optic region. At the moment, remaining contents of the eggs, mainly vitellin reserve slendered due to the evolution of embryonic structures and turn into dark grey color.

Stage 7. Post-nauplius with eye individualization (190-233 hours)

Eyes separated from the optic region, prolonged, turn into an elliptical form and separated from the cephalic region but still stuck on their base.

Stage 8. Final post-nauplius with eye condensation (233-280 hours)

Diameter of eye enlarged with their color intensification and eyelashes arise above each eyes. Well developed and segmented maxillipeds visible in this phase and overlapped the abdomen. Whole egg space was obtained by the embryo. The vitellin vesicles intensified and were more visible in the cephalic region of the embryo.

Stage 9. Pre-hatching (280-305 hours)

Cephalothorax dark grey part was reduced significantly. The heart was fully evident from the vitellin mass and compression of the heart was significantly active than during the previous phases.

Among the 12 breeding trials, the larvae survived up to 18 days for the first 10 events. Thereafter several breeding trials and errors as well as trouble shooting it was possible to produce PL of the prawn species. The PL were stocked in the nursery pond for nursery rearing. Till report the Dimua prawn attains 6.87±0.55 cm length and 2.06±0.87 g body weight after 45 days rearing in the nursery pond. Water quality parameters such as temperature, salinity, pH, dissolved oxygen and ammonia were being recorded weekly. Water temperature was recorded within 20 to 36°C, pH 7.52 to 8.56, salinity 4 to 13 ppt, dissolved oxygen 5.12 to 7.24 ppm and ammonia 0.00 to 0.10 ppm. The water quality parameters of the breeding tank and broodstock ponds are being measured regularly. The broodstock inducement with enriched artificial (commercial diet) and natural feed (earthworms) is ongoing. For histological analysis, gonad of different

stages will be collected. Shrimp is contributing significantly to the national economy of Bangladesh through export earnings and creating employment opportunity. It is the second largest foreign exchange earning source in Bangladesh. But due to low salinity during the majority of the year and frequent mass mortality of this shrimp caused by invasion of WSSV and AHPND/EMS, farmers have become very much cautious about stocking of this shrimp in their ghers and many of the bagda farmers have already intended to shift their culture pattern and searching for suitable species for stocking. Brackishwater prawn farming is one of major activities play important role in socio-economic development of the fishers and increase export earnings of the country. Indigenous prawns have huge domestic and export demand for their taste and ability to purchase. One of the major problems for the commercial prawn farming is the non-availability of the required quantity of quality seeds since it primarily depends on the natural collection of seeds which is uncertain, unscientific and mostly limited supply. Existing hatcheries are not producing seeds up to their installed capacity due to various constraints such as non-availability of healthy broodstock, mid larval cycle disease and mass mortality of larvae leading to low yield. Therefore, the biology, maturation, breeding, life cycle etc. was studied for the successful breeding and culture of the studied prawn in captivity.

Discussion and conclusion

The present study gives baseline information about maturation, breeding and larval development of *M. villosimanus* which concludes that this species could be used as another potential candidate species to commercial brackishwater prawn culture in Bangladesh. This species required 24-28 days to obtain post-larvae from hatchlings which is comparatively a less period than other *Macrobrachium* species with a higher survival rate. As this species is having good market value and consumed by the local people in Khulna division of Bangladesh, the establishment of hatcheries will facilitate utilization of inland and low saline for prawn culture in South-west coastal region of Bangladesh (Figure 4).

A. Brood Dimua Prawn B. Broodstock Pond C. Post-Larvae

D. Monitoring Larvae E. Stocking in Nursery F. Monitoring PL

Figure 4. Observation of brood and larval development

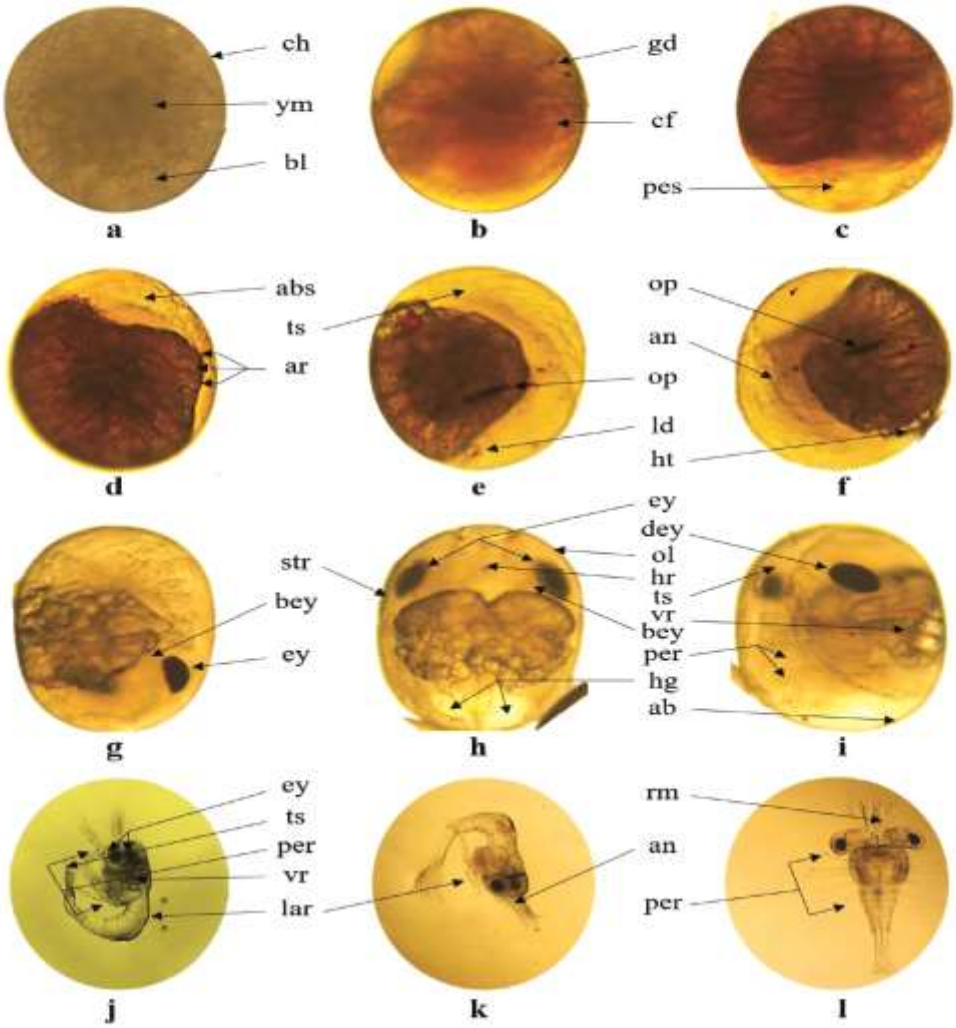


Figure 5. Embryonic stages in brackishwater prawn *M. villosimanus*. a. fertilization; b. cleavage; c. blastula; d. gastrula, making up of blastopores; e. nauplius with black spot; f. post-nauplius with heart beats; g. post-nauplius with eyes pigmentation; h. post-nauplius with eyes condensation; i. pre-hatching; j. freshly hatched larva; k. two days larvae; l. five days larvae. ab. abdomen; abs. abdominal segment; an. antennae; ar. abdominal region; bey. base of the eye; bl. blastomeres; cf. cleavage sillon; ch. chorion; dey. developed eye; ey. eye; gd. germinal disc; hg. heart growth; hr. head region; ht. heart; lar. larva; ld. lipid droplet; ol. optical lobe; op. ocular pigment; per. pereiopods; pes. primitive embryonic structure; rm. rostrum; str. streaks; ts. telson; vr. vitellin reserve; ym. yolk mass.

Improvement of soft-shell mud crab (*Scylla olivacea*) culture technique in south-west coastal region of Bangladesh

Researchers
A.K.M. Shafiqul Alam Rubel, SSO
Debashis Kumar Mondal, SSO
Md. Hashmi Sakib, SSO
Md. Masudur Rahman, SO
Rafia Afrin, SO

Objectives

- To observe the effect of environmental conditions (salinity variations and aeration) on molting efficiency of mud crab.
- To observe the effect of physical stress (limb trimming) on molting of mud crab.
- To compare the performance of soft-shell shedding between hatchery produced and natural crablets.

Achievement

Experiment 1. Effect of various stocking densities on soft-shell shedding of mud crab

The experiment was conducted in Brackishwater old hatchery complex and earthen ponds. Cemented cisterns (each cistern 7 m^2) and earthen ponds (0.1 ha each) were prepared for the experiment. The experiment was conducted with 3 Treatments depending on density variations, *viz*., T_1 (1 crab/ box), T_2 (2 crabs/ box) and T_3 (3 crabs/ box). There was a controlled Treatment T_1. Overall experimental design has been displayed in Table 1.

Table 1. Experimental design of different stocking densities in soft shell shedding.

Treatment	Treatment details	Limb cut	Box size	Crab size (g)	Replica
T_1	1 crab/1 box (C)	both chelipeds	(25×15×15) cm^3	40 - 50	3
T_2	2 crab/1 box	both chelipeds	(25×15×15) cm^3	40 - 50	3
T_3	3 crab/1 box	both ch elipeds	(25×15×15) cm^3	40 - 50	3

Crablets were collected/purchased from local markets and stocked according to design. Twenty crabs (1 crab/1 box) of 40-50 g were stocked in T_1 (control) in each replication. In contrast, in each replication, 40 crabs (2 crab/1 box) of 40-50 g were stocked in T_2 and 60 crabs (3 crab/1 box) of 40-50 g were stocked in T_3. Chopped Tilapias were distributed as feed at every 2-3 days interval and 30% of tank water was being exchanged in every week by considering the water quality.

Crabs were monitored at every 6 hours interval for soft-shell shedding. Molting was observed by the naked eye. Initial weight and weight after soft-shell shedding were recorded. Water quality parameters such as salinity, temperature, pH, dissolved oxygen, alkalinity, and ammonia were monitored and recorded on a weekly basis.

In the pond, the water quality parameters were recorded on a weekly basis during the culture period (Table 2). The salinity level was in between 2.00 and 4.5 ppt. Though the recorded temperature of this experiment has fluctuated slightly in every week, it ranged between 24.92°C and 28.43°C. At the same time, presence of suitable range of dissolved oxygen was also very essential in case of oxidation of various chemical compounds and respiration process, which leads to increase feed intake, growth and moulting. In the present study, the range of dissolved oxygen was 5.00-8.98 ppm. The recorded values of other parameters like pH, ammonia, and total alkalinity varied between 7.32-8.50, 0-1.00 ppm and 97-144 ppm, respectively throughout the experimental period.

Table 2. Water quality parameters throughout the culture period.

	Temperature (°C)	pH	Dissolved oxygen (ppm)	Total ammonia (ppm)	Alkalinity (ppm)	Salinity (ppt)
Minimum	24.92	7.32	5	0	97	2
Maximum	28.43	8.50	8.98	1.0	144	4.5

Stocking density has some critical feedback on growth rate, survival rate, weight gain and duration of an aquatic species. In this study, it was found that the mean weight gain (%) and increased carapace width (%) of T_1 was 43.12±4.39 and 15.52±1.92, T_2 was 39.07±1.06 and 14.84±1.51 and T_3 was 34.88±6.62 and 12.93±1.59 (Figure 1). In T_1, growth performance was better than rest two as there was one crab in a single box. So that they faced no competition for food and no cannibalism was occurred there. In T_3, there were three crabs in a single box. So, they got little space and faced competition for food which might hamper their growth and percentage of total weight gain. On the other hand, in T_2, mean weight gain and increase of carapace wide was near to T_1 and attained the second highest values.

However, if we consider the shedding period, T_2 showed better response than T_1 and T_3. The lowest performance was observed in T_2 as it took highest days to produce soft-shell (47 days). Likewise, T_1 showed 45 days for growing soft-shell in the farm. Therefore, the quickest soft-shell production was found in T_2 as displayed in Table 3. The overall shedding performance of female was better than male throughout the culture period and it was 55.55% male and 44.44% female in T_1, 44.44% male and 55.55% female in T_2 and 33.33% male and 66.66% female in T_3.

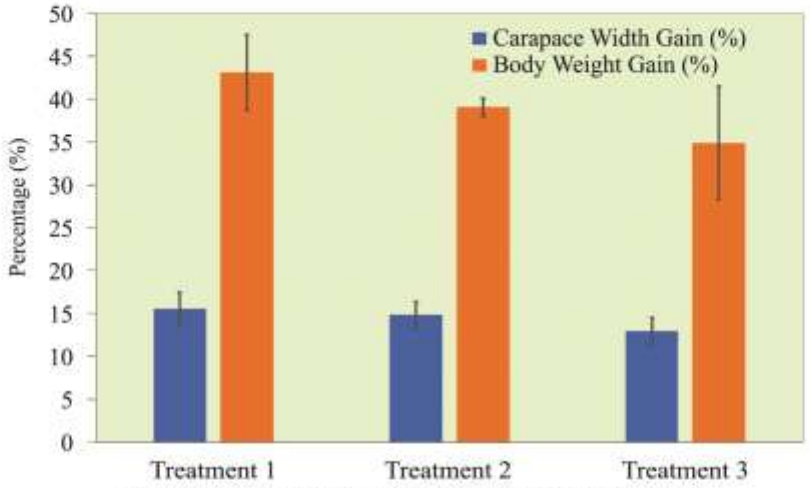


Figure 1. Carapace width and body weight gain percentage of soft-shell.

Table 3. Duration of shedding performance, male and female ratio during soft-shell production.

Treatments	Shedding period	Male (%)	Female (%)
T_1	45.66± 2.44	55.56	44.44
T_2	39.66±2.54	44.44	55.56
T_3	47.44±1.5	33.33	66.66

Effects of stocking density on overall survival rate of mud crab during soft-shell farming has been presented in Figure 2. The overall survival rate (%) of soft-shell mud crab was 53.33±2.88, 61.67±1.44, and 51.66±1.66 in T_1, T_2 and T_3, respectively. The survival rate in T_3 was the lowest among the all Treatments, whereas, the highest survival rate was calculated in Treatment 2.

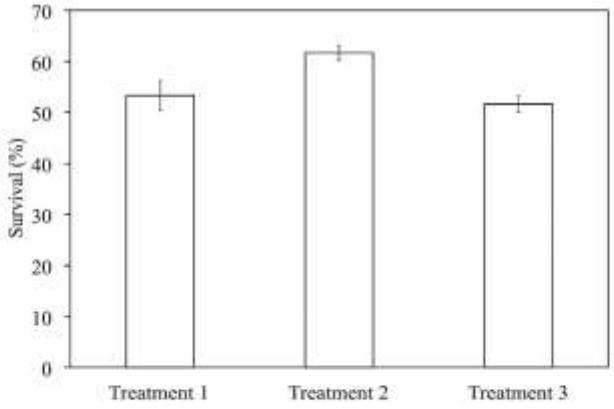


Figure 2. Effects of stocking density on survival rate in soft-shell farming.

Cemented cisterns were used to conduct indoor study for producing soft-shell mud crab. The recorded physico-chemical parameters, namely salinity, temperature, pH, dissolved oxygen, total ammonia and alkalinity of water during the culture period has been presented in Table 4. Generally, all the parameters of the cisterns were within the acceptable ranges for brackish water aquaculture of crab. The temperature of the experimental ponds ranged between 27℃ and 29.5℃. Salinity is considered as one of the most fundamental factors for mud crab culture. The salinity level was kept at 15 ppt in all cisterns. The recorded pH of this study ranged from 7.82 to 8.65. Ammonia, dissolved oxygen, and total alkalinity levels of the cisterns were found within a range of 0-0.25, 7.4-9.10, and 84-116 ppm, respectively.

Table 4. Water quality parameters throughout the culture period.

Parameters	Treatment -1 (1 crab/ box)	Treratment -2 (2 crab/ box)	Treatment -3 (3 crab/ box)
Salinity (ppt)	15	15	15
Temperature (^{0}C)	27-29.5	27-29.5	27-29.5
pH	8.22-8.65	8.06-8.3	7.82-8.09
Ammonia (ppm)	0.0-0.25	0.0-0.25	0.0-0.25
Dissolved oxygen (ppm)	8.2-9.0	8.1-9.1	7.4-8.7
Total alkalinity (ppm)	90-112	84-128	86-116

Stocking density has major effects on growth rate, survival rate, weight gain and duration of shedding of soft-shell. In the present study, it was found that the mean weight gain (%) and increase of carapace width (%) was 48.79±3.89 and 15.85±3.36, in T_1; 48.65±5.10 and 15.73±2.91 in T_2 and 29.94±7.53 and 10.30±1.64 in T_3 (Figure 3). In T_1, growth performance was better than rest two Treatments. In T_2, growth performance was very close to T_1. However, T_3 exhibited poorest performence among all these Treatments.

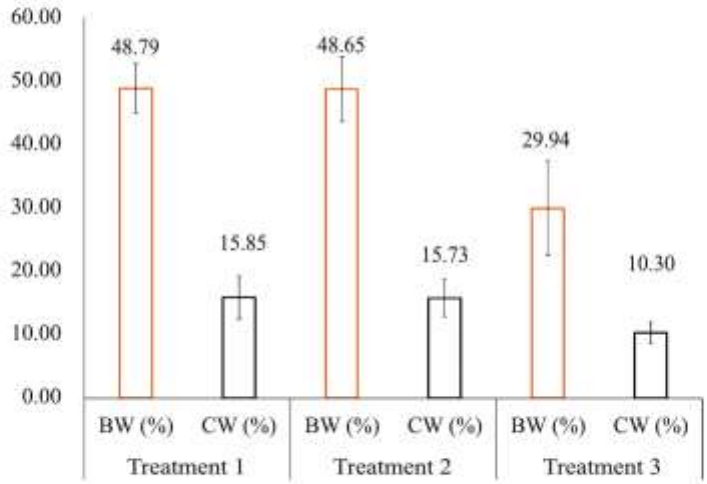


Figure 3. Body weight and carapace width gain (%) of soft-shell mud crab.

Figure 4 represents the overall duration of shedding performance during the experimental period. T_2 showed better response than T_1 and T_3. The lowest performance was observed in T_2 as it took highest days to produce soft-shell (35.56 days). Likewise, T_1 showed 31.89 days for growing soft-shell in the farm. Therefore, the quickest soft-shell production was found in T_2 (30.78 days).

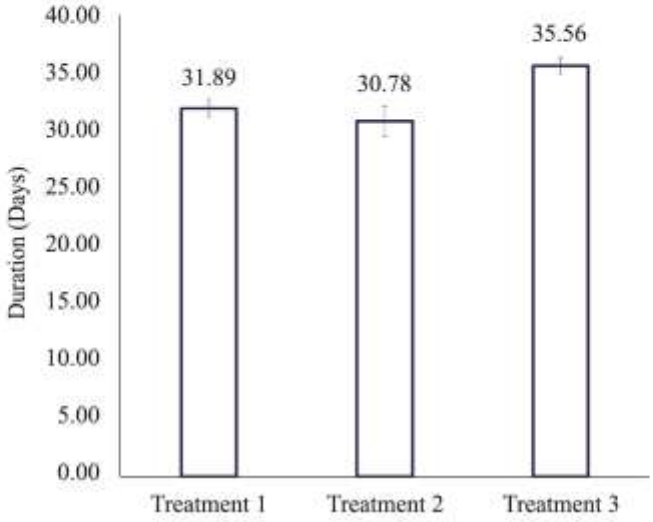


Figure 4. Duration of shedding performance.

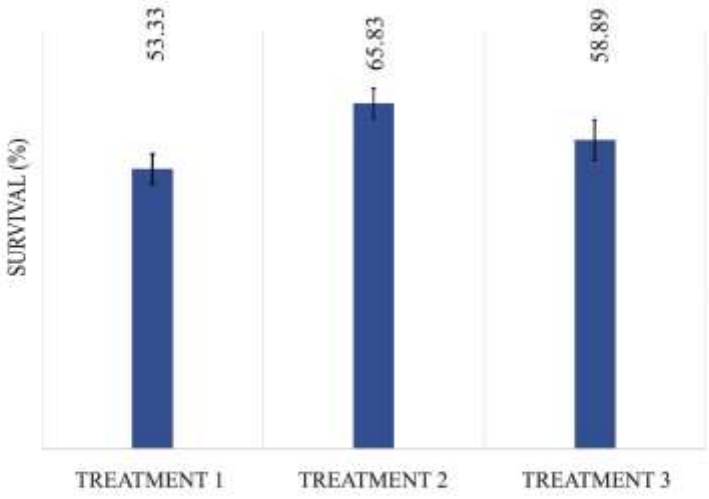


Figure 5. Effect of stocking density on survival rate.

Effect of stocking density on overall survival rate of mud crab in different Treatments during soft-shell farming has been presented in Figure 5. The overall survival rate (%) of soft-shell mud crab was 53.33±2.89, 65.83±2.89, and 58.89±3.85 in T_1, T_2 and T_3, respectively. The survival rate in T_3 was the lowest among all Treatments, whereas the highest survival rate was calculated in Treatment 2.

Experiment 2. Effect of removal of chelipeds on soft-shell shedding of mud crab

Cemented cisterns (Each Cistern 7 m^2) and earthen ponds (0.1 ha each) were prepared for this experiment. At first, the experiment was conducted and repeated in the brackishwater earthen pond. This trial was conducted with 3 Treatments depending on the number of cheliped removal, *viz*., T_1 (no cheliped removal), T_2 (removal of single cheliped) and T_3 (removal of both chelipeds). There was a control Treatment (T_1) with a density of 2 crabs/ 1 box. Overall experimental design has been shown in Table 5.

Table 5. Experimental design for removal of chelipeds in soft shell shedding.

Treatment	Treatment details	Stocking	Box size	Crab size (g)	Replica
T_1	No removal of cheliped (C)	2 crab/1 box	(25×15×15) cm^3	40 - 50	3
T_2	Removal of single cheliped	2 crab/1 box	(25×15×15) cm^3	40 - 50	3
T_3	Removal of both cheliped	2 crab/1 box	(25×15×15) cm^3	40 - 50	3

Crablets were collected/purchased from local market and were stocked according to the design. Fourty crabs (2 crab/1 box) of 40-50 g were stocked for each replication of all Treatments. Chopped Tilapia were supplied as food at every 2-3 days interval and 30% of tank water was exchanged in every week by considering the physico-chemical parameters of water. Crabs were monitored at every 6 hours interval for soft-shell shedding. Moulting was observed through both naked eyes. Initial weight and weight after soft-shell shedding was recorded. Water quality parameter such as salinity, temperature, pH, dissolved oxygen, alkalinity and ammonia were monitored and recorded on a weekly basis.

Temperature, salinity, dissolved oxygen, pH, ammonia and total alkalinity of pond water were recorded in every week during the culture period (Table 6). The salinity level in the present experiment was between 4-12 ppt. Temperature ranged between 28.8°C and 34°C. The range of dissolved oxygen was 5.80 to 9.70 ppm which was in suitable range for soft-shell shedding. The recorded values of other parameters like pH, ammonia, and total alkalinity varied between 7.78-8.95, 0-0.25 ppm and 87-128 ppm, respectively during the trial.

Table 6. Water quality parameters throughout the culture period.

	Temperature (°C)	pH	Dissolve oxygen (ppm)	Total ammonia (ppm)	Alkalinity (ppm)	Salinity (ppt)
Minimum	28.8	7.78	5.8	0	87	4
Maximum	34	8.95	9.7	0.25	128	12

Generally, chelipeds has some effects on growth rate, survival rate, weight gain and duration of shedding of mud crab. In the present study, it was found that the mean weight gain (%) and increase of carapace width (%) of T_1 was 32.41±11.10 and 14.48±6.78, T_2 was 47.13±11.45 and 14.81±4.09 and T_3 was 72.41±10.93 and 21.21±2.97 (Figure 6). In T_3, growth performance was better than rest two Treatments.

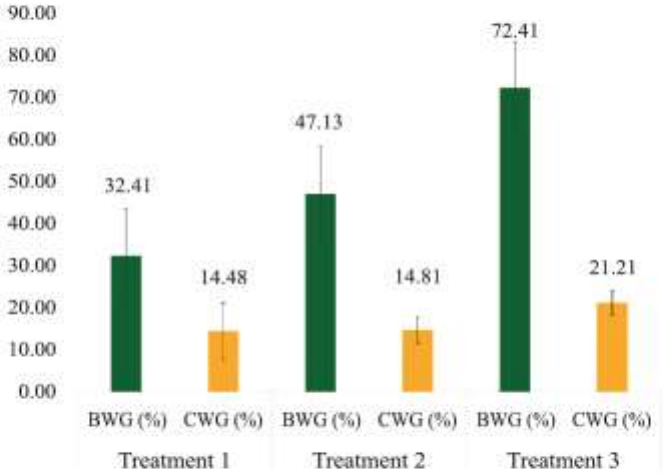


Figure 6. Carapace width and body weight gain percentage of soft-shell.

Again, if we consider the shedding period, T_3 showed better response (21.44 days) than T_2 (29.56 days) and T_1 (34.44 days) as displayed in Figure 7. The shedding period in T_3 was the lowest among all Treatments, whereas, the highest shedding period was calculated in Treatment 1. Shedding efficiency was found the highest in Treatment 3.

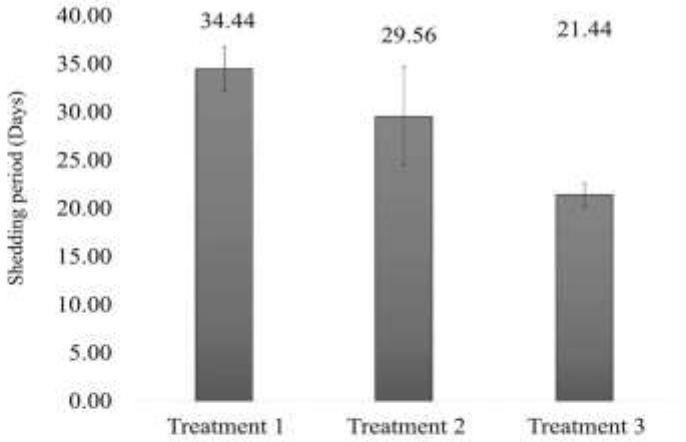


Figure 7. Duration of shedding performance.

Effect of cheliped removal on overall survival rate of mud crab during soft-shell farming has been presented in Figure 8. The overall survival rate (%) of soft-shell mud crab of experiment was 44.17, 50, and 60 in T_1, T_2, and T_3 respectively. The survival rate in T1 was the lowest among the all Treatments, whereas, the highest survival rate was calculated in Treatment 3.

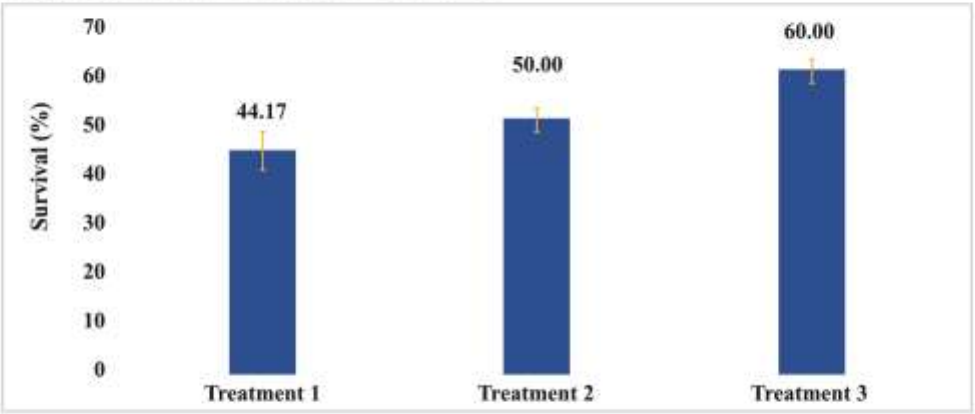


Figure 8. Effect of cutting chelipeds on survival rate in soft-shell farming.

Thereafter, cemented cisterns were used to conduct indoor study for producing soft-shell mud crab. The recorded physico-chemical parameters, namely salinity, temperature, pH, dissolved oxygen, total ammonia and alkalinity of water during the culture period in this research has been presented in Table 7. Generally, all the parameters of the cisterns were within the acceptable ranges for brackish water aquaculture of crab. The temperature of the cisterns ranged between 27.1°C and 31.1°C. Salinity is considered as one of the most fundamental factors for mud crab culture. The salinity level was kept at 15 ppt in all cisterns. The recorded pH of this study ranged from 7.85 to 8.65. Ammonia, dissolved oxygen, and total alkalinity levels of the cisterns were found within a range of 0-1, 5.4-8.6, and 120-182 ppm, respectively.

Table 7. Water quality parameters throughout the culture period.

Parameters	Treatment -1 (2 crab/ box)	Treratment -2 (2 crab/ box)	Treatment -3 (2 crab/ box)
Salinity (ppt)	15	15	15
Temperature (^{0}C)	27.1-31.1	27.2-29.6	27.3-29.6
pH	8.22-8.65	7.91-8.32	7.85-8.25
Ammonia (ppm)	0-1	0-1	0-1
Dissolved oxygen (ppm)	5.8-8.6	5.7-8.43	5.4-8.55
Total alkalinity (ppm)	120-182	124-158	120-155

Generally, chelipeds has some crucial effects on growth rate, survival rate, weight gain and duration of shedding of mud crab. In the present study, it was found that the mean weight gain (%) and increase of carapace width (%) of T_1 were 22.54±5.66 and 13.41±1.63, T_2 were 35.48±5.32 and 20.28±5.33 and T_3 was 53.84±8.37 and 28.12±3.03 (Figure 9). In T_3, growth performance was better than rest two Treatments, whereas the lowest value was calculated in Treatment 1 for both the parameters.

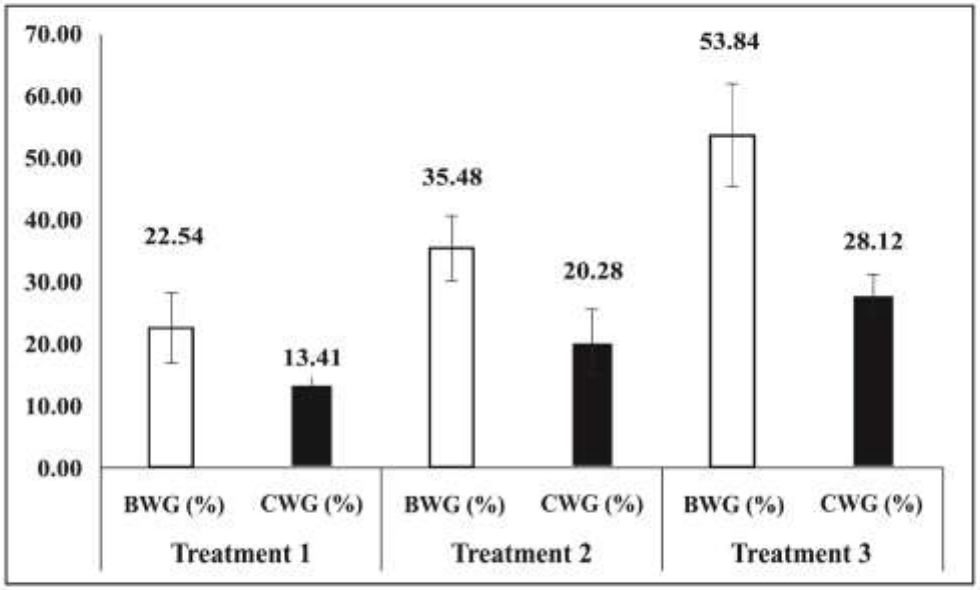


Figure 9. Body weight and carapace width gain (%) of soft-shell mud crab.

Figure 10 represents the overall duration of shedding performance during the experimental period. T_3 showed better response than T_1 and T_2. The lowest performance was observed in the T_1 as it took highest days to produce soft-shell (34.44 days). Likewise, T_2 showed 29.22 days for growing soft-shell in the farm. Therefore, the quickest soft-shell production was found in T_3 (20.33 days).

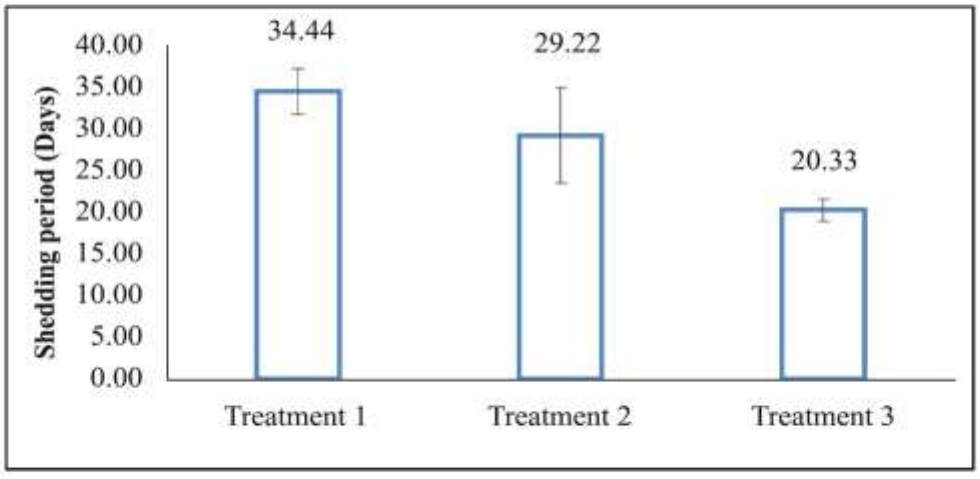


Figure 10. Duration of shedding performance.

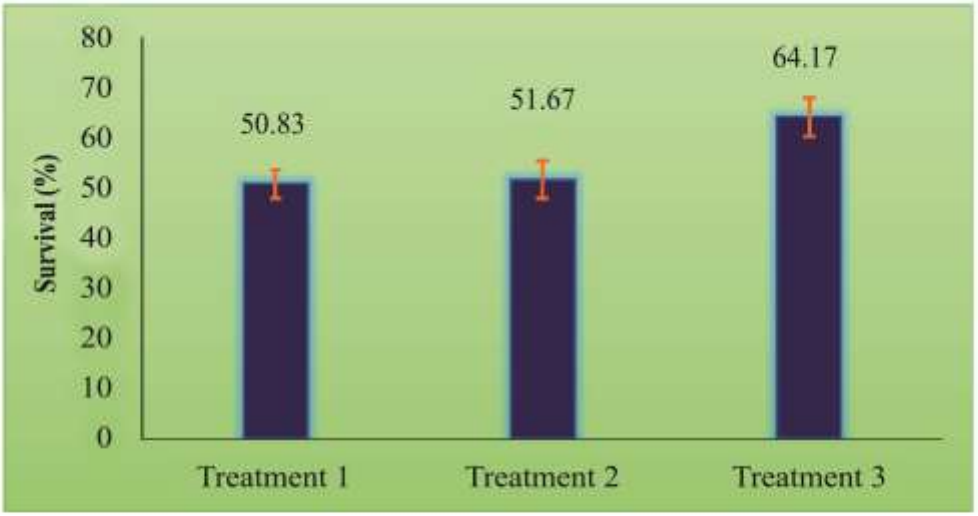


Figure 11. Effect of stocking density on survival rate.

Effect of cheliped removal on overall survival rate of mud crab during soft-shell farming has been presented in Figure 11. The overall survival rate (%) of soft-shell mud crab of experiment was 50.83, 51.67, and 64.17 in T_1, T_2, and T_3 respectively. The survival rate in T_1 was the lowest among all Treatments, whereas, the highest survival rate was calculated in Treatment 3.

Discussion and conclusion

Soft-shell mud crab farming is very new concept in Bangladesh, and it is solely dependent on single stocking as mud crab is very aggressive. Thus, chelipeds removal has also some significant effects on shedding efficiency and survival of soft-shell. Soft-shell mud crab aquaculture can be a new horizon for ensuring global food security. Development of soft-shell mud crab farming in controlled condition is the primary challenge in rural farms. Therefore, this study on soft-shell shedding considering pond and hatchery (controlled) condition deserve considerable attention.

In terms of stocking density, double stocking in a single box performed well and took 30-39 days for moulting. Survival rate was found between 61-65%. However, removal of cheliped expedited molting rate of immature crabs prior stocking. Cutting of both chelipeds took only 21 days to produce soft-shell during the culture period. However, the survival rate was a bit low (60%) as they are highly cannibalistic in nature. Generally, crabs are very cannibalistic in nature. These experiments give birth new ways to cope with low productivity, mortality and financial loss. However, by considering present findings soft-shell production can be increased in the same cultivable land without expanding extra land area.

A. Sludge removing. B. Liming. C. Building a farm.

D. Stocking crabs. E. Monitoring water. F. Soft-shell crab.

Figure 12. Managament practices in soft-shell mud crab farming

Domestication, breeding and seed production of some commercially important brackishwater fish

Researchers

Dr. Md. Latiful Islam, PSO
Debashis Kumer Mondal, SSO
Shawon Ahmmed, SO
Md. Abu Naser Mision, SO
Shahnaj Parvin, SO

Objectives

- To enhance the survival rate of Chitra and Datina fish seed
- To domesticate the commercially important fish in brackishwater environment
- To observe the food-feeding habit and reproductive biology of the fish
- To develop induced breeding, seed production and nursery technique of the fish

Achievements

Experiment-1. Captive breeding and optimization of feeding regimen in larvae rearing of Datina (P. hasta) for enhancement of survival rat

Larvae rearing protocol of *P. hasta* was optimized and sub-sequent nursery rearing was done to enhance the survival of the fish. Breeding was done with LHRHa hormone at the dose of 50.25 µg/kg fish for female and male fish. As the fish spawn and hatched the larvae was stocked in the larvae rearing tanks at the rate

of 30 larvae/l and standard green water (microalgae; 0.5×105/ml) was used as culture medium. The experiment was designed with five different Treatments *viz.*, feeding of rotifer for 3 days=T_1, 5 days=T_2, 7 days=T_3, 9 days=T_4, 11 days=T_5 followed by "Artemia+rotifer" feeding for next 7 days and "Artemia+Copepod+commercial diet" feeding up to 30 days for each Treatment. The trial was conducted in 100 liter plastic buckets with 3 replications of each. Water salinity was fixed at 25 ppt and feeding was started after 3 days of hatching. The survival rate under different feeding regimen has been expressed in Table 1. The survival rate in T_4 (18.61±1.71%) and T_5 (17.78±1.04%) were significantly higher ($p<0.05$) than T_1 (0.00±0.00%), T_2 (2.22±0.39%) and T_3 (8.33±0.68%); though, there was no significant difference between T_4 and T_5. Nevertheless, T_4 showed maximum survival rate among all the Treatments indicated the best combination of feeding regimen for larval rearing of Datina (*P. hasta*) fish.

Table 1. Experimental design for optimization of feeding regimen of Datina larvae.

Treatment	Feeding regimen and duration			Survival rate (%)
	Rotifer	Artemia+rotifer	Artemia+Copipod+ commercial diet	
T_1	for 3 days	for next 7 days	for up to 30 days	0.00 ± 0.00^{d}
T_2	for 5 days	for next 7 days	for up to 30 days	2.22 ± 0.39^{c}
T_3	for 7 days	for next 7 days	for up to 30 days	8.33 ± 0.68^{b}
T_4	for 9 days	for next 7 days	for up to 30 days	18.61 ± 1.71^{a}
T_5	for 11 days	for next 7 days	for up to 30 days	17.78 ± 1.04^{a}

Experiment-2. Evaluation of different commercial diets for the nursery rearing of Datina (P. hasta) fry

The feeding trail was carried out in 15 circular fiber glass tanks (300 L capacity each) under five different Treatments *viz.*, T_1 = feeding with CP feed, T_2 = feeding with Mega feed, T_3 = feeding with Tongway feed, T_4 = feeding with Quality feed, and T_5 = feeding with Naurish feed. The same aged (30 days) uniform size (1.53±0.20 cm) of Datina fry were collected from BS hatchery and were randomly distributed into each Treatment and replications accordingly. Feeding was done at the rate of 5% body weight. Weekly samplings of fish was performed to adjust the daily feed ration for the following week and to estimate the growth performance. Water of the rearing tanks was exchanged at the rate of 20-25% at weekly basis. As stipulated in Table 2, the gained average body weight after 75 days of nursery rearing in T_1 (Length. 7.05±0.15 cm and weight. 5.76±0.23 g) were significantly higher ($p<0.05$) than other Treatments. Meanwhile, Tongway feed (4.98±0.15 g) and Mega feed (4.53±0.14 g) stood second in terms of body weight gain. Survival was more or less similar in all the Treatments with the highest (74%) in CP feeding Treatments. However, Datina as a carnivorous fish needed high protein levels for proper growth and CP feed provided the requirement.

Table 2. Growth performance of Datina fry after 75 days of nursery rearing.

Treatment	Treatment details	stocking density/ton	Water salinity	Average body Length (cm)	Average body weight (g)	Survival (%)
T_1	CP feed	500	10-15	7.05 ± 0.15^{a}	5.76 ± 0.23^{a}	74%
T_2	Mega feed	500	10-15	6.10 ± 0.28^{b}	4.53 ± 0.14^{b}	72%
T_3	Tongway feed	500	10-15	6.65 ± 0.20^{b}	4.98 ± 0.13^{b}	72%
T_4	Quality feed	500	10-15	4.81 ± 0.17^{c}	2.18 ± 0.21^{c}	68%
T_5	Naurish feed	500	10-15	5.03 ± 0.22^{c}	2.31 ± 0.34^{c}	71%

Experiment-3. Domestication and brood development of Bhangon (Mugil cephalus), Royna/Rekha (Datnioides polota), Kain Magur (Plotosus canius), Bele (Glossogobius giuris) and Taposi (Polynemus paradiseus) under controlled condition

Fry (0.1g) and/or sub-adult (40 g) of *M. cephalus*, *D. polota*, *P. canius* and *G. giuris* was collected from river and reared in 0.01-0.1 ha on-station ponds. Stocking density was maintained at 10 fish/decimal. Growth performance and water quality variables was monitored weekly basis and maturity/gonad development will be monitored after one year age with the frequency of weekly basis. Tongway commercial floating and sinking feed was applied for rearing of all fish at 25%-5% of BW. Unfortunately, it was not possible to collect any live fish of *P. paradiseus* from any source due it's high sensitivity.

Till report, the average length and body weight of *M. cephalus* in the domestication pond were 9.42±0.78 cm and 8.947±1.12 g, respectively. However, sub-adult (40 g) of *D. polota* were collected from the local and wild sources and stocked in the on station (0.01-0.1 ha) broodstock domestication ponds at a stocking density of 10 fish/decimal. Till report, the gained average length and body weight of *D. polota* in the domestication pond were 11.23±3.12 cm and 182.35±27.44 g, respectively. In addition, fingerlings of *G. giuris* were collected and stocked in broodstock pond (0.1 ha) at a stocking density of 10 fish/decimal. Till report, the gained average length and body weight of *G. giuris* in the domestication pond were 12.27±2.21 cm and 28.12±4.98 g, respectively. About 20 individuals of *P. canius* were stocked in the broodstock pond (0.1 ha). But it is very difficult to collect the regular data on growth performance and gonadal development as this species habituated into the burrows under the pond bottom ground. In case of *P. paradiseus*, about 10 dead fish samples were collected in every month for histological analysis. But, the researchers could not find any live samples from any sources. Moreover, the researchers are still trying to collect the fry or sub-adult of *P. canius*, *G. giuris* and *P. paradiseus* for the domestication and broodstock development. As soon as, the researchers could collect, it will be stocked in the domestication ponds and necessary data will be gathered.

Experiment. 4. Food and feeding habits of brackishwater finfish (Bhangon, Royna, Kain Magur, Bele and Taposi)

Fish of different size and sexes was collected from the natural sources in Paikgacha region. The stomach was dissected and stomach content was measured qualitatively and quantitatively. The different food fractions were measured by the counting method. The food items composed of microscopic organisms were counted in aliquot samples on a Sedgewick Rafter chamber then total food items were converted into 100.

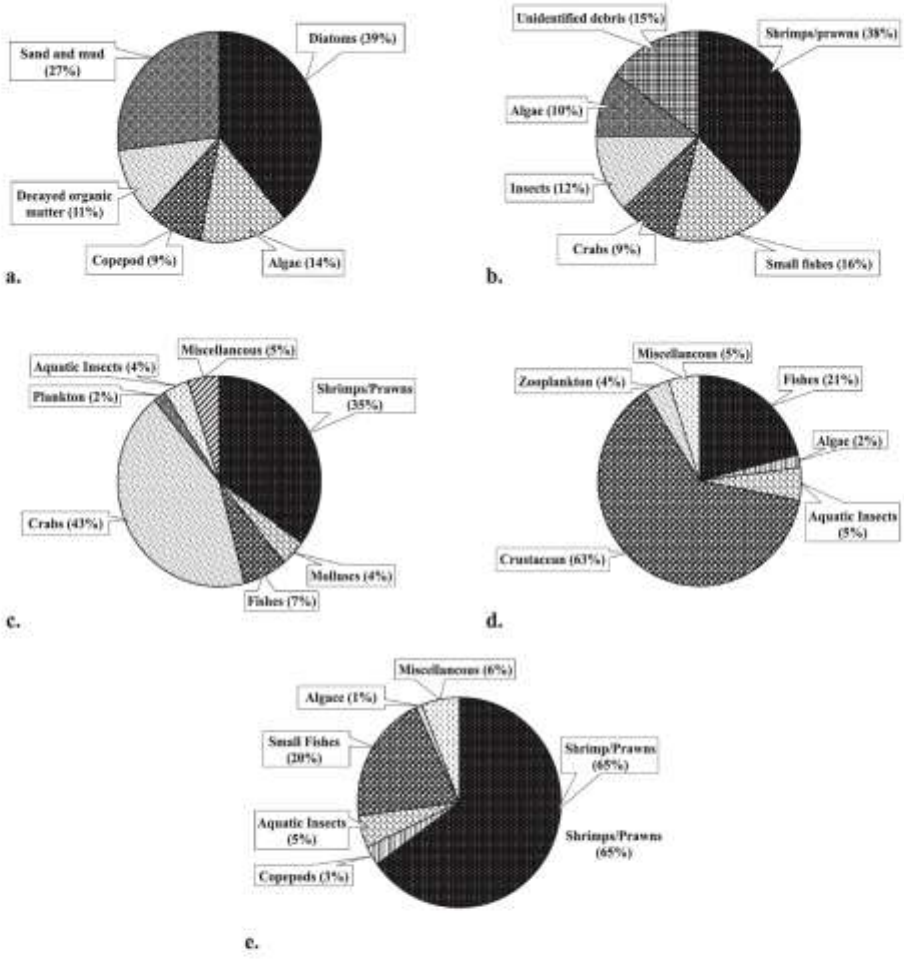


Figure 1: Percentage of different food materials of *M. cephalus* (a); *D. polota* (b); *P. canius* (c); *G. giuris* (d); and *P. paradiseus* (e)

The average percentage composition of food items in the stomach of *M. cephalus* were diatoms 39%, algae 14%, copepods 9%, decayed organic matter 11% and sand and mud 27% (Figure 1.a). Dietary components of *M. cephalus* proved undoubtedly that the fish obtains its diet consisting mainly of fresh and decaying algae from the benthic zones. So, the fish could be defined as herbivore to omnivore.

D. polota feed on shrimps/prawns, small fish, crabs, insects and algae. Of these food items, the highest percentage of shrimps/prawns (38%) and the lowest of crabs (9%) were observed in the stomach of *D. polota.* However, small fish (16%), insects (12%) and algae (10%) and unidentified debris (15%) were also observed in the alimentary canal. Therefore, the feeding habit of *D. polota* was determines as carnivorous (Figure 1.b).

The stomach contents in *P. canius* were composed of six major groups *viz*; crabs (43%), shrimps/prawns (35%), fish (7%), molluscs (4%), aquatic insects (4%), plankton (2%) and unidentified items (5%). Maximum level of stomach fullness of *P. canius* was found with crabs, prawns and fish. The diet composition revealed a wide range of animal origin prey items indicating that *P. canius* as a carnivorous and crusher fish (Figure 1.c).

Based on qualitative and quantitative analysis of gut contents, *G. giuris* has been categorized as carnivorous. Food items mainly consist of fish, crustaceans, insects, zooplankton and algae. The main food items in guts were recorded as Crustacean (63%), small fish (21%), aquatic insects (5%), zooplankton (4%), algae (2%) and miscellaneous (5%) by volume (Figure 1.d).

The average percentage composition of food items in the stomach of *P. paradiseus* (Figure 1.e) were shrimps/prawns (65%), small fish (20%), copepods (3%), aquatic insects (5%), algae (1%) and unidentified items (6%). Dietary components of *P. paradiseus* proved undoubtedly that the fish obtains its diet consisting mainly of animal origin. So the fish was found to be carnivorous.

Experiment. 5. Determination of maturity and peak spawning season of the proposed fin fish

Fish samples both male and female were collected fortnightly from the local market, fishermen and brackishwater ponds. The collected fish were sorted according to sex. The fish were dissected and gonad/testes were gently removed, dried with the help of blotting paper and weighed. Then the gonads of fish were preserved in well labeled vials containing bouin's fixative for histological studies. Gonad somatic index (GSI) of the male and female fish of the collected samples was determined separately following the equation. GSI= (Weight of gonad / Weight of fish) × 100

M. cephalus

A total of 51 female Bhangon fish samples were studied from November 2021 to July 2022 of which samples for the month of May to July were eligible for the fecundity estimation. The highest gonad weight 111.23±9.34 g and gonadosomatic index 8.88±0.76 were identified in the month of May and the lowest value was recorded in the month of November (GW = 0.56±0.06 and GSI= 0.14±0.01). Highest fecundity 3780265±850960 egg/fish was observed in July (Table 3).

Table 3. Month-wise TL, BW, GW, GSI and Fecundity of *Mugil cephalus* in the south-western coastal region of Bangladesh.

Month	Sex (n)	TL (cm) Mean ± SD	BW (g) Mean ± SD	GW (g) Mean ± SD	GSI Mean ± SD	Fecundity Mean ± SD (n)
November	F (5)	33.5±3.22	423.42±49.81	0.56±0.06	0.14±0.01	-
December	F (4)	31.2 ± 1.41	316.69±32.88	0.64±0.07	0.21±0.02	-
January	F (6)	44.80±5.61	880.45±57.46	3.25±0.28	0.37±0.04	-
February	F (5)	49.6±1.83	1467.05±131.52	6.19±0.08	0.42±0.04	-
March	F (7)	44.7±2.47	900.57±185.65	3.96±2.250	0.43±0.18	-
April	F (6)	45.62±3.83	1145.65±143.22	7.27±0.13	0.64±0.11	-
May	F (6)	42.6±3.47	889.15±76.46	111.23±9.34	8.88±0.76	2317033±12000
June	F (7)	43.50±4.60	1045.51±341.20	69.06±10.26	0.06±0.01	2864650±550300
July	F (5)	43.08±5.91	1010.45±245.83	110.23±15.96	8.09±1.03	3780265±850960

Note. n = number of fish; TL = Total Length; BW = Body Weight; GW = Gonad weight

D. polota

A total of 57 female Royna fish samples were studied from November 2021 to July 2022 of which twenty-five fish samples were eligible for the fecundity estimation. The highest gonad weight 64.03±6.15 g, gonadosomatic index 18.24±3.01 and fecundity 486605±24112 egg/fish were identified in the month of May and the lowest value was recorded in the month of November (GW=0.65±0.11 and GSI=0.18±0.05) (Table 4).

Table 4. Month-wise TL, BW, GW, GSI and Fecundity of *Datnioides polota* in the south-western coastal region of Bangladesh.

Month	Sex (n)	TL (cm) Mean ± SD	BW (g) Mean ± SD	GW (g) Mean ± SD	GSI Mean ± SD	Fecundity Mean ± SD (n)
November	F (6)	28.27±5.22	357.83±53.43	0.65±0.11	0.18±0.05	-
December	F (5)	31.2±1.41	316.69±32.88	0.64±0.07	0.21±0.02	-
January	F (3)	14.34±0.98	68.36±16.89	0.27±0.07	0.43±0.21	-
February	F (4)	20.67±3.41	259.16±56.72	1.56±1.41	0.73±0.61	-
March	F (14)	19.35±3.75	167.68±86.79	3.92±1.16	2.34±0.44	-
April	F (8)	21.35±4.17	205.76±73.22	7.87±1.15	4.15±0.41	198456±56487
May	F (6)	20.41±3.13	216.33±56.18	64.03±6.15	18.24±3.01	486605±24112
June	F (4)	20.93±3.82	210.59±69.27	60.08±6.89	16.89±2.81	430400±22300
July	F (7)	18.94±3.57	198.13±70.53	40.24±5.73	12.22±1.89	370200±34300

Note. n = number of fish; TL = Total Length; BW = Body Weight; GW = Gonad weight

P. canius

A total of 73 female Kain Magur samples were studied from November 2021 to July 2022. No ovary was traced until May and the ovary eligible for the fecundity estimation was noticed in the month June. The highest gonad weight 58.85±33.20 g was in the month of July, highest gonadosomatic index 6.68±1.09 and fecundity 1010±80 egg/fish was in June. Lowest value was recorded in the month of November (GW=0.47±0.01 and GSI=0.12±0.01) (Table 5). It is difficult to find the large sized female Kain Magur fish in nature and nearby fish market.

Table 5. Month-wise TL, BW, GW, GSI and Fecundity of *Plotosus canius* in the south-western coastal region of Bangladesh.

Month	Sex (n)	TL (cm) Mean ± SD	BW (g) Mean ± SD	GW (g) Mean ± SD	GSI Mean ± SD	Fecundity Mean ± SD (n)
November	F (4)	40.70±1.13	391.90±30.46	0.47±0.01	0.12±0.01	-
December	F (10)	24.63±3.48	134.14±40.99	0.65±0.59	0.46±0.85	-
January	F (12)	28.27±8.25	162.77±142.87	0.86±0.32	0.64±0.45	-
February	F (7)	32.20±5.18	214.91±94.19	0.77±0.58	0.69±0.74	-
March	F (10)	27.63±7.01	238.82±88.87	1.75±0.05	0.74±0.16	-
April	F (7)	42.25±13.22	448.98±94.34	3.45±0.05	0.89±0.12	-
May	F (6)	39.20±3.18	514.91±64.44	6.77±1.78	1.32±0.34	-
June	F (9)	37.89±7.89	380.41±139.56	50.61±28.32	6.68±1.09	1010±80
July	F (8)	42.53±8.12	457.39±235.41	58.85±33.20	5.45±1.93	819.50±51

Note. n = number of fish; TL = Total Length; BW = Body Weight; GW = Gonad weight

G. giuris

A total of 72 female Bele fish samples were studied from November 2021 to July 2022 of which twenty-nine fish samples were eligible for the fecundity estimation in the month of November-December 2021 and June-July 2022. The highest gonad weight 4.78±2.51 g and gonadosomatic index 7.73±2.11 were observed in the month of November 2021 and the lowest value was recorded in the month of February 2022 (GW=0.18±0.05 and GSI=0.15±0.09) (Table 6). The average fecundity was documented 79501±7565 eggs/fish in the December followed by 67282±5238 eggs/fish in November 2021 (Table 6).

Table 6. Month-wise TL, BW, GW, GSI and Fecundity of *Glossogobius giuris* in the south-western coastal region of Bangladesh.

Month	Sex (n)	TL (cm) Mean ± SD	BW (g) Mean ± SD	GW (g) Mean ± SD	GSI Mean ± SD	Fecundity Mean ± SD (n)
November	F (11)	14.21±5.58	38.69±49.22	4.78±2.51	7.73±2.11	67282±5238
December	F (3)	25.20±1.35	129.88±13.21	4.64±1.24	3.42±4.91	79501±7565
January	F (7)	13.70±1.38	25.56±7.39	0.22±0.08	0.34±0.30	-
February	F (7)	20.17±6.04	80.49±36.84	0.18±0.05	0.15±0.09	-
March	F (13)	27.63±7.01	238.82±88.87	1.75±0.05	0.74±0.16	-
April	F (7)	19.45±6.36	76.29±39.51	0.22±0.14	0.26±0.14	-
May	F (9)	13.70±1.38	25.56±7.39	0.22±0.08	0.06±0.01	-
June	F (8)	16.91±0.89	43.35±5.12	1.00±0.36	2.36±0.86	22012.00±2593.73
July	F (7)	22.60±4.49	109.92±14.08	2.55±1.06	3.71±0.76	8931.33±1824.43

Note. n = number of fish; TL = Total Length; BW = Body Weight; GW = Gonad weight

P. paradiseus

A total of 112 female Taposi fish samples were studied from November 2021 to July 2022 of which 76 fish samples were eligible for the fecundity estimation in the month of November 2021 and March to July 2022. The highest gonad weight 8.12±2.57 g and gonadosomatic index 11.10±0.91 were identified in the month of June 2022 and the lowest value was recorded in January 2022 (GW=0.15± 0.06 and GSI=0.28±0.07) (Table 7). The average highest fecundity was documented 36324±6438 eggs/fish in the May 2022 followed by 32410±7790 eggs/fish in June2022 (Table 7).

Table 7. Month-wise TL, BW, GW, GSI and Fecundity of *P. paradiseus* in the south-western coastal region of Bangladesh.

Month	Sex (n)	TL (cm) Mean ± SD	BW (g) Mean ± SD	GW (g) Mean ± SD	GSI Mean ± SD	Fecundity Mean ± SD (n)
November	F (10)	18.14±1.80	34.35± 0.17	0.14±0.11	0.45± 0.29	8129±1565
December	F (10)	20.82±2.14	61.16±17.34	0.22±0.11	0.34±0.11	
January	F (15)	20.31±1.96	53.70±16.92	0.15± 0.06	0.28±0.07	
February	F (11)	20.41±2.41	58.72±21.45	0.19±0.09	0.33±0.07	-
March	F (17)	21.30±1.20	69.12±14.58	0.49±0.55	0.76±0.94	17048±7643
April	F (12)	17.22±1.46	28.81±9.37	1.32±1.57	4.48±4.25	18571±5187
May	F (11)	21.41±3.43	67.72±23.35	7.29±2.22	10.88±2.07	36324±6438
June	F (12)	21.2±1.90	73.22±22.03	8.12±2.57	11.10±0.91	32410±7790
July	F (14)	17.8±2.07	34.24±16.61	2.01±2.19	5.08±2.91	14834±8308

Note. n = number of fish; TL = Total Length; BW = Body Weight; GW = Gonad weight

However, year-round sample collection will be continued for the analysis of GSI and histological analysis of gonad maturation stages to find out the peak breeding seasons of the studied fish species.

A total of 8, 5, 10, 12 and 17 gonad samples of Bhangon, Royna, Kain Magur, Bele and Taposi fish, respectively have been sent for histological analysis. Meanwhile, some more samples of each species have already been preserved. However, histological analysis of gonad samples of the fish species is under processing. In the meantime, histological analysis of gonad of Taposi fish has been completed and the results have been summarized in bellow.

Histological observation of female gonad of *Polynemus paradiseus*

The development of oocytes can be divided into different developmental stages. The findings of this study illustrated that oocytes were developed in a synchronized manner. Paired ovaries with matured oocytes were observed at the mature stage. In the present study, month-wise gonadal developmental stages were observed histologically in female *Polynemus paradiseus*.

i. Chromatin Nuclear Stage (CNS)

This is a preliminary stage containing chromatin threads. It is identified by the youngest and undeveloped oocytes (UO). These oocytes are rarely seen in maturity. Besides other stages, the chromatin nuclear stage was ascertained during November-December (Figure 2.a)

ii. Early Perinucleolar Stage (EPNS)

The real development of the oocyte begins with this stage. At this stage, along with oocyte growth, the nucleus starts to enlarge and numerous nucleoli are found around the circumference of the nucleus, which indicates an immature oocyte. This stage was observed in the months of December (Figure 2.b).

iii. Late Perinucleolar Stage (LPNS)

This stage differs from the previous one by the expansion of the oocyte. A great number of nucleoli were apparently viewed throughout the nucleus. A follicular layer was developed around the oocyte. Chorion formation starts at this stage. This stage was mostly spotted in the month of January-February.

iv. Yolk Vesicle Stage (YVS)

This stage was identified by the initial evolvement of yolk vesicles (globules) in the border of the oocytes. They were first shaped as an individual rows that seems colorless when the slides were stained with haematoxylin and eosin. These YV developed as tiny forms but they increased in size and number without ceasing, which indicates a maturing oocyte. This stage was mostly spotted in the month of February-March.

v. Early Yolk Granule Stage (EYGS)

The final oocyte maturation was marked by the formation of yolk granules in oocytes, with completely developed yolk vesicles. They were stained in light pink with haematoxylin and eosin. Most of the oocytes of this stage were observed in April-May.

vi. Late Yolk Granule Stage (LYGS)

With the advancement of yolk granule stage, both the diameter of the oocytes and the number of yolk granules augmented sharply, and oil droplets appeared within the cytoplasm. The yolk granules are densely packed and occupy almost the entire oocyte. The yolk granules were deep pink in color, with haematoxylin and eosin. LYGS was detected mostly during April-May and may be the upcoming month in when the ovary was fully matured.

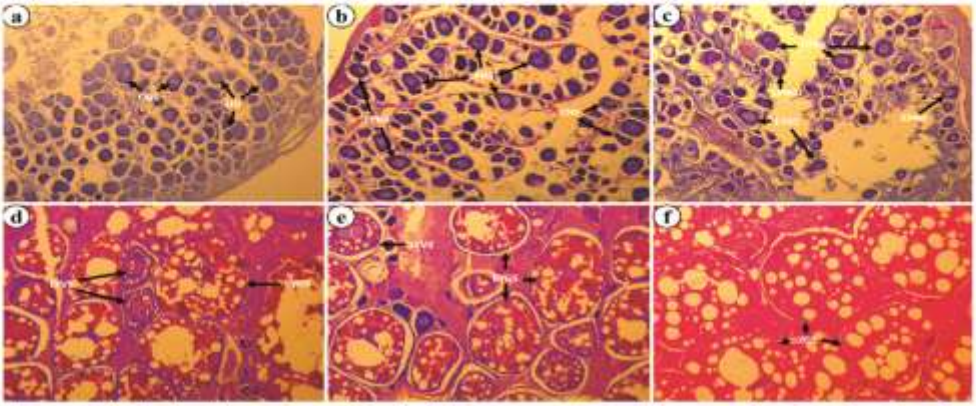


Figure 2. Histological observation of female *Polynemus paradiseus*. Haematoxylin-eosin stained sections of P. paradiseus ovary at 10x magnification. (a) Chromatin Nuclear Stage (CNS); (b) Early Perinucleolar Stage (EPNS); (c) Late Perinucleolar Stage (LPNS); (d) Late Yolk Vesicle Stage (LYVS), Early Yolk Granule Stage (EYGS); (e) Late Yolk Vesicle Stage (LYVS), Early Yolk Granule Stage (EYGS); (f) Late Yolk Granule Stage (LYGS).

Experiment. 6. Induced breeding of Bhangon, Royna, Kain Magur, Bele and Taposi

Preliminary success was achieved for Royna and Kain Magur breeding. The larvae of Royna survived for 10 days (Figure 3) whereas the eggs of Kain Magur partially fertilized and then stopped the progress after 12 hrs (Figure 4). However, complete success will be achieved in near future using the present experience as the hormone, hormonal dose and latency period has been optimized.

Figure 3. Preliminary success on Royna/Rekha breeding