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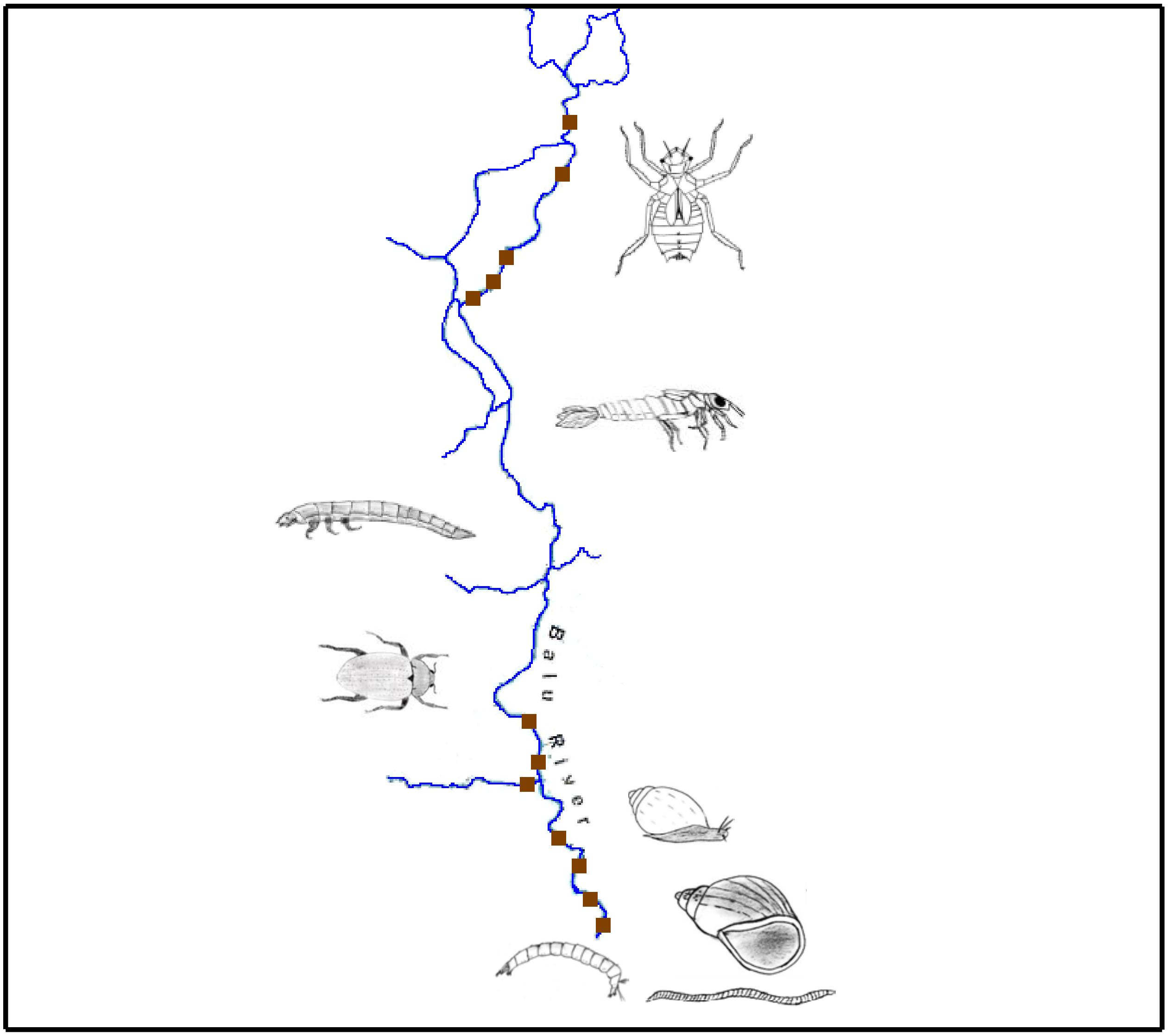

The Use of Macroinvertebrates in Water Quality Monitoring: A Preliminary Study on the Bioassessment Methods for Watercourses in Bangladesh

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Master of Science Thesis
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THIS WORK IS DEDICATED TO
MY BELOVED MOTHER, MOMTAZ BEGEUM

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ABSTRACT

Biological assessment methods based on macroinvertebrates, in conjunction with traditional physical and chemical methods have been widely used as an integral part of water quality monitoring in many countries. Biological assessment of the health of biotic communities in receiving waters offers several important advantages over chemical-based approaches. Because organisms integrate environmental conditions over a long period of time as they are constantly exposed to their ambient environment, where as physical and chemical analysis is instantaneous in time when the sample was collected. At the same time, biological assessment methods often are cheaper than chemical methods. Water quality monitoring in Bangladesh is limited to physicochemical analysis. Therefore, application of biological assessment method for river water quality is still a new field in Bangladesh.

The study area chosen for this research project is located in the greater Dhaka district where the Balu river flows from north to south towards the Shitalakshya river. Macroinvertebrates were sampled from different sites of the river ranging from relatively unpolluted to grossly polluted sites during the period of October to December 2005 and at the same time associated physicochemical parameters of the water quality were measured. Macroinvertebrates were identified up to family level with little exception. Taxa richness, diversity and biotic indices were calculated and evaluated with respect to physicochemical water quality variables, using graphical methods.

The results show that biological assessment based on macroinvertebrate communities provides useful information about water quality by displaying response to pollution. Of the five indices tested four found to be promising namely, Taxa richness, BMWP, BMWP-ASPT and Community Loss Index to use in Bangladesh. These four indices performed well in the assessment of the study sites and the description of the pollution gradient adequately. However, local modifications e.g. inclusion of local taxa and modification of the sensitivity scores of the indicator organisms are necessary to improve the performance of the biotic indices.

Multiple indices have been recommended for assessment of organic pollution status of the rivers in Bangladesh using macroinvertebrates. These indices appear to be suitable indicators for assessing the progress towards achievement of Millennium Development Goal 7: Target 9 in protection and restoration of freshwater ecosystems in Bangladesh

Keywords: Water quality assessment, water quality monitoring, macroinvertebrates, biological assessment, taxa richness, diversity, biotic indices, Millennium Development Goal.

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LIST OF ABBREVIATIONS

Abbreviation	Full meaning
ASPT	Average Score per Taxon
BMWP	Biological Monitoring Working Parties
BOD_5	Biochemical Oxygen Demand
CLI	Community Loss Index
DO	Dissolved Oxygen
EC	Electrical Conductivity
EPT	Ephemeroptera, Plecoptera and Trichoptera
MA	Millennium Ecosystem Assessment
MDG	Millennium Development Goal
NH_4-N	Nitrogen as ammonium
NO_3-N	Nitrogen as nitrate
PCA	Principal Component Analysis
PO_4-P	Phosphorus as orthophosphate
TV	Tolerance Value

1. INTRODUCTION

1.1 GENERAL

The environment, economic growth and development of Bangladesh are all highly influenced by water - its regional and seasonal availability, and the quality of surface and groundwater. Spatial and seasonal availability of surface and groundwater is highly responsive to the monsoon climate and physiography of the country. In terms of quality, the surface water of the country is unprotected from untreated industrial effluents and municipal wastewater, runoff pollution from chemical fertilizers and pesticides, and oil spillage in the coastal area from the operation of sea and river ports (UNEP, 2001).

The Government of Bangladesh has been implementing the National Water Management Plan, starting from 2000 for the next 25 years, with the intention of operationalising the directives given by the National Water Policy, 1999. One of the core objectives of this plan is: 'clean water in sufficient quantities for multi-purpose use and preservation of the aquatic and water dependent ecosystems'. In order to achieve this objective, a national water quality monitoring programme has been identified as one of the main elements of measures for environmental protection and improvement.

Routine surface water quality monitoring in Bangladesh is limited to physicochemical analysis. The Department of Environment as a regulatory body of the Government of Bangladesh has been collecting data on surface water quality since 1980, and currently, it is monitoring water quality at 69 stations for different physico-chemical parameters of water. But, this monitoring program lacks clear objectives and methodologies.

Biological assessment and monitoring is rapidly emerging as a standard river quality assessment tool to be used in conjunction with more traditional physical and chemical methods. In order to achieve and maintain the highest water quality in lakes, rivers, and streams, environmental experts are using the resident organisms living in these waters as sensitive indicators of change. Among the aquatic organisms, macroinvertebrates are the most preferred to develop biological assessment methods (Metcalfe, 1989). The presence or absence of benthic macroinvertebrates has been shown to be a good indicator of both chronic and episodic impacts of human disturbance to river conditions (Rosenberg & Resh, 1993).

Over the past 40 years numerous bioassessment methods have been developed viz., indicator organisms, diversity indices, similarity indices, multimetric methods, biotic indices, saprobic indices etc. Most of the methods have been developed for western countries. Some methods were tested in different regions from where they were developed and promising results were obtained (Armitage *et al*., 1983). Considering the important role of biological methods in water quality monitoring, a study is designed to evaluate several bioassessment methods with respect to river water pollution in Bangladesh.

1.2 BACKGROUND

Biomonitoring based on community structure of freshwater organisms can be traced back to the pioneering work of two German scientists, R. Kolkwitz and M. Marsson, in the early 1900s, who have provided a scientific and practical method for classifying the impacts of organic pollution of running water by combining chemical and biological aspects (Knoben *et al.,* 1995). With the development of saprobic index and indicator organisms of saprobity, the saprobic system was, and is up till now, applied in many European countries.

In Europe, the first development in the use of benthic communities for water quality assessment, apart from the saprobic system, arose in the UK and was first presented in the Trent Biotic Index. Since the late seventies, biotic, saprobic and diversity index methods based on macroinvertebrates have been in use in some western European countries and afterwards many other countries followed suit (Knoben *et al.*, 1995).

RIVPACS is a system for biological assessment of rivers that has been used in the UK since 1990. It permits the prediction of fauna using various combinations of environmental variables, which then allows a direct comparison with actual biological assemblages from the site. Before that time, biological surveillance of the UK Rivers was performed by means of the BMWP score (Biological Monitoring Working Party) and ASPT (Average Score Per Taxon), which can be considered members of the group of biotic indices and biotic scores (Armitage *et al.*, 1983).

While the multivariate prediction method has been developed in the UK, continuation and development of indices or metrics has occurred in the USA. The application of variety of selected metrics has led to the development of multimetric approach where numerous metrics are applied to benthic macroinvertebrate data to assess the status of the community in response to some disturbance. The United States Environmental Protection Agency (USEPA) has adopted a mutimetric approach in the Rapid Bioassessment Protocols (Plafkin *et al.*, 1989).

In recent years there has been an upsurge of interest for the biological monitoring of water quality in several developing countries. Some techniques for bioassessment emphasize a low cost approach, which makes them immediately attractive for use in developing countries, and other features enhance this suitability (important features have been highlighted in chapter 2).

In India, the Central Pollution Control Board (CPCB) has derived Biological Water Quality Criteria (BWQC) for water quality evaluation with the objective of assessing the actual health of water bodies. This system is based on the range of saprobic values and diversity of the benthic macroinvertebrate families with respect to water quality.

The taxonomy of benthic macroinvertebrates in tropical ecosystems is identified as an area requiring further work. As a first step towards development of a biological water quality assessment system, developing countries can adopt family level identification of benthic macroinvertebrates and at the same time should evaluate the available biological assessment methods. To be incorporated into such a system, assessment methods must accurately and reliably reflect environmental degradation (Thorne & Williams, 1997).

This study was carried out in the context of achieving the Millennium Development Goal 7 that aims to ‘ensure environmental sustainability’, and its Target 9, which calls for ‘integrating the principles of sustainable development into country policies and programs and reversing the loss of environmental resources’.

1.3 PROBLEM STATEMENT

Bangladesh is the lower riparian of three major river systems, the *Ganges-Padma*, the *Brahmaputra-Jamuna* and the *Meghna.* These lowland floodplain rivers are characterised by relatively deep, fine sediment (mud, silt, sand) bottom, flat slopes, substantial meandering, susceptible to erosion, and channel shifting.

Most of the bioassessment techniques have been developed and applied in temperate countries for their streams and rivers dominated by cobble or gravel substrates. But some of those assessment methods have been found also to be suitable for tropical lowland rivers.

There are two fundamental problems in applying bioassessment techniques to assess water quality using macroinvertebrates in Bangladesh. First, we know very little about the natural assemblages of invertebrates that occur in running water habitats and how they respond to changes in the environment. Second, no study about suitable methods for assessing water quality using macroinvertebrates in Bangladesh has been reported. This represents a serious gap in the knowledge required for the development of biological monitoring for the surface water quality of the rivers in Bangladesh.

1.4 RESEARCH OBJECTIVES

The overall objective of this research is to investigate the possibility of using macroinvertebrates as a bio-indicator for assessment of organic pollution status of the rivers in Bangladesh and to relate the bioassessment programme to the achievement of Millennium Development Goals in Bangladesh. The specific objectives of this research are:

1. To present a preliminary analysis of the macroinvertebrate communities along the Balu river system in Bangladesh.
2. To verify the applicability of some selected bio-assessment methods for assessing the organic pollution status of the Balu river.
3. To make recommendations for the biological water quality assessment methods which are potentially suitable for the lowland river systems in Bangladesh.
4. To relate water quality assessment and monitoring programme to the attainment of the seventh millennium development goal: ‘Ensure environmental Sustainability’ and its Target 9: “Integrating the principles of sustainable development into country policies and programs and reversing the loss of environmental resources” in Bangladesh.

In order to reach these objectives, the following research questions have been formulated for this research project:

- What are the assemblages of macroinvertebrates occurring in the different pollution gradients of the Balu river just after the spot where it runs through the swamsp of Beel Belai and to one just before it joins the Shitalakshya River near Demra?
- What are the possibilities to use taxa richness, biotic systems, diversity and similarity/loss methods of bioassessment to evaluate organic pollution in the Balu river?
- What are the suitable biological water quality assessment methods that can be used for evaluation of organic pollution for lowland river systems in Bangladesh?
- How can Bangladesh use the biological water quality information to achieve the MDG Target 9 in the protection and restoration of freshwater ecosystems?

2. REVIEW OF LITERATURE

2.1 DEFINITION RELATED TO WATER QUALITY

"Water quality" is a term used to express the suitability of water to sustain various uses or processes. Any particular use will have certain requirements for the physical, chemical or biological characteristics of water; for example limits on the concentrations of toxic substances for drinking water use, or restrictions on temperature and pH ranges for water supporting invertebrate communities. Consequently, water quality can be defined by a range of variables, which limit water use (Chapman & Jackson, 1996).

There is increasing recognition that natural ecosystems have a legitimate place in the consideration of options for water quality management. This is both for their intrinsic value and because they are sensitive indicators of changes or deterioration in overall water quality, providing a useful addition to physical, chemical and other information (Chapman & Jackson, 1996).

2.2 GENERAL CONCEPT OF WATER QUALITY MONITORING

There are several definitions of monitoring (GESAMP, 1980); one of the useful definitions given by GESAMP (1980) is: "Monitoring is the process of repetitive observing for defined purposes on one or more elements of the environment according to prearranged schedules in space and time and using comparable methodologies for environmental sensing and data collection". Monitoring provides factual information concerning the present state and the past trends in environmental behaviour (GESAMP, 1980).

The Arizona Department of Environmental Quality (ADEQ, 2005) defines water quality monitoring as "an integrated activity for evaluating the physical, chemical and biological character of water ecosystems in relation to human health, ecological conditions and designated uses". Although there are many definitions of water quality monitoring, the main reason for monitoring water quality has been, traditionally, the need to verify whether the observed water quality is suitable for intended uses. However, monitoring has also evolved to determine trends in the quality of the aquatic environment and how the environment is affected by the release of contaminants, by other human activities, and/or by waste treatment operations (Meybeck *et al*., 1996).

Assessment is an integral part of monitoring. Water quality assessment is the overall process of evaluation of the physical, chemical and biological nature of the water, whereas water quality monitoring is the collection of the relevant information (Meybeck *et al*., 1992).

Survey, surveillance and monitoring are types of environmental observation programmes, which are often not distinguished from one another and sometimes all three may be referred to as monitoring. General definitions of the above stated programmes have been proposed (Meybeck *et al*., 1992) which may also be modified and interpreted for the aquatic environment as follows:

Term	Definition
MONITORING	Long-term, standardised measurement and observation of the aquatic environment in order to define status and trends.
SURVEY	A finite duration, intensive programme to measure and observe the quality of the aquatic environment for a specific purpose.
SURVEILLANCE	Continuous, specific measurement and observation for the purpose of water quality management and operational activities.

The major types of monitoring methods used in water quality monitoring are hydrological, physico-chemical and biological monitoring. Traditionally, pollution control agencies all over the world solely relied on chemical and physical approach to regulate discharges of toxic pollutants. This process involved setting limits to loads and concentrations of a limited number of priority pollutants in the discharges to water systems, based on their potential toxicity. Signaling and control was established with monitoring activities on chemical concentrations in surface waters and in effluents (de Zwart & Kramer, undated).

Other facet of chemical assessment is that it does not provide direct information of the effects on the biological quality or ecosystem health of water bodies (Knoben *et al.*, 1995). The ultimate goal of environmental protection for aquatic systems is to warrant the sustenance of the ecosystems as a whole, including flora and fauna. For management point of view, it is often focused on human interactions related to public health viz., fishing, swimming, drinking water supply etc. It is therefore, comprehensible that for the prediction and detection of ecological effects of pollution inputs to receiving water bodies, biological monitoring methods need to be developed to compliment the physico-chemical monitoring methodologies (de Zwart & Kramer, undated).

2.3 BIOLOGICAL MONITORING

According to Rosenberg (1998), biological monitoring is the systematic use of living organisms or their responses to determine the quality of the environment. Biomonitoring encompasses two different aspects: the first provides a measure of ecological quality and the second uses the response of biota to changes in water quality as method of assessing such changes (Calow & Pettes, 1994).

Direct biological assessment of the health of biotic communities in receiving waters offers several important advantages over chemical-based approach and some of them are as follows:

- Organisms integrate environmental conditions over a long period of time as they are constantly exposed to their ambient environment, where as physical and chemical analysis is instantaneous in time when the sample was collected (Chapman & Jackson, 1996).
- Biological communities also integrate the effects of multiple stresses and demonstrate cumulative impact (Plafkin *et al.*, 1989).
- It is recognized that not all impacts are chemical in nature; biological assessment may also be able to detect the impact of flow alterations, habitat destruction, overharvesting of biological resources etc. (Karr, 1981).

- Many biological approaches can be cheaper than chemical methods in terms of equipment, but would normally place heavy demands on field and laboratory personnel.
- It has been observed that the public in general has the tendency to like biomonitoring better than the chemical approach. Because of the possible effects being observed in biota are closer to human living and feeling. This aspect is very important in terms of the impact of environmental management (de Zwart & Kramer, undated).

Water pollution is essentially a biological problem. Chemical measurements are like taking snapshots of the ecosystem, whereas biological measurements are like making videotape. The ultimate purpose of environmental assessment and regulation is the maintenance of biological integrity, so setting water- and sediment- quality objectives should involve biological as well as chemical criteria (Mandaville, 2002).

Biomonitoring has some disadvantages as well. It is sometimes extremely difficult to relate an observed effect to specific aspects of pollution through biological effect measurements. The response of organisms may be affected by their natural cycles, such as life stage and reproductive condition. Consequently, like other techniques, biomonitoring methods should be developed and interpreted by experienced biologists (Chapman & Jackson, 1996).

The response of biological communities or of the individual organism can be monitored in a variety of ways to indicate effects on the ecosystem. According to Chapman & Jackson (1996), the biological assessment methods can be divided into five major categories:

- Ecological methods: These methods include information on species composition, density, diversity, availability of indicator species etc.
- Physiological and biochemical methods: These methods are based on community metabolism (such as oxygen production or consumption, growth rates) or biochemical effects in individuals or communities (such as enzyme inhibition).
- Controlled biotests: Measuring toxic effects (death, growth rates, reproductive capacity) on organisms under defined laboratory conditions or the effects on behaviour *in situ* or in controlled environments.
- Contaminants in biological tissues: This type of assessment is based on measurements of the accumulation of specific contaminates in the tissues of organism living in the environment (passive monitoring) or deliberately exposed in the environment (active monitoring).
- Histological and morphological methods: The observation of cellular changes or morphological changes such as gill damage or skin lesions in fish.

2.4 BENTHIC MACROINVERTEBRATES IN WATER QUALITY MONITORING

Macroinvertebrates are a diverse array of animals without backbones operationally defined as those that are retained by a sieve or mesh with pore size 0.2-0.5 mm, as used most frequently in stream sampling devices. Stream macroinvertebrates include various groups of worms (flatworm, eelworms, segmented roundworms), molluscs (snails and bivalves), crustaceans (shrimps, crayfish, other shrimp-like group), mites, and above all, insects. Especially important among the latter are the larval stages of mayflies (Ephemeroptera), stoneflies (Plecoptera), caddisflies (Trichoptera), beetles (Coleoptera) and true flies (Diptera) (New Zealand Ministry for Environment, 1999).

The central role of macroinvertebrates in food webs of streams and rivers means they contain information on the energy base of the ecosystem, water quality, habitat diversity, and the availability of appropriate kind of food to support population of native fish and trout. They can be seen as integrators of information on stream ecosystems structure and function as well as water quality (New Zealand Ministry for Environment, 1999).

Benthic macroinvertebrates have been used as indicators of river quality for the past fifty years. There are compelling reasons for the apparent popularity of freshwater macroinvertebrates in current biomonitoring practice; they offer a number of advantages (Mandaville, 2002):

- they are ubiquitous, so they are affected by perturbations in many different habitats,
- they are species rich, so the large number of species produces a range of responses,
- they are sedentary, which allows determination of the spatial extent of a perturbation,
- they are long-lived, which allows temporal changes in abundance and age structure to be followed, and
- they integrate conditions temporally, so like any biotic group, they provide evidence of conditions over long periods of time.

Tropical communities of benthic macroinvertebrates are similar to temperate communities, with many of same families. One major group of aquatic invertebrates, aquatic insects, which are valuable indicator organism, are considered to be no more diverse in tropics than in temperate zone, in contrast to fish and terrestrial insects (Throne & Williams, 1997).

However, macroinvertebrates also have disadvantages, but these can be mostly overcome by proper experimental design. For example, macroinvertebrates do not respond to all impacts; the distribution and abundance of macroinvertebrates may be affected by factors in addition to the perturbation in question; and the distribution and abundance of macroinvertebrates vary seasonally (Mandaville, 2002).

2.4.1 Sampling of benthic macroinvertebrates

There are three general guidelines for sampling biota given by the International Organization for Standardization (ISO), which are as follows:

1. Guidance on handnet sampling of aquatic benthic macroinvertebrates (ISO 7828:1985).

Hand net or kick-net is used for sampling in shallow water down to a depth of about 1.5 meter. This is qualitative sampling technique that provides data on the presence/absence/ diversity and relative abundance of taxa depending on sampling effort and mesh size.

2. Guidance on the design and use of quantitative samplers for benthic macroinvertebrates on stony substrata in shallow freshwaters (ISO 8265:1988).

This a guidance on equipment and procedures for the quantitative sampling of benthic macroinvertebrates in shallow waters of depth less than 0.5 m up to 1 m and velocity higher than 0.1 m/s. These samplers are know as quadrate samplers in which the bed of the water body is isolated and the disturbance of the substrate leads to release the invertebrates into the net by the water current.

3. Sampling in deep waters for macroinvertebrates – Guidance on the use of colonization, qualitative and quantitative samples (ISO 9391:1993).

Colonization samplers are used for rivers of depth greater than one meter. They are designed to provide a collection of macroinvertebrates capable of living and colonizing in artificial substrates and therefore, reflecting directly the water quality of the sample site.

2.4.2 Approaches used to assess the water quality using macroinvertebrates

- **The saprobic system**

The saprobic systems are based upon the observation that species composition as well as species numbers are different over a gradient of self-purification after organic inputs, ranging from complete oxidation to predominance of reduction processes. As a result, a zonation in the aquatic communities can be distinguished reflecting the degree of saprobity. Every species has a specific tolerance to decomposing organic substances and thus, the oxygen content, which is expressed in a saprobic indicator. The saprobic index is numerical evaluation of the presence of indicator species and their respective saprobic values (Knoben *et al.*, 1995).

- **Biotic indices and biotic scores**

Biotic indices and biotic scores combine features of both the diversity approach as well as the saprobic approach. The biotic indices are based on two principles:

a) Macroinvertebrate groups Plecoptera (stoneflies), Ephemeroptera (mayflies), Trichoptera (caddisflies), Gammarus, Aselus, red Chironomids and Tubificidae disappear in the order mentioned as pollution increases;

b) The number of taxonomic groups is reduced as organic pollution increases.

A biotic index is a qualitative measure whereas most biotic scores include a measure of abundance and thus are semi-quantitative (Knoben *et al.*, 1995).

- **Diversity indices**

Diversity is a basic feature of the structure of a community or ecosystem. The basic assumption is that disturbance of the water ecosystem or communities under stress leads to a reduction in diversity. The extent of reduction to the diversity depends on the degree of pollution. This is not necessarily always true, as mild pollution can be associated with an increase in diversity, before the characteristic decline with more severe pollution as described by Pinder & Farr (1987; cited in Throne & Williams, 1997).

Diversity indices generally require a species or taxa number component (not necessarily identified) and an evenness component (or a component that measures the abundance of taxa), and are thus an index of community structure not function. The most widely used diversity index is the Shannon-Wiener index, because it is stable in any spatial distribution and insensitive to rare species (Calow & Petts, 1994). Its formula is as follows:

$$H= -\sum(N_i / N)*\ln(N_i/N)$$

Where H= index value, N= total number of individuals of all species collected, and N_i = number of individual belonging to the i-th species.

The sequential comparison index (SCI) is a kind of diversity index developed to fill the need for a rapid numerical method of assessing the biological consequences of pollution. The sequential comparison index (SCI) of Cairns *et al.* (1971) is particularly attractive for use in developing countries as it requires no formal taxonomic expertise and is based on the "sign test" and the "theory of runs". That is, it relies on the innate ability of the user to recognize differences in size, shape and colour (signs) of organisms. It is an expression of community structure since it depends upon both the species richness of the community and on the distribution of individuals among the species. Only two individuals are compared at a time. The current individual need only be compared to the previous one. If it looks similar it is part of the same "run"; if not, it is part of a new run. Differences in the shape, color and size of the organisms are sequentially encountered in a sample (Mackie, 1998).

The formula of the sequential diversity index (DI) is as follows (de Zwart & Trivedi, 1992):

$$DI= \text{No. of runs}/ \text{Total no. of organisms}$$

Where a run is a set of organisms that looks similar

- **Richness**

Richness measures are extremely simple and quick to calculate and have been identified as effective rapid assessment metrics for use with macroinvertebrates (Resh & Jackson, 1993). For simplification, both the total number of families and the number of families belonging to the order Ephemeroptera, Plecoptera and Trichoptera (EPT richness) are evaluated. These three orders are generally considered to be sensitive to organic pollution.

- **Rapid Bioassessment Protocol (RBP)**

Rapid Bioassessment Protocol combines the assessment of the biological condition or quality with the assessment of habitat quality. This implies that the method can be considered as an ecological assessment method (Plafkin *et al.*, 1989). RBP IV consists of a certain number of diversity indices and a number of comparative indices, called metrics. These metrics assess the biological condition of the benthic community, divided in three categories: structure, community balance and functional feeding group. The basic principal of the method is the comparison with a reference site or a set of reference data. Here the reference site is a mean situation of a set of unaffected sites (Knoben *et al.*, 1995). Following is the some metrics used in RBP.

<u>Structure metrics:</u>

1. Taxa Richness = total number of taxa collected at the site.
2. EPT Index = number of genera belonging to the Orders Ephemeroptera, Plecoptera, and Trichoptera which were collected.
3. Community Loss Index = (taxa richness at the reference site - taxa common to reference and sampling sites)/taxa richness at the sampling site.

<u>Community balance metrics</u>

4. Percent Contribution of Dominant Taxa = percent contribution of the dominant taxa to the total number of organisms collected.

5.Family Biotic Index (modified) = S (xi ti)/n, where:

xi = number of individuals within a genus,

ti = tolerance value for the taxa,

n = total number of organisms in the sample.

<u>Functional feeding group metrics:</u>

6. Ratio of Scrapers/Filtering Collectors = number of scrapers/(number of scrapers + number of filtering collectors).
7. Quantitative Similarity Index = (Functional Feeding Groups or FFG's) - compares two communities in terms of presence or absence of FFG's, also taking relative abundance into account.

- **River invertebrate Prediction and Classification System (RIVPACS)**

RIVPACS has been used in nationwide biological assessment of rivers in United Kingdom in 1990. RIVPACS uses the physical, geographical and chemical characteristics of a monitoring site to predict what the natural macroinvertebrate fauna of that site would be in the absence of environmental stress. Comparison of the predicted macroinvertebrate communities with those observed during the biological sampling and analytical programme allows the calculation of ecological quality indices (EQIs). Based on RIVPACS methodology, Australian RIVPACS (AusRivAs) for rivers was developed as part of the Monitoring River Health Initiative (RHI) in Australia.

- **Biological Monitoring Working Party (BMWP) Score System**

Biological Monitoring Working Party score (BMWP) is a biotic score system which uses identification of organism to family level. This system reduces the effort and taxonomic expertise necessary for routine biological assessments based on indicator organisms. The Biological Monitoring Working Party score (BMWP) has been standardised by the International Organization for Standardization (ISO). It can be used to reflect the impact of organic pollution, such as results from sewage disposal or farm waste. This score was devised in the UK but was not specific to any single river catchment or geographical area. This scoring system was tried in tropical rivers and was found quite suitable (de Zwart and Trivedi, 1992; Thorne & Willams, 1997)

In this method, invertebrates are collected from different habitats (e.g. gravel, silt, weed beds) at representative sites of river stretches. The organisms are identified to the family level and then each family is allocated a score between one and ten. The most sensitive organisms, such as mayfly nymphs score ten, molluscs score three and the least sensitive worms score one (Table 2.1).

The BMWP score is calculated by summing the scores for each family represented in the sample. The number of taxa gives an indication of the diversity of the community (high diversity usually indicates a healthy environment) (Friedrich *et al.*, 1996).

A weakness of the BMWP system, in common with many other score systems, is the effect of sampling effort. A prolonged sampling period can be expected, under most circumstance, to produce a higher final score than a sample taken quickly. To overcome this inherent weakness of the BMWP system, it became common practice to calculate the Average Score Per Taxa (ASPT) by dividing the BMWP Score by the number of taxa.

Table 2.1 The biological scores allocated to groups of organisms by the Biological Monitoring Working Party (BMWP) score (Chapman & Jackson, 1996; Friedrich *et al.*, 1996)

Common Name	**Family**	**Score**
Mayflies Stoneflies Bugs Caddisflies	Siphlonuridae, Heptageniidae, Leptophlebiidae, Ephemerellidae, Potamanthidae, Ephemeridae. Taeniopterygidae, Leuctridae, Capniidae, Perlodidae, Perlidae, Chloroperlidae Aphelocheiridae Phryganeidae, Molannidae, Beraeidae, Odontoceridae, Leptoceridae, Goeridae, Lepidostomatidae, Brachycentridae, Sericostomatidae	10
Dragonflies Damselflies Caddisflies Crustaceans	Gomphidae, Cordulegasteridae, Aeshnidae, Corduliidae, Libellulidae Lestidae, Agriidae Psychomyiidae (Ecnomidae), Phylopotamidae Astacidae	8
Mayflies Stoneflies Caddisflies	Caenidae Nemouridae Rhyacophilidae (Glossosomatidae), Polycentropodidae, Limnephilidae	7
Snails Mussels Caddisflies Damselflies Crustaceans	Neritidae, Viviparidae Ancylidae (Acroloxidae), Unionidae Hydroptilidae (Crangonyctidae) Coenagriidae, Platycnemididae Gammaridae, Corophiidae	6
Bugs Beetles Caddisflies Trueflies	Hydrometridae, Gerridae, Nepidae, Naucoridae, Notonectidae, Pleidae, Corixidae Haliplidae, Hygrobiidae, Dytiscidae (Noteridae), Gyrinidae, Hydrophilidae (Hydraenidae), Elmidae, Clambidae, Scirtidae, Dryopidae Hydropsychidae Tipulidae, Simuliidae Planariidae (Dogesiidae), Dendrocoelidae	5
Mayflies Alderflies Leeches	Baetidae Sialidae Pisicolidae	4
Snail Cockles Hoglice Leeches	Valvatidae, Hydrobiidae (Bithyniidae), Lymnaeidae, Physidae, Planorbidae Sphaeriidae Asellidae Glossiphoniidae, Hirudinidae, Erpobdellidae,	3
Midges	Chironomidae	2
Worms	Oligochaeta (Whole class)	1

2.5 WATER QUALITY AND THE MILLENNIUM DEVELOPMENT GOALS

At the Millennium Summit in September 2000 the states of the United Nations reaffirmed their commitment to working toward a world in which sustaining development and eliminating poverty would have the highest priority. The Millennium Development Goals grew out of the Millennium Declaration adopted by the United Nations in 2000. The goals have been commonly accepted as a framework for measuring development progress.

The eight Millennium Development Goals (MDGs) lay out a comprehensive set of outcome targets for sustainable development derived from the recognition that the reduction of poverty in all its forms requires not only a genuine partnership between rich and poor countries, but also integrated responses that address the interrelated challenges of poverty, hunger, disease, gender inequality, lack of education, and environmental degradation. The MDGs are national goals that need to be concretized, operationalized, and achieved in each country.

The eight Millennium Development Goals:

Goal 1 Eradicate extreme poverty and hunger
Goal 2 Achieve universal primary education
Goal 3 Promote gender equality and empower women
Goal 4 Reduce child mortality
Goal 5 Improve maternal health
Goal 6 Combat HIV/AIDS, malaria and other diseases
Goal 7 Ensure environmental sustainability
Goal 8 Develop a global partnership for development

Table 2.4 Targets of the Millennium Development Goal no.7

Goal 7. Ensure environmental sustainability	
Target 9	Integrate the principles of sustainable development into country policies and programmes and reverse the loss of environmental resources
Target 10	Halve, by 2015, the proportion of people without sustainable access to safe drinking water and sanitation
Target 11	By 2020, to have achieved a significant improvement in the lives of at least 100 million slum dwellers

The seventh Millennium Development Goal aims to ensure environmental sustainability, with its first target being the integration of principles of sustainable development into country policies and programmes and the reversal of the loss of environmental resources. With respect to water resources development and management, it has immediate repercussions for the MDG targets relating to the reduction of poverty and hunger, the promotion of gender equality and the empowerment of women, the reduction of child mortality, combating key communicable diseases, better access to safe drinking water as has been outlined in the World Summit on Sustainable Development held in Johannesburg, 2003 (WHO, 2005).

3. RESEARCH METHODOLOGY

3.1 STUDY AREA

For this study, sampling was carried out in the Balu River in Bangladesh (Fig. 3.1). This river runs mainly through the extensive swamps of Belai Beel and those east of Dhaka, joining the Shitalakshya River near Demraghat in the downstream. In the upstream, it has a narrow connection with the Shitalakshya through the Suti Nadi and also by way of the Tongi Khal with the Turag River that passes through the northwest of Dhaka. Although it carries floodwater from the Shitalakshya and the Turag during the flood season, the Balu is of importance mainly for local drainage and access by boats.

3.1.1 Climate

Bangladesh has a subtropical monsoon climate characterized by wide seasonal variations in rainfall, moderately warm temperatures, and high humidity. Regional climatic differences in this flat country are minor. Three seasons are generally recognized: a hot, humid summer from March to June; a cool, rainy monsoon season from June to October; and a cool, dry winter from October to March. In summer, temperature ranges between 34°C and 21°C (maximum and minimum average). In winter, average maximum and minimum temperature are 29°C and 11°C, respectively.

The single most dominant element of the climate of Bangladesh is the rainfall. Because of the country's location in the tropical monsoon region, the amount of rainfall is very high. However, there is a distinct seasonal pattern in the annual cycle of rainfall. The winter season is very dry, and accounts for only 2-4% of the total annual rainfall. Rainfall during this season varies from less than 2 cm to slightly over 4 cm. As the winter season progresses into the pre-monsoon hot season, rainfall increases. The amount of rainfall in this season varies from about 20 cm to slightly over 80 cm, which accounts for 10-25% of the total annual rainfall. The monsoon season accounts for 70-85% of annual total rainfall that amount to 100 cm to over 200 cm. Geographic distribution of annual rainfall shows a variation from 150 cm in the west-central part of the country to more than 400 cm in the north-eastern and south-eastern parts.

Floods are more or less a recurring phenomenon in Bangladesh and often have been within tolerable limits, but occasionally they become devastating. The study area suffers from monsoon flood that visits during the period between June and October and inundates vast areas and causes huge losses to lives and properties.

3.1.2 Population and Landuse

Dhaka, the biggest city in Bangladesh, lies on the west bank of the Balu downstream with its urban hinterland. Population density of this area is very high, which is about 7500 persons per square kilometre. The east bank and the upstream have the population density ranging from 700 to 1500 persons per square kilometre (BBS, 1999). The land use is dominated by intensive agriculture-mostly rice cultivation. Extensive vegetable cultivation has been common in the areas proximity to the Dhaka city. Riparian zones are vegetated with wood and fruit trees, shrubs, flood resistance plant species, phragmites etc. From Trimohoni towards downstream areas, riverbank vegetation has gradually disappeared and has been replaced by human settlements (Fig. 3.2).

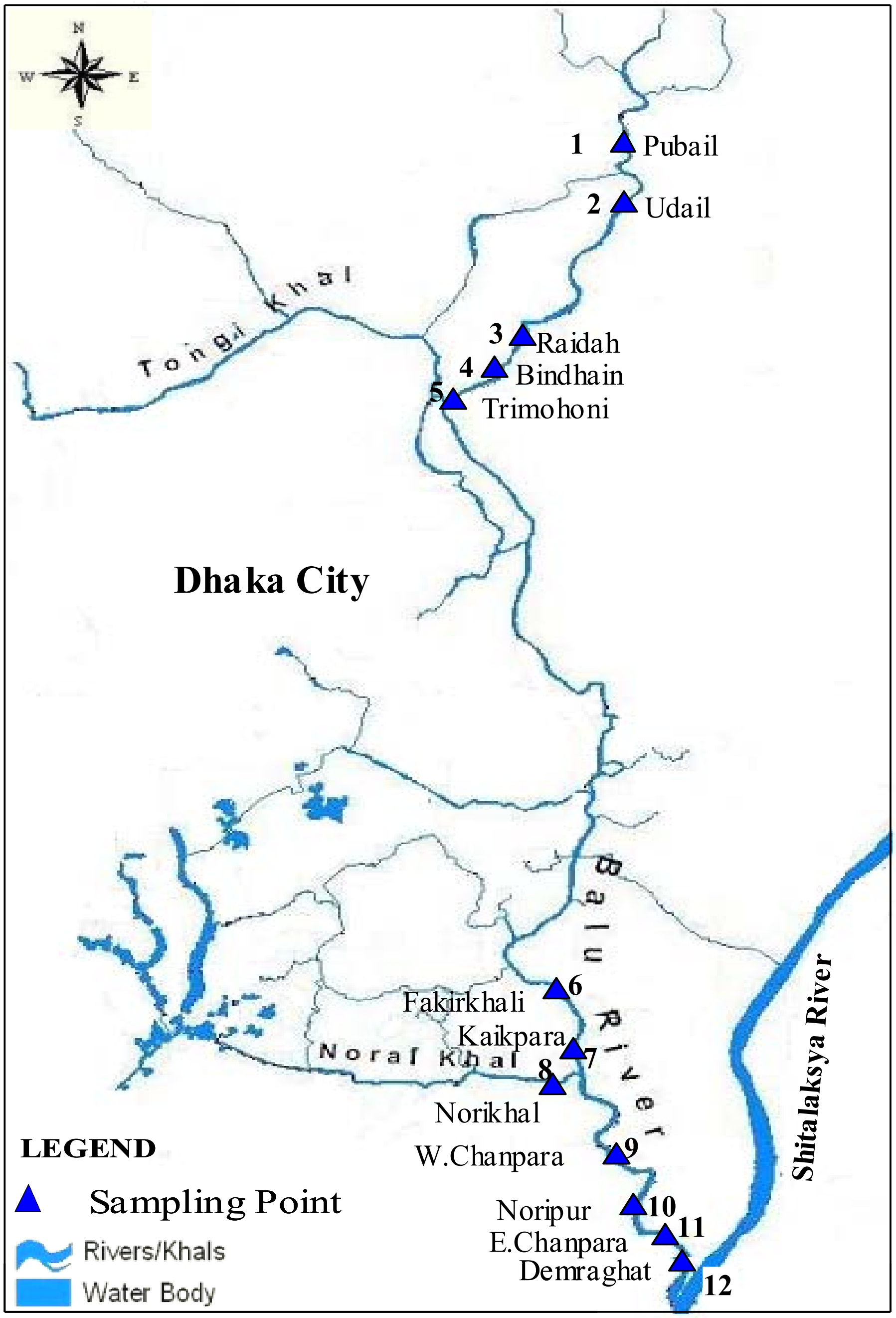
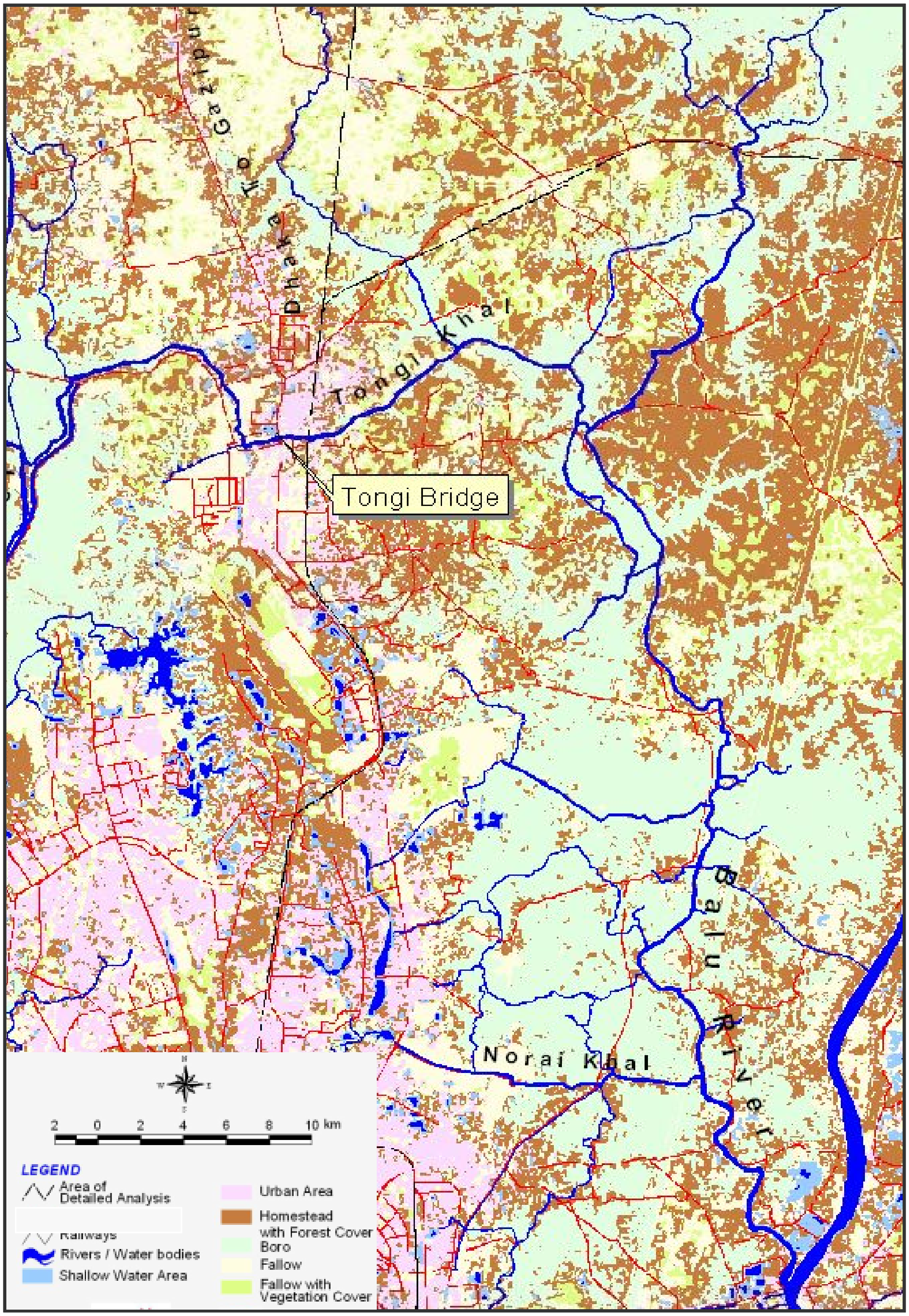


Fig 3.1 Location of the 12 study sites in the River Balu and Norikhal in Bangladesh

Sampling point	Balu River							Tributary to the Balu	Balu River			
	1	2	3	4	5	6	7	8	9	10	11	12
Downstream distance (km)	0	1.5	4.5	6	7	21	22	24	26	28	29	30



Source: Bangladesh Environment Management Project

Fig. 3.2 Land use of the study area in the greater Dhaka District in Bangladesh

3.1.3 Water Pollution

Many canals that originate from swamps/wetlands in the east and southern parts of the Dhaka City receive urban run-off, untreated industrial effluent and sewage while flowing through different areas of the city. They join with each other and finally drain into the Balu River. Although many of these canals have been filled up, few still continue their existence. Norikhal is one of the main canals that discharges huge untreated municipal waste into the River Balu. During the dry time, water of this canal becomes murky and creates a big stink.

Before entering Dhaka City, the Balu River is affected by rural land use run-off containing organic and biodegradable materials. The upper part of the Balu proximity to Beel Belai is relatively unpolluted as habitation and agriculture are limited. The lower reach of the Balu especially after the spot where Norikhal meets with it flows through heavily populated suburb of the Dhaka City and is close to many dwellings. These areas are progressively more polluted by domestic waste, untreated sewage and effluent from a jute mill at Demraghat.

3.2 SAMPLING SITES

The sampling locations were chosen to represent an entire pollution gradient ranging from unimpacted sites to grossly polluted sites. A total of 12 sites were selected in the study area as shown in Fig. 3.1. Site numbers 1-5 are in the upstream of the Balu River and were chosen as they are considered to be unimpacted by the influence of the city. Distance between site number 1 and 5 is 7 km. These sites are located relatively in low populated rural areas in which habitation and agricultural intensity are limited. However, it is unlikely to get sites, which are completely unimpacted by human influences to be used as reference sites, but the least impacted sites are available to be used as a reference sites for the evaluation of other sites (Davis & Simon, 1995).

The most polluted site in the study area is number 8, which is located in the Norikhal, a big canal that carries wastewater from the east of the Dhaka City and drains into the Balu. Site number 6 and 7 are in the Balu River, 3 and 2 km upstream of the confluence with the Norikhal, respectively. These two sites are influenced by domestic pollution in varying degree. Sites 9 –12 are all located within 6 km downstream of the Norikhal. These areas are heavily populated and close to numerous dwellings. These four sites suffer from local pollution as well as effluent discharged through the Norikhal. Site 12 is close to the confluence of the Balu with the River Shitalakshya.

3.3 SAMPLING PERIOD

Sampling was carried out between October and December 2005. These sampling periods in Bangladesh belong to the end of rainy monsoon season (June through October) and beginning of the cool dry winter season (November through February). Samples were taken twice from each site in about one-month interval so that data of wet and dry season are distinguishable. A summary of the physicochemical data for both seasons is given in Appendix 1.

(a)

(b)

(c)

Fig. 3.3 Sampling sites in the River Balu: Trimohoni in dry season (a), Fakirkhali in wet season (b), and riverbank vegetation at Raidah (c).

3.4 PARAMETERS CHOSEN AND MEASUREMENTS

The parameters for water quality assessment were selected in accordance with the objectives of the study programme. Here the main focus is: to evaluate primarily organic pollution of river water using macroinvertebrates. A field data protocol was used for documentation of sampling and analysis (after de Zwart & Trivedi, 1992; Appendix –3).

In addition to water quality parameters, the following environmental variables were considered.

- Habitat variables:
  - Substrate characteristics (examined by eye/finger test): mud, sand, silt, plants, stones etc.
  - Habitat characteristics (estimated by eye): floating macrophyte, submerged macrophyte, bank vegetation, algae etc.
- Field observation of landuse, point and non-point sources of pollution.

Table 3.1 Description of the sampling sites

Site name	Land use	Pollution Source	Bank vegetation	Aquatic vegetation	Substrate type
Pubail	Agriculture, wetland & sparse settlements	Agriculture	Floodplain shrub and herb	Water spinach, Coontail, Fanwort, etc.	Detritus & clay
Udail	Agriculture & sparse settlements	Agriculture	Patch forest, shrub and herb	Hyacinth, reeds, phragmites, etc.	Detritus & clay
Raidah	Agriculture, permanent vegetation & sparse settlements	Agriculture	Patch forest, shrub and herb	Hyacinth, reeds, phragmites, etc.	Detritus & clay
Bindhain	Agriculture, permanent vegetation & sparse settlements	Agriculture	Patch forest, shrub and herb	Hyacinth, reeds, phragmites etc.	Detritus & clay
Trimohoni	Agriculture, barren lowland & sparse settlements	Agriculture	Floodplain shrub and herb	Hyacinth, reeds, hydrilla, etc.	Clay
Fakirkhali	Agriculture, on-the-bank settlements (moderate density)	Agriculture & sewerage from hanging toilet	Homestead plants	Hyacinth, reeds, phragmites, etc.	Detritus & clay
Kaikpara	Agriculture, on-the-bank settlements (moderate density)	Agriculture & sewerage from hanging toilet	Homestead plants	Hyacinth, reeds, phragmites, etc.	Detritus & clay
Norikhal	Agriculture, on-the-bank settlements (moderate density)	Municipal wastewater	Grass	Hyacinth, reeds, phragmites, etc.	Clay
W.Chanpara	Sub-urban (right bank), dense settlement and agriculture (left bank)	Agriculture & sewerage from hanging toilet	Homestead plants	Hyacinth, reeds, spinach, etc.	Detritus & clay
Noripur	Sub-urban (right bank), dense settlement and agriculture (left bank)	Agriculture & sewerage from hanging toilet	Homestead plants	Hyacinth, reeds, etc.	Detritus & clay
E.Chanpara	Sub-urban (right bank), dense settlement	Sewerage from hanging toilet	Homestead plants	Water hyacinth & reeds.	Detritus & clay
Demraghat	Sub-urban	Jute mill & Municipal wastewater	Absent	Water hyacinth & reeds.	Detritus & clay

3.4.1 Hydrological Parameters

A rough estimate of the mean velocity of a sampling site of the river was obtained by measuring the travel time of an orange along a known distance. Water depths was measured by using measuring pole and river width was estimated by eye or measuring the width of the bridge. Together with a measurement of the cross-sectional area, this will provide an estimate of flow of a sampling site (Guinot *et al*., 2005). During the wet season, water depth was very high and river flooded the vast area of the both side of its bank, and therefore, measurement of cross-sectional area was not possible with simple pole or naked eye. Hydrological data were taken during the dry period in two sites.

3.4.2 Physicochemical Parameters

In order to aid the evaluation of the pollution status of the selected study sites, and therefore, the pollution gradient, eight physicochemical parameters were chosen: Temperature, pH, biochemical oxygen demand (BOD_5), ammonium-nitrogen, nitrate-nitrogen, phosphate-phosphorus, electrical conductivity (EC) and dissolved oxygen (DO). These parameters are likely to be influenced by the input of and/or recovery from organic pollution (Thorne & Williams, 1997). Temperature, pH, conductivity and dissolved oxygen was measured *in situ* using portable meters. BOD was determined by subtracting the endpoint oxygen concentration after 5 days confinement in a closed bottle at 20^{o} C from the initial oxygen concentration. The concentration of NH_4-N, NO_3-N and PO_4-P was measured *in situ* using field kit (Dr. Lange's field kit).

3.4.3 Biological Sampling

Samples of benthic macroinvertebrates were taken from each site by using macrofauna net. Considering the high water depth (> 10 m) in the wet season, initially biological sampling was tried out with the Van Veen grab sampler. During the study, it was found that grab sampler is effective to collect fauna from sediment, but ineffective to catch macrofauna attached to plants. Eventually, macrofauna net was used to collect macroinvertebrates from marginal vegetation as well as from bottom sediment.

The number of samples taken from each site was decided by producing a curve relating the number of species encountered with the number of samples taken (de Zwart and Trivedi, 1992). The sampling efforts were continued until the curve became asymptotic.

3.4.4 Identification of Macrofauna

The identification of macroinvertebrates was achieved on site at family level with the use of benthic fauna family identification keys (de Zwart and Trivedi, 1992) and locally available books on freshwater fauna of Bangladesh. But the exceptions were worm and bristle worm; they were identified at class level. Most of the taxa were identified *in situ*, but some of biological samples were preserved in 4% formalin for posterior identification.

3.4.5 Statistical Analysis

Wet and dry season physicochemical data were assessed using a paired *t*-test in order to detect seasonal differences (Results of paired *t*-test is given in Appendix-4). The relationship between biological indices and five important physicochemical parameters (BOD, DO, NH_4-N, PO_4-P and conductivity) was determined by Spearman's correlation coefficient method.

3.4.6 Chemical Water Quality Assessment

The waters in the study area of the Balu River are affected by untreated domestic waste and rural land use run-off containing organic, biodegradable material as well as urban effluents. As a result, the changes in macroinvertebrate communities associated with this complex type of pollution can be caused in many ways.

Although the chemical water quality data were collected in the wet and dry season, the further analysis was carried out using only the dry season data. Because of the dilution of pollutants in the wet season, most of the sites showed very low level of pollution. During this time, variation in chemical parameters among the sites is very small and therefore, the size of differences between the sites was lost in relation to chemical water quality.

In order to compare the pollution status of each site, a simple integrated assessment of the five important parameters (BOD, DO, NH_4-N, PO_4-P and conductivity) was carried out. NO_3-N values were excluded from pollution status assessment for the reason that most of sites showed very low NO_3-N, which was below the measuring limit of the Dr. Lange's field kit.

Every site was ranked for each of the five parameters and an average rank for each site was calculated. For DO, lowest value received the highest rank.

A water quality class was assigned to each site so that sites having similar water quality can be referred easily. These classes ranged from 1 to 4 and reflected unpolluted, slightly polluted, moderately polluted and grossly polluted condition. These chemical classes of quality assessment scheme broadly correspond to the UK Water Council (1981) classes used in Britain (Thorne & Williams, 1997).

3.4.7 Biological Assessment Methods

Of the numerous methods available for biological assessment of water quality on the basis of macroinvertebrates, those potentially suitable for tropical lowland floodplain river were tested for this research program are listed in Table 3.1.

Table 3.2 Methods of biological assessment applied to the Balu River System

System/Method	Index/Score	References
Richness	No. of families	Thorne & Williams, 1997
	EPT richness	
Biotic Systems	BMWP	Thorne & Williams, 1997 de Zwart &Trivedi, 1992
	BMWP-ASPT	
Diversity and similarity/loss	Sequential Comparison Index	de Zwart &Trivedi, 1992
	Community Loss Index	Thorne & Williams, 1997

3.5 EVALUATION OF THE BIOLOGIAL ASSESSMENT METHODS

To evaluate the bioassessment methods against a pollution gradient, simple graphical method was used similar to those described by Thorne and Williams (1997). Average rank of each site calculated from rank of each of five parameters provided a good indication of pollution gradient, as it existed in the field. The response of metrics to environmental degradation was achieved by plotting score/index values against the gradient and by inspecting the plots.

According to Fore *et al.* (1996; as cited in Thorne and Williams, 1997), graphical methods are fundamentally advantageous over statistical techniques with respect to showing in detail about how indices respond; identifying outliers, and illustrating the range over which the indices are most sensitive.

3.6 RECOMMENDATION FOR BIOLOGICAL ASSESSMENT METHODS

On the basis of the following criterion biological assessment methods of water quality for rivers were selected:

- Indices, which separated the four water quality classes to a reasonable degree and showed a clear relationship with the pollution gradient.

  This was manifested in the graph. When index values were plotted against the pollution gradient (mean chemical rank) there was reasonable separation of the most and least disturbed sites and intermediate sites were placed between them.

3.7 DETERMINATION OF TOLERANCE VALUES FOR BANGLADESHI RIVER MACROINVERTEBRATES

An approach based on principal components analysis (PCA) on physicochemical data to represent the pollution gradient was used to develop tolerance values (TVs) for taxa occurring in the Balu River and Norikhal. All the 12 sites were selected for the analysis. The variables included in the PCA were DO, BOD_5, NH_4-N, PO_4-P and electrical conductivity. All five water chemistry variables were transformed with $\log_{10}$ to reduce skewness. The PCA was run using a correlation matrix. The PC1 axis scores were used for TVs calculation (Appendix 2B).

According to USEPA, TVs based on PCA are only developed for taxa with at least 25 observations in each data set (Blocksom & Winters, 2005). As a first step towards the development of TVs for Bangladeshi river macroinvertebrates, the rule for minimum observations limit was not considered as because the maximum observation of any taxa in this study were twelve. Simuliidae family was excluded from the process of developing final tolerance value. Because it's initial TV, which is used to calculate final TV, was found as an outlier. Grubbs' test was used to detect outliers in a univariate data set. The following formula was used for calculating initial TV of a taxon.

$$TV_{initial} = \frac{\sum_i (proportion_i \times PC1_i)}{\sum_i proportion_i} \qquad \text{(Equation 1.1)}$$

Where $proportion_i$ is the mean proportion of a given taxon in sample i and $PC1_i$ is the PC1 axis score at that same site. Then, the initial TV was rescaled to a 0-10 range, with 10 as most tolerant and 0 as least tolerant, using the following formula.

$$TV_{final} = \frac{TV_{init} - TV_{min}}{TV_{max} - TV_{min}} \times 10 \qquad \text{(Equation 1.2)}$$

Where TV_{init} is the initial TV value of a given taxon, calculated by using equation 1.1, TV_{min} is the minimum TV_{init} value across all taxa and TV_{max} is the maximum TV_{init} value across all taxa.

The scale of a 0-10 range was used to measure taxa tolerance where 10 indicates the most tolerant and 0 is the least tolerant taxa.). In order to correspond to BMWP scores, TVs were again rescaled to a range from 1-10 where most sensitive organisms score ten and least sensitive organisms score one.

4. RESULTS

4.1 PHYSICOCHEMICAL PARAMETERS

The results of the physicochemical analysis in the River Balu and Norikhal are presented in Figs. 4.1, 4.2 and 4.3 for wet (Oct., 2005) and dry seasons (Nov.-Dec., 2005). All the physicochemical variables except pH exhibited significant seasonal differences (Paired *t*-test, $p = 0.05$). Mean temperature, conductivity and DO were higher in the wet season than the dry season and the converse applied for BOD_5, NH_4-N and PO_4-P.

Temperature

Water temperature ranged from 27.1 to 30.9°C in the wet season, and in the dry season was between 24.7 and 26.1°C. Temperatures in the upstream were relatively lower than those in downstream sites (Fig.4.1-a).

pH

Both in the wet and the dry season, pH values of all the 12 sites remained about neutral: 7.0-7.6. No variation was observed between the wet and the dry period regarding pH values (Fig.4.1-b).

Electrical Conductivity (EC)

All the sampling sites showed low level of electric conductivity. EC values varied between 71 and 125 μScm^{-1} in the wet season, and this range in the dry season was from 73 to 208μScm^{-1}. In the wet season, the highest EC level was recorded at Norikhal but in dry season, Demraghat had the highest conductivity (Fig.4.1-c).

Dissolved Oxygen (DO)

DO values followed the similar pattern in both dry and wet season. The most upstream site had the highest DO value but it gradually decreased towards the downstream sites. At Norikhal, DO value decreased to its lowest level and increased considerably at the immediate downstream site although it followed a decreasing trend again in the downstream direction. In the wet period, DO values in the Balu River oscillated between 6.2 and 2.9 mgl^{-1} and those were between 4.8-0.8 mgl^{-1} in the dry period (Fig.4.2-a). On the other hand, DO levels in the Norikhal were 0.6 and 0.1 mgl^{-1} in the wet and dry season, respectively.

Biochemical Oxygen Demand (BOD_5 20°C)

In the wet season, BOD_5 values in all sites were below 4 mgl^{-1} except Noripur where this value was 5.4 mgl^{-1}. During the dry season, the lowest BOD_5 level was measured at Pubail and Udail, the most upstream sites and the downstream sties ranged between 6.5-10.5 mgl^{-1}. During this season BOD_5 level was very high in the Norikhal point with 30.5 mgl^{-1} (Fig.4.2-b).

(a)

(b)

(c)

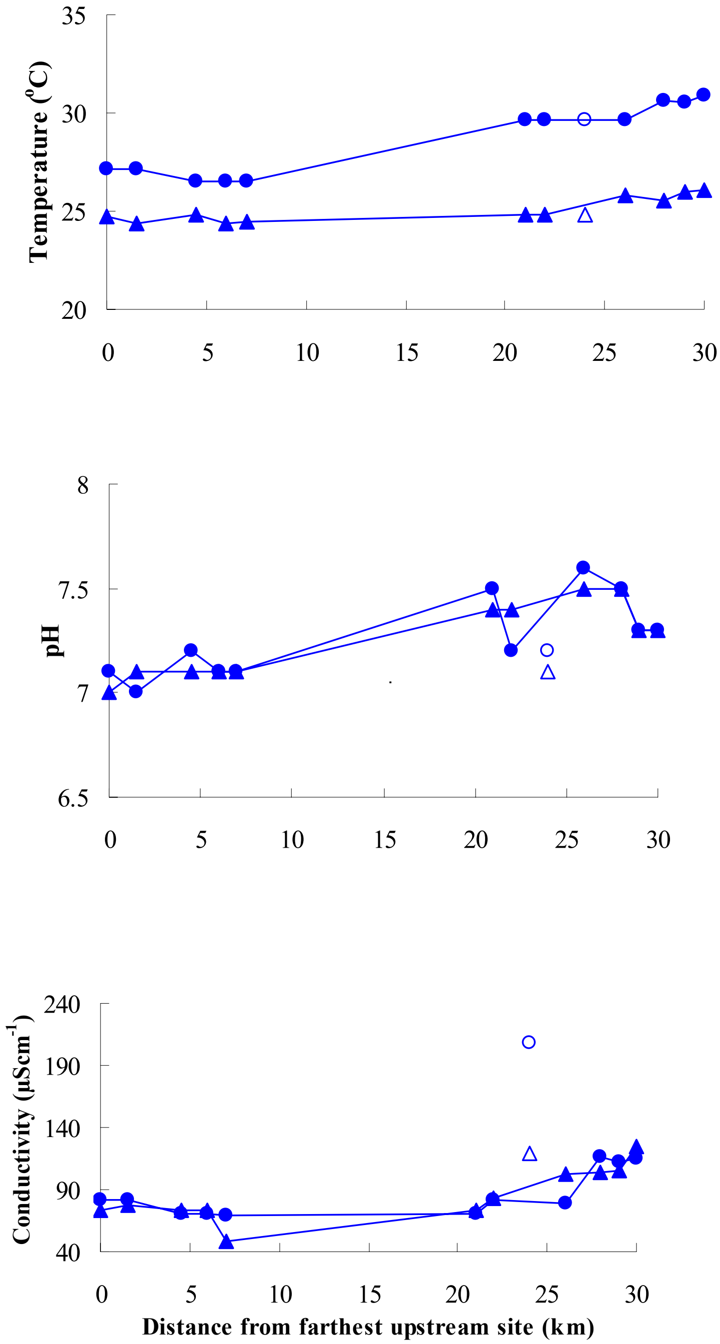


Fig. 4.1 Seasonal and spatial patterns of water temperature (a), pH (b) and conductivity (c) in the River Balu and Norikhal- a canal of the Balu River in Bangladesh. Wet (Oct.'05) and dry (Nov-Dec'05) season values in the River Balu represented by ● and ▲, respectively while those in the Norikhal represented by ○ and △, respectively.

(a)

(b)

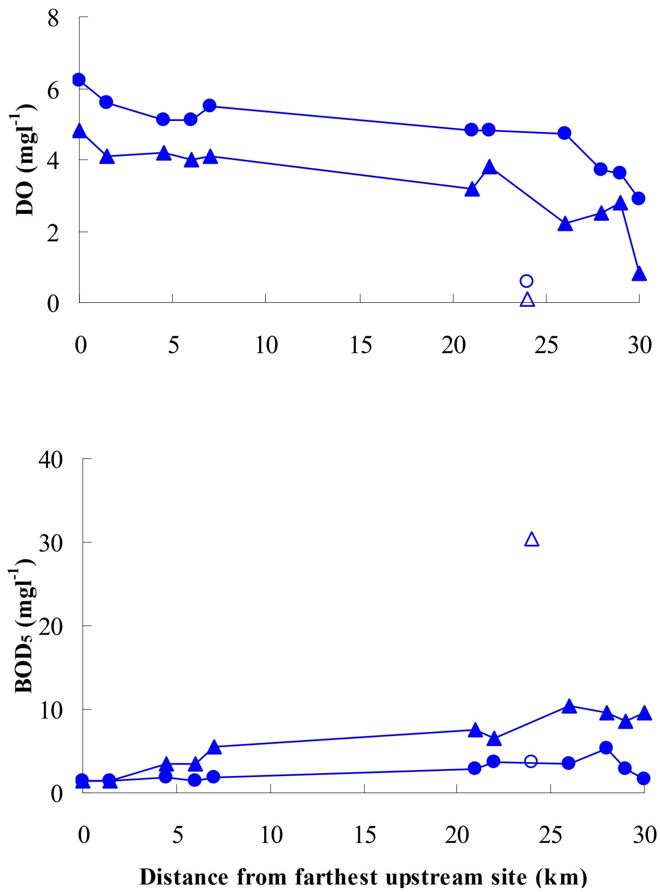


Fig. 4.2 Seasonal and spatial patterns of DO (a) and BOD_5 (b) in the River Balu and Norikhal- a canal of the Balu River in Bangladesh. Wet (Oct'05) and dry season (Nov.-Dec.'05) values in the River Balu represented by ● and ▲, respectively while those in the Norikhal represented by ○ and △, respectively.

Ammonium Nitrogen (NH_4^+-N)

In the wet season, NH_4-N level varied between 0.02-0.97 mgl^{-1} in the sampling sties of the Balu River while the Norikhal had a concentration of 2.83 mgl^{-1}. NH_4-N level in the dry season considerably increased in the downstream sites starting from immediate upstream point of Norikhal. During this time, the values ranged from 0.1 to 0.34 mgl^{-1} in the upstream sites of the Balu and its downstream sites were between 2.18-3.95 mgl^{-1}. Norikhal showed very high concentration of NH_4-N, 11.09 mgl^{-1} (Fig.4.3-a).

Nitrate Nitrogen (NO_3^--N)

NO_3-N values in majority of the sites were below the detection limit of the Dr. Langes's field kit (0.23-13.5 mgl^{-1}). Among the sites where NO_3-N values were measured, Norikhal showed the highest value of 0.76 mgl^{-1} followed by Noripur and Demraghat, which were 0.40 and 0.30 mgl^{-1}, respectively in the wet season. During the dry season, field kit could only measure the value of Norikhal and Noripur both at 0.23 mgl^{-1} and for rest of sites they were below the detection limit (Fig.4.3-b).

Orthophosphate (PO_4^{-3}-P)

Relatively high levels of phosphate were found in most of sites both in the wet and dry seasons. However, phosphate values did not display any definite pattern. In the wet season these values fluctuated between 0.05-0.91 mgl^{-1} and those in dry season between 0.05 -0.81 mgl^{-1} in the sampling locations of the Balu river. In the dry season, Norikhal had a phosphate concentration of 1.07 mgl^{-1} which is almost 2.5 times higher than that observed in the wet season (Fig.4.3-c).

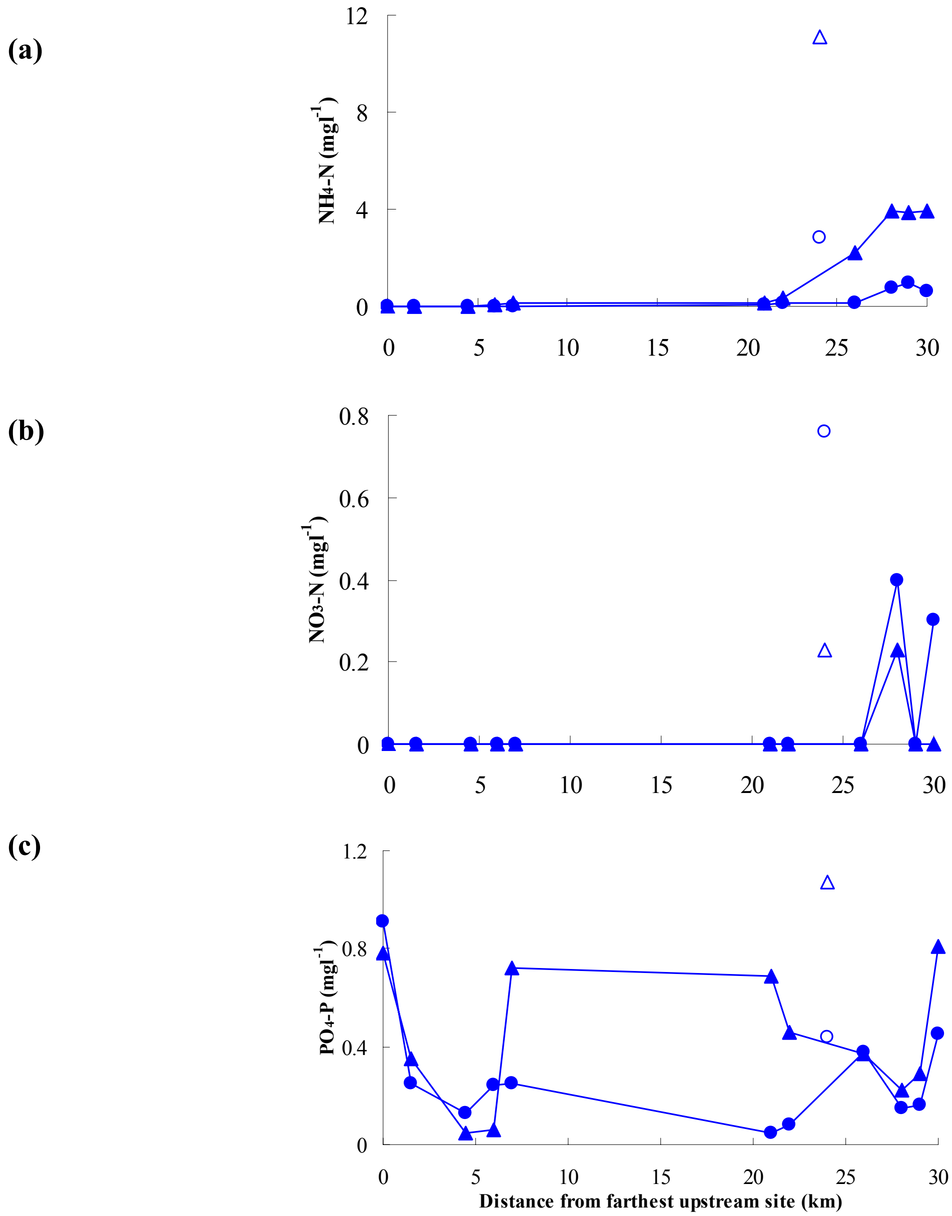


Fig. 4.3 Seasonal and spatial patterns of nutrients: ammonium-N (a), nitrate-N (b) and Orthophosphate (c) in the Balu River and Norikhal- a canal of the Balu in Bangladesh. Wet (Oct.'05) and dry (Nov.-Dec,'05) season values in the River Balu represented by ● and ▲, respectively while those in the Norikhal represented by ○ and △, respectively.

4.2 CHEMICAL WATER QUALITY CLASSIFICATION

Table 4.1 shows the rank of the study sites for each of the five parameters and average rank for each site. The mean rank of the study sites was between 2.4 and 11.8. Based on the average chemical rank, a scale with the values ranging from 2 to 12 was used to rate the quality of water. The minimum and maximum values of the scale is close to lowest and highest mean chemical rank of the study sites, respectively. The scale was then equally divided into four with the interval of 2.5 and sites were categorized as class1 (2-4.5), class 2 (4.5-7), class 3 (7-9.5) and class 4 (9.5-12). The water quality class 1, 2, 3 and 4 reflects unpolluted, slightly polluted, moderately polluted and grossly polluted, correspondingly. The mean ranks of the study sites are plotted against those four classes in Fig. 4.4.

The first four sites (site no. 1- 4) had the lowest mean chemical ranks and ranged from 2.4 to 3.4. All these four sites were in water quality class 1. Based on mean rank (4.6 - 6.6), site number 5, 6 and 7 were the next group of sites with similar water quality and were in water quality class 2. Site nos. 9-12 were in chemical water quality class 3 with the mean rank between 7.8 and 8.6. Demraghat and Norikhal had the highest mean chemical rank of 10.6 and 11.8, respectively and were rated as class 4 sites. Fig. 4.5 shows mean chemical rank and water quality class (WQC) for each site.

Table 4.1 Chemical water quality class based on mean chemical rank of physicochemical parameters for each site in the River Balu and Norikhal in dry season (Nov.-Dec.'05).

Sampling site	BOD_5 (mgl^{-1})	Rank	NH_4-N (mgl^{-1})	Rank	PO_4-P (mgl^{-1})	Rank	DO (mgl^{-1})	Rank	Conductivity (µScm-1)	Rank	Mean Rank	Class
Pubail	1.5	1	0.01	1	0.78	10	4.8	1	74	3	3.2	1
Udail	1.5	1	0.02	2	0.35	5	4.1	3	78	6	3.4	1
Raidah	3.5	3	0.03	3	0.05	1	4.2	2	74	3	2.4	1
Bindhain	3.5	3	0.04	4	0.06	2	4.0	5	74	3	3.4	1
Trimohoni	5.5	5	0.12	5	0.72	9	4.1	3	48	1	4.6	2
Fakirkhali	7.5	7	0.15	6	0.69	8	3.2	7	73	2	6	2
Kaikpara	6.5	6	0.34	7	0.46	7	3.8	6	83	7	6.6	2
Norikhal	30.5	12	11.09	12	1.07	12	0.1	12	119	11	11.8	4
W.Chanpara	10.5	11	2.18	8	0.37	6	2.2	10	103	8	8.6	3
Noripur	9.5	9	3.95	11	0.22	3	2.5	9	104	9	8.2	3
E.Chanpara	8.5	8	3.88	9	0.29	4	2.8	8	105	10	7.8	3
Demraghat	9.5	9	3.90	10	0.81	11	0.8	11	125	12	10.6	4

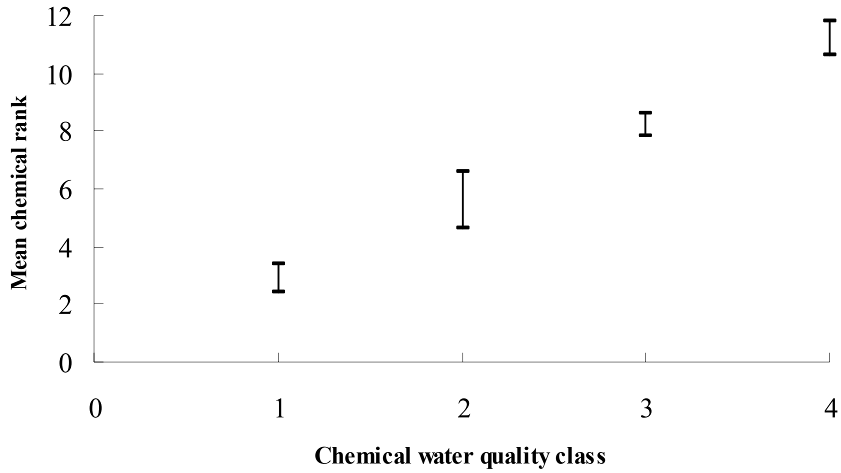


Fig. 4.4 Range of mean chemical ranks for sites in each chemical water quality class

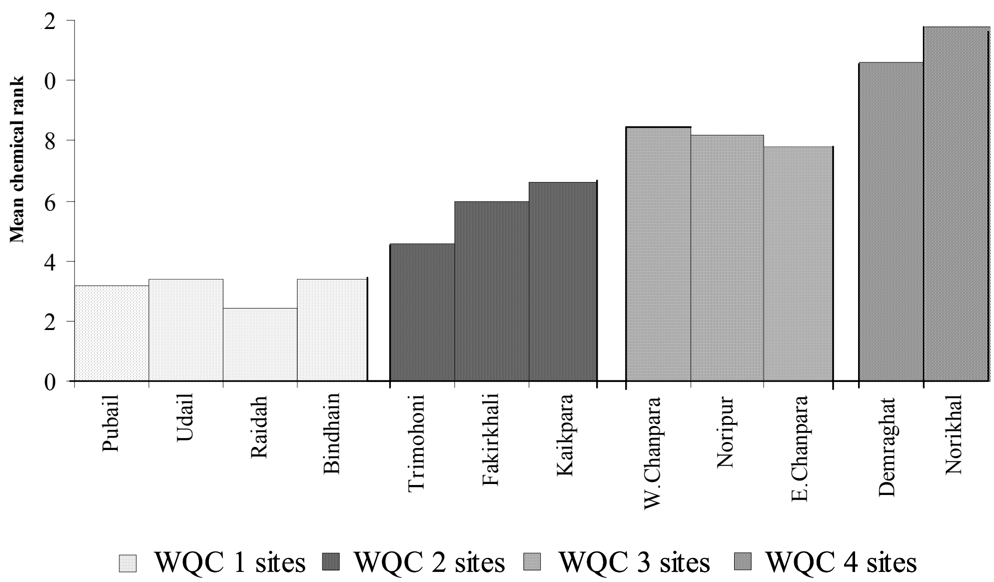


Fig. 4.5 Mean chemical rank and water quality class (WQC) for each site

4.3 HYDROLOGICAL FACTORS

Hydrological parameters were measured during the dry seasons at Raidah-a upstream site and at a downstream site proximity to Fakirkhali. The river width observed at both sides was approximately 75 meter, but the depth at Raidah was 1.5 meters while in downstream site it was around 3.0 meters. Similarly velocity and discharge at lower reach of the river were found higher than those at higher reach. Raidah had the surface velocity and discharge of 0.26 ms^{-1} and 29 m^3s^{-1} and the same at downstream site were 0.40 ms^{-1} and 100 m^3s^{-1}, respectively.

4.4 BIOLOGICAL DATA

Taxa Richness

The complete list of taxa sampled during October through December in the Balu River and Norikhal is given in Table 4.2. A total of 31 taxa were found in the study sites. The highest number of taxa (24) was observed in Pubail and Raidah while the lowest number of taxa (10) was found in Norikhal. The range of the number of taxa in slightly polluted sites was from 17 to 13, but in moderately polluted sites was 14 (Fig. 4.6). Of the total of 31 taxa, 94% were sampled in class-1, 65% in class 2, 55% in class 3 and 42% in class 4 sites (Fig. 4.7).

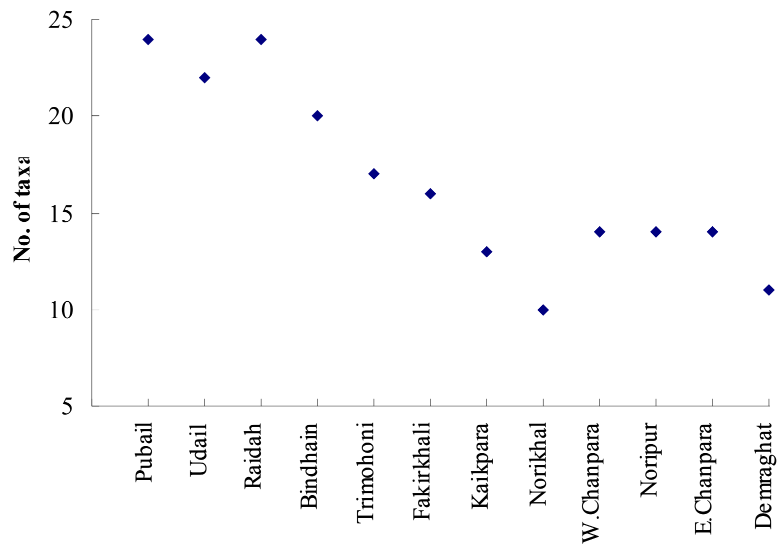


Fig. 4.6 Number of macroinvertebrate taxa in different study sites in the River Balu and Norikhal in Bangladesh between Nov.-Dec.'05

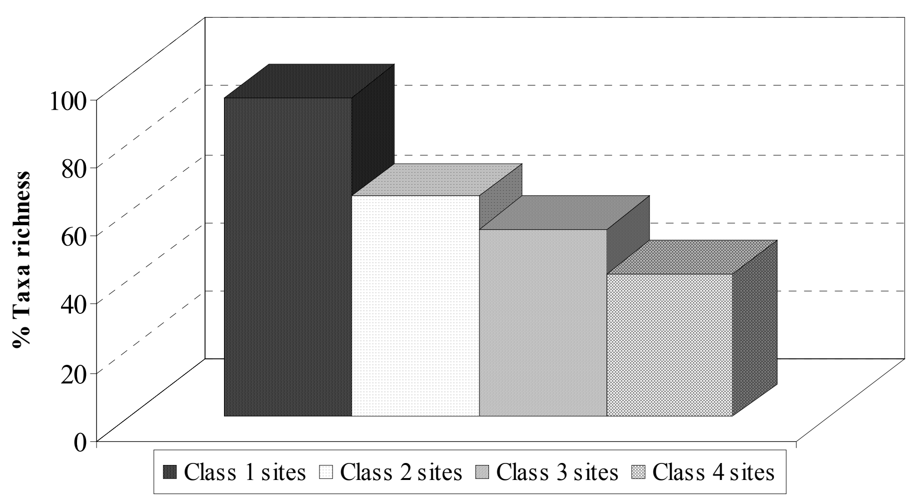


Fig. 4.7 Percentage of taxa richness for study sites in each water quality class

Table 4.2 Inventory of macroinvertebrate taxa found in different study sites in the River Balu and Norikhal in Bangladesh between Nov.-Dec.'05

Common Name & Order	Taxa	**Class 1 sites**				**Class 2 sites**			**Class 3 sites**			**Class 4 sites**	
		Pubail	Udail	Raidah	Bindhain	Trimohoni	Fakirkhali	Kaikpara	W.Chanpara	Noripur	E.Chanpara	Demraghat	Norikhal
Mayflies (Ephemeroptera)	Baetidae	X		X									
Caddisflies (Trichoptera)	Beraeidae	X		X									
	Molannidae	X	X	X	X								
	Sericostomatidae	X		X									
	Lepidostomatidae			X		X							
	Philopotamiidae	X											
Dragonflies (Odonata)	Corduliidae	X	X	X	X	X							
	Libellulidae	X	X	X	X	X	X	X	X	X			
Damselflies (Odonata)	Coenagriidae	X	X	X			X						
Waterbeetles (Coleoptera)	Hydrophilidae	X											
	Coleoptera larva	X	X										
Waterbugs (Hemiptera)	Hydrometridae			X		X	X		X	X	X	X	X
	Gerridae	X	X	X	X	X							
	Belostomatidae	X	X	X	X	X	X	X		X	X	X	X
Blackflies (Diptera)	Simulade								X			X	X
Midges (Diptera)	Chironomidae	X	X	X	X	X	X	X	X	X	X	X	X
Leeches(Hirudinea)	Glossiphoniidae								X	X	X		
Bristle Worms	Polychaeta		X		X		X		X	X			
Worms	Oligochaeta	X	X	X	X	X	X	X	X	X	X	X	X
Snails (Class: Gastropoda)	Viviparidae	X	X	X	X								
	Pilidae	X	X	X	X	X	X	X	X	X	X	X	X
	Bithynidae	X	X	X	X	X	X	X	X	X	X	X	X
	Thiaridae	X	X	X	X	X	X	X	X	X	X	X	
	Hydrobiidae	X	X	X	X	X	X	X	X		X		
	Neritidae		X		X								
	Lymnaeidae	X	X	X	X			X	X		X		X
	Planorbidae	X	X	X	X	X	X	X	X	X	X		X
Mussels (Class: Bivalvia)	Unionidae	X	X	X	X	X	X	X		X	X	X	
	Sphaeridae	X	X	X	X	X	X	X		X	X	X	X
Crustacea	Palaemonidae	X	X	X	X	X	X	X	X	X	X	X	
	Alpheidae		X	X	X	X	X						
Total no. of Taxa		**24**	**22**	**24**	**20**	**17**	**16**	**13**	**14**	**14**	**14**	**11**	**10**

Composition of Taxa

The biggest group of macroinvertebrates in the study area with respect to number of taxa was snails, which accounted for 33% of the total macrofauna encountered followed by caddisflies, 13%. Mayflies, damselflies, blackflies, normal worm and bristle worm all together contributed for 20% in the taxa composition (Fig. 4.8).

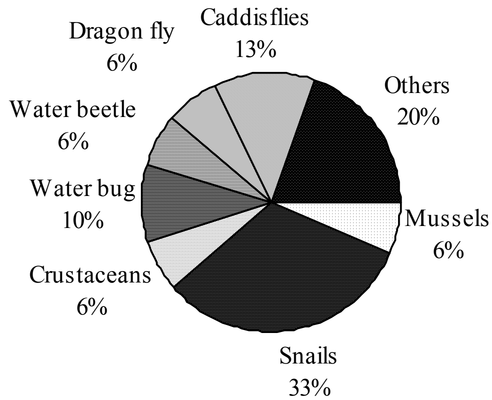


Fig. 4.8 Composition of macroinvertebrates from all study sites in the River Balu and Norikhal in Bangladesh

Distribution of Taxa

Fig. 4.9 shows the distribution of macrofauna taxa in the Balu river and its tributary, Norikhal. A total of thirty-one macroinvertebrate taxa were found in the study sites of which four taxa where common in all twelve sites, viz., Chironomidae, Oligochaeta, Pilidae and Bithyniidae. Next most common taxa were Belostomatidae, Thiaridae, Planorbidae, Sphaeridae and Palaemonidae, which were found in eleven sites.

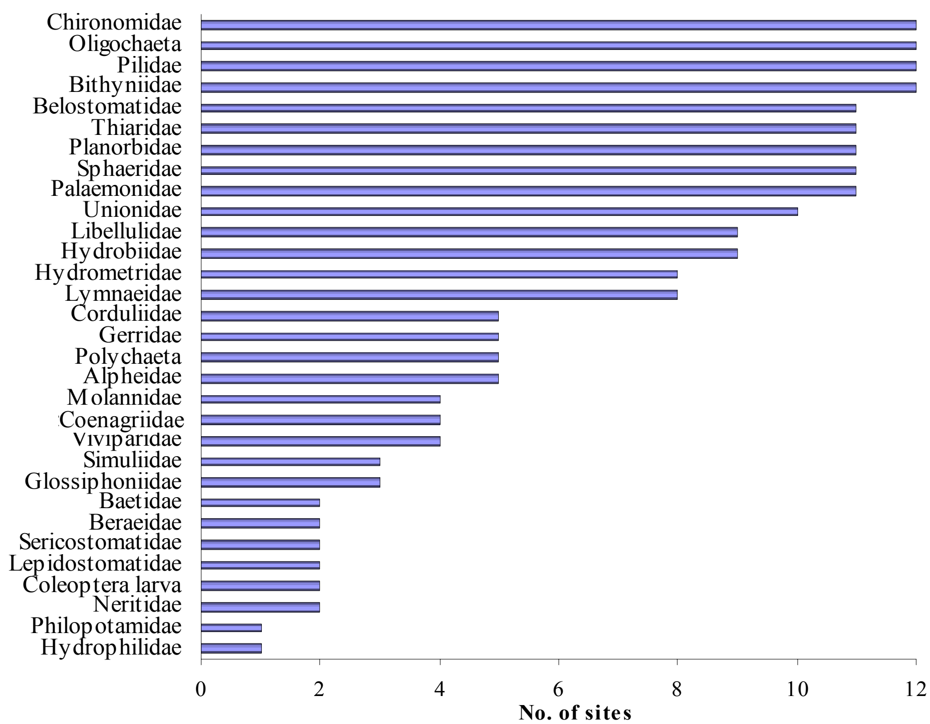


Fig. 4.9 Distribution of macroinvertebrate taxa in the River Balu and Norikhal in Bangladesh

EPT Richness

Six families belonging to the orders Ephemeroptera and Trichoptera were found in the study area. No Plecoptera taxon was encountered in any sites. Mayflies and Caddisflies were restricted to only class 1 and 2 sites. Trimohoni is the only class 2 sites where Lepidostomatidae, the lone Trichoptera family was found.

Biological Monitoring Working Parity (BMWP) Score and Average Score Per Taxon (ASPT)

The total taxa encountered in the study area were 31 of which the number of BMWP taxa was 24 (Fig. 4.10). The local taxa were Belostomatidae, Polychaeta, Pilidae, Thiaridae, Bithynidae, Palaemonidae and Alpheidae.

The most upstream site Pubail showed the highest BMWP score of 109 while Norikhal and Demraghat jointly scored the lowest, 25 (Fig. 4.11a). ASPT did not follow the same pattern as BMWP score. Norikhal had the lowest ASPT score of 3.13 while Raidah showed the highest score of 5.58 (Fig. 4.11b).

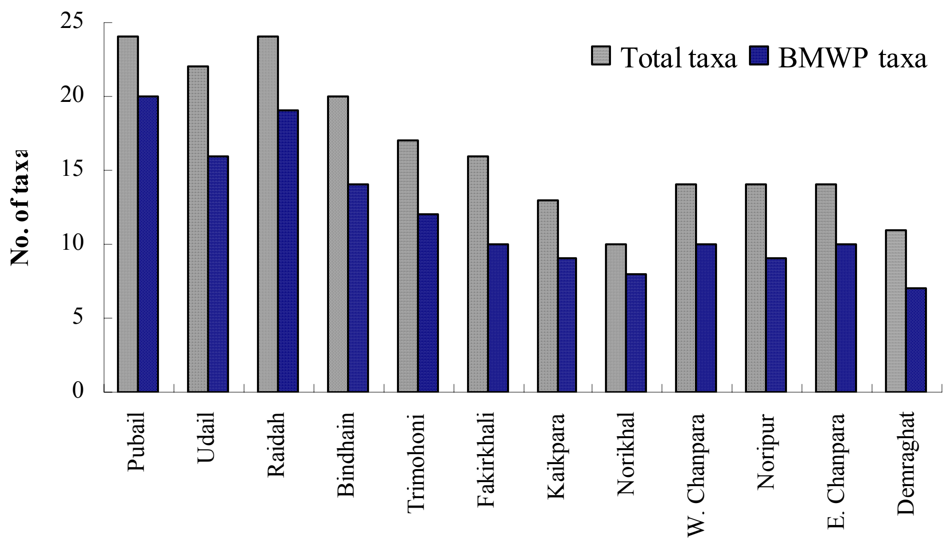


Fig.4.10 Barchart to show total and BMWP taxa richness in different study areas

(a)

(b)

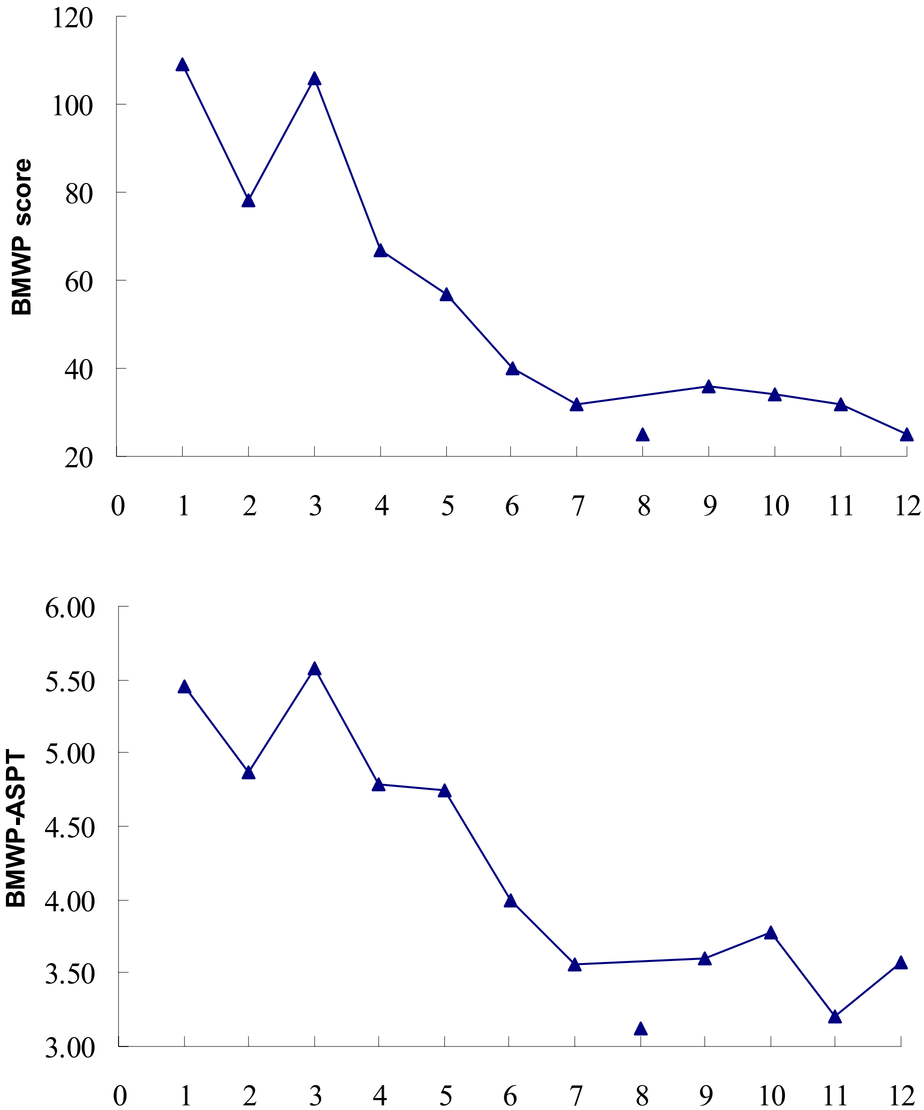


Fig. 4.11 Graphs to show biotic index score. BMWP score (a) and ASPT (b)

Community Loss Index (CLI)

Community Loss Index displayed a gradual upward trend towards downstream sites. CLI values were calculated with respect to two class 1 sites with the lowest mean chemical ranks namely, Pubail and Raidah. These two sites were considered as reference sites. All class 1 sites showed the lowest community loss values and the class 4 site had the highest. And those of the class 2 and 3 sites were intermediate.

When Pubail was considered as reference site, index points of class 1 sites were <0.4 followed by class 2 of 0.59 - 0.85 followed by class 3 sites with a range from 0.86 to 1.00. Demraghat and Norikhal, the class 4 sites had the highest community loss values of 1.36 and 1.60, respectively (Fig. 4.12).

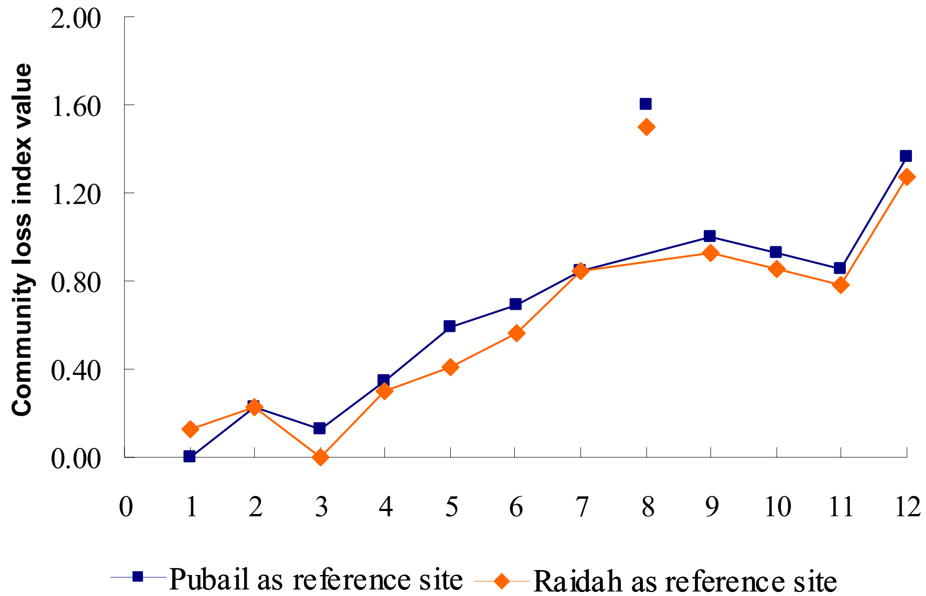


Fig. 4.12 Community loss index for each site

Sequential Comparison Index

Sequential comparison index values were almost same in all study sites. The highest index value of 0.89 was observed in Raidah and the lowest value of 0.72 was in Demraghat and Norikhal (Fig. 4.13). The average SCI value was 0.76 with a standard deviation of 0.05.

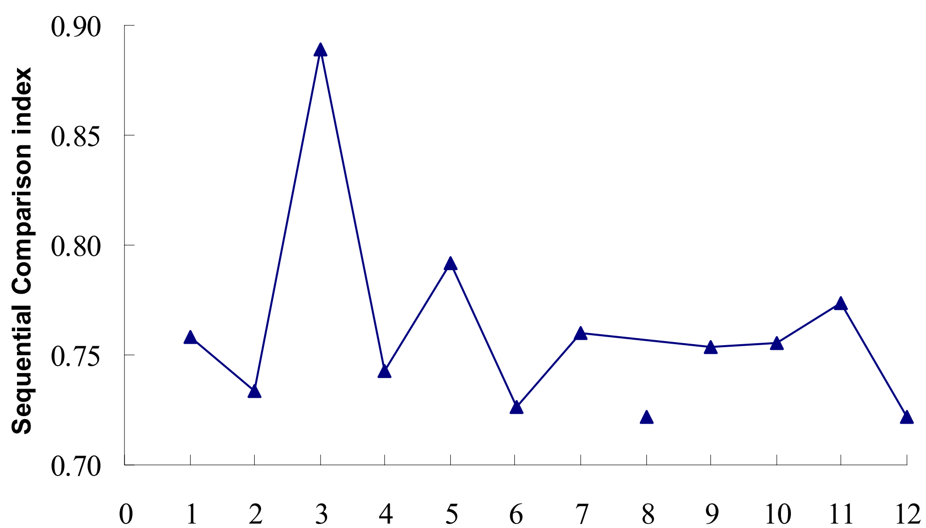


Fig. 4.13 Sequential comparison index values from different study sites

4.5 THE RELATIONSHIP OF BIOLOGICAL PARAMETERS WITH PHYSICO-CHEMICAL PARAMETERS

A significant Spearman's correlation existed ($p<0.01$) with all measured biological indices and physicochemical parameters except orthophosphate. Taxa richness, BMWP, BMWP-ASPT and SCI had significantly positive correlation with DO, but negative correlation with BOD_5, NH_4-N and conductivity. On the other hand, CLI followed the reverse trend; it had a significant negative correlation with DO and a positive correlation with BOD_5, NH_4-N and conductivity.

Table 4.3 Spearman's correlation coefficient (r) of biological indices and physicochemical parameters. * Indicates correlation is significant at the 0.01 level (2-tailed).

Parameter	DO (r)	BOD_5 (r)	NH_4-N (r)	PO_4-P (r)	Conductivity (r)
Taxa richness	0.908*	-.869*	-.917*	-.455	-.753*
BMWP	0.896*	-.848*	-.926*	-.393	-.777*
ASPT	0.889*	-.860*	-.885*	-.363	-.750*
SCI	0.903*	-.860*	-.908*	-.486	-.752*
CLI					
Pubail as reference site	-.981*	.963*	.958*	0.378	.775*
Raidah as reference site	-.960*	.935*	.930*	0.462	.754*

4.6 EVALUATION OF THE BIOLOGICAL INDICES

Graphs (Fig. 4.14 & 4.15) were used to evaluate the responses of indices to pollution gradient. A total of five indices were tested against average chemical ranks used as the gradient and of them, four displayed predicted response to the pollution gradients and reasonably separated four water quality classes (Table 4.4).

In the case of the measures of richness, values of taxa richness index declined gradually along the gradient from lest polluted sites (class 1 sites) to grossly polluted sites (class 4 sites) and separated the four classes of sites satisfactorily (Fig. 4.14a). Although EPT richness was a candidate index to be tested, it was eventually excluded because no Plecoptera taxon was found at any site.

Both biotic measures namely, BMWP and BMWP-ASPT met the test criteria through displaying a clear relationship with the pollution gradient and were able to distinguish sites in four water quality classes (Fig 4.14b & 4.14c). In case of BMWP-ASPT, class 3 and class 4 were distinguished less effectively (Fig. 4.14c).

Sequential comparison index, the only diversity index tested showed almost no relationship with the environmental gradient and poorly distinguished all the sites in 4 water quality classes (Fig. 4.15a). In contrast, community loss index performed well. According to prediction, community loss index value increased gradually from water quality class 1 sites towards the sites in class 4 and achieved considerable separation of these four classes (Fig. 4.15b).

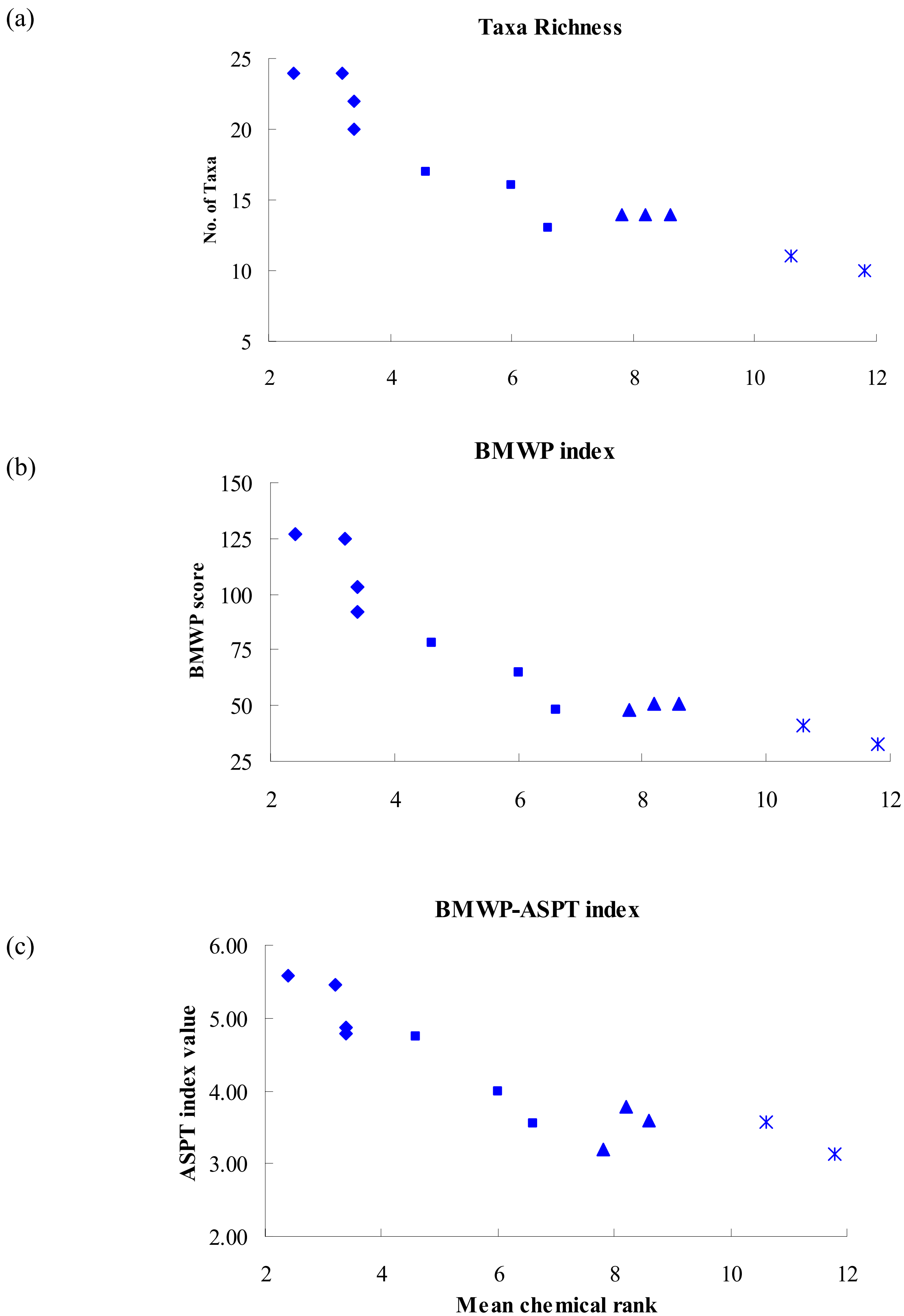


Fig. 4.14 Responses of indices to environmental pollution gradient. Class 1-4 represented by ◆, ■ , ▲ and ✳, respectively.

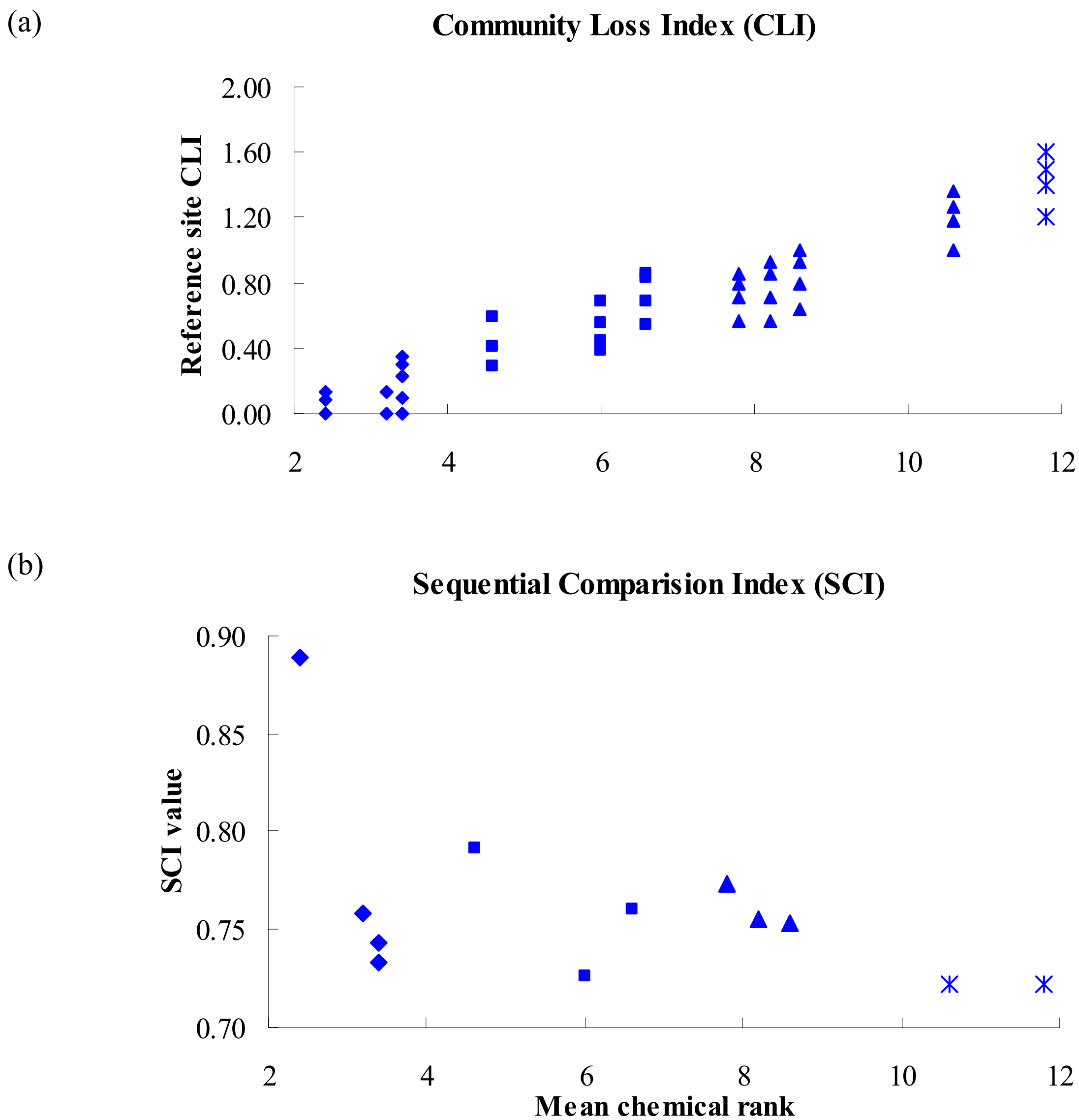


Fig. 4.15 Responses of indices to environmental pollution gradient. Class 1-4 represented by ◆, ■, ▲ and ✳, respectively

Table 4.4 Predicted response to the pollution gradient of the test indices

System/Methods	Index/Score	Predicted response	Meets test criteria
Richness	Taxa richness	Decrease	Yes
Biotic system	BWMP	Decrease	Yes
	BWMP-ASPT	Decrease	Yes
Diversity and similarity/loss	Sequential comparison index	Decrease	No
	Community loss index	Increase	Yes

4.7 DETERMINATION OF TOLERACE VALUES FOR BANGLADESHI RIVER MACROINVERTIBRATES

The principal component analysis (PCA) resulted in one axis with eigenvalues larger than 1. There was a very large first principal component (PC1 axis) explaining over 67% of the total variation in the data (Table 4.5). All of the variables were significantly correlated with the first principal component, but orthophosphate had weaker correlation than the other variables (Table 4.6).

Table 4.5 Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	3.362	67.237	67.237	3.362	67.237	67.237
2	.899	17.984	85.222			
3	.409	8.178	93.400			
4	.302	6.046	99.445			
5	.028	.555	100.000			

Table 4.6 Spearman's rank correlation between physicochemical parameters used in PCA and first principal component (PC1).

Variable	PC1
Ammonium Nitrogen	0.940
Biochemical Oxygen Demand	0.912
Dissolved Oxygen	-0.883
Electrical Conductivity	0.804
Orthophosphate	0.471

Across all taxa, minimum TV_{init} (TV_{min}) was -1.032 and maximum TV_{init} (TV_{max}) was 0.594. The initial tolerance values (TV_{init}) of all the taxa except Simuliidae are given in Table 4.7 TV_{init} of Simuliidae taxon was excluded because its value was detected as outlier.

Table 4.7 Initial tolerance values of the taxa occurring in the River Balu and Norikhal in Bangladesh

Name of Taxa	TV_{init}	Name of Taxa	TV_{init}	Name of Taxa	TV_{init}
Baetidae	-1.013	Coleoptera larva	-1.006	Lymnaeidae	0.215
Beraeidae	-1.013	Hydrometridae	0.594	Planorbidae	0.137
Molannidae	-0.980	Gerridae	-0.909	Unionidae	-0.091
Sericostomatidae	-1.013	Belostomatidae	0.220	Sphaeridae	0.220
Lepidostomatidae	-0.817	Pilidae	0.253	Palaemonidae	-0.021
Philopotamidae	-1.032	Chironomidae	0.253	Thiaridae	-0.021
Corduliidae	-0.909	Glossiphoniidae	0.530	Hydrobiidae	-0.297
Libellulidae	-0.294	Polychaeta	-0.072	Neritidae	-0.951
Coenagriidae	-0.721	Oligochaeta	0.253	Alpheidae	-0.704
Hydrophilidae	-1.032	Viviparidae	-0.980	Bithyniidae	0.253
Simuliidae	1.358				

The tolerance values of 30 taxa were calculated of which 28 are in family level and 2 are in higher level. The scale of a 0-10 range was used to measure taxa tolerance where 10 indicates the most tolerant and 0 is the least tolerant taxa (Table 4.8).

Table 4.8 Tolerance values for macroinvertebrate taxa scaled to a 0-10 range. The greater the tolerance of taxa to pollution, the higher the tolerance scores.

Tolerance value	Name of taxa
0	Baetidae, Beraeidae, Molannidae, Sericostomatidae, Philopotamidae Hydrophilidae, Coleoptera larva, Viviparidae, Neritidae
1	Lepidostomatidae, Gerridae, Corduliidae
2	Coenagriidae, Alpheidae
5	Libellulidae, Hydrobiidae
6	Polychaeta, Unionidae, Thiaridae, Palaemonidae
7	Planorbidae
8	Lymnaeidae, Sphaeridae, Belostomatidae, Pilidae, Bithyniidae, Oligochaeta, Chironomidae
10	Hydrometridae, Glossiphoniidae

5. DISCUSSION AND CONCLUSIONS

This study clearly shows that biological assessment based on macroinvertebrate communities provides useful information about water quality by displaying response to pollution. Biotic community-type indices such as taxa richness, BMWP, BMWP-ASPT and Community Loss Index were identified the most promising assessment endpoints for measurement of organic pollution in lowland floodplain rivers in Bangladesh.

5.1 PHYSICOCHEMICAL RESULTS

The physicochemical data confirm the spatial and temporal variability in pollution across the study sites in the Balu River. Monsoon rainfall in the wet season accompanied by flood has a strong effect on the dilution of the pollution in the river system. During the wet season, low concentration of nitrogenous pollutants (NH_4-N and NO_3-N) was observed across all the study areas and at the same time, oxygen levels were reasonably high. With respect to PO_4-P, the contrary applies for 50% of the sampling sites where the concentration values ranged from 0.91 to 0.25 mgl^{-1} and for the rest of the sites phosphate pollution was minimal.

On the contrary, during the dry season when water level receded, study sites displayed a marked variation in concentration of nutrients, dissolved oxygen, biochemical oxygen demand, etc. These apparent variation in water chemistry during the dry period were mostly influenced by landscape factors such as land use/land cover patterns, land management activities such as forest harvest, agriculture or residential development as well as domestic sewage/waste water discharge.

Upstream sites where land use is mostly dominated by agriculture and/or patch of woodland had low levels of NH_4-N, NO_3-N, BOD_5 but comparatively high levels of DO. Relatively low settlement density, presence of riparian vegetation, absence of industrial/household discharge and little agricultural runoff (during the dry season) were important explanatory factors for low concentration of nitrogen-related parameters and relatively higher values for DO.

Urban landuse, discharge of municipal waste water through the Norikhal as well as effluent discharge from a jute mill at Demraghat were the dominant factors accounted for high level of pollutants (nutrients) and low level of DO in the downstream sites.

During the dry season, moderate to high concentration of PO_4-P was observed in 75% of the study sites, which appeared to be regulated by factors that operate at different temporal and /or spatial scales. Explanatory factors could include river flow, primary productivity, macroinvertebrate and microbial organic matter processing, storm transport, riparian characteristics, crop type, crop management practices, or relative position of land use elements in the catchments (Johnson *et al.*, 1997).

In all sampling sites, EC values were found well below the World Health Organization (WHO) recommended desirable upper limit for drinking and protection of ecosystem. The WHO recommended 800 μScm^{-1} limit for drinking water and 1500 μScm^{-1} recommended for the protection of aquatic ecosystems. Among all twelve sites, Norikhal had the highest salinity level of 208 μScm^{-1} measured in the wet season.

The pollution status of the study sites was determined by ranking method. Calculating the mean rank for each site with respect to physicochemical parameters appears to be an effective method for summarizing these data although the parameters may be of varying importance.

Four discrete water quality classes were assigned to the sampling sties arbitrarily but it matches well with the mean ranks. Variation in chemical parameters, ranks and mean ranks within the sites of class 1, 2 and 3 is relatively small. In the case of sites belonging to class 4, there is large variation in pollutants concentration, but small changes in ranks and final mean ranks. However, this ranking method demonstrates the gradient of pollution and gives a good idea of the relative status of the sites, which is reasonable for the evaluation of the metrics (Thorne and Williams, 1997).

5.2 BIOLOGICAL DATA

The present study gives an overall picture of the macroinvertebrate communities occurring along the pollution gradient of the river Balu and its tributary, the Norikhal, which was previously lacking.

Dry season is the appropriate time for macroinvertebrate sampling in lowland floodplain rivers in Bangladesh. Because during the wet season, both water depth and velocity of the river remain high and when water overflows the banks macrofauna disperse in the floodplain, it causes serious problems in collecting the whole range of bottom fauna.

In the present study, hand nets were used for collection of aquatic macrofauna, which proved an effective method for sampling near to the riverbank (bank to 1 m water depth) and water submergent vegetation zones, but effectiveness declines with increasing water depth especially when the height of the water column in the channel exceeds 1 meter. This problem can be compensated by increasing the length of the handle (dip net).

Macroinvertebrates were identified up to family level except normal worm and bristle worm, they have been identified at class level. The change in the number of macroinvertebrate families (higher level in the case of worms) along the pollution gradient was observed in this study. It is important to note that before setting out to collect macrofauna, adequate knowledge has to be gained about the local taxa-their identification keys, habitat preferences, etc. At the family level, the macrofauna communities of the Balu River appear to be similar in their make-up and response to pollution to communities in the temperate European and North American countries. This observation corresponds closely with the views of Thorne and Williams (1997) that tropical communities of macrobenthos are broadly similar to those in temperate countries with a few exceptions.

Sampling sites belonging to water quality class 1 displayed highest taxa richness and accordingly, the number of taxa in class 4 sites was the lowest. Certain taxa were found throughout the gradients namely, Chironomidae, Oligochaeta, Pilidae and Bithynidae, which are tolerant. The Ephemeroptera and Trichoptera are the most sensitive orders and these were chiefly restricted to class 1 sites. Lepidostomatidae is the only family belonging to the Trichoptera order found only one location of the class 2 sites. The class Gastropoda is abundant in the Balu River. A total of 8 Gastropoda taxa were found from different sampling sites of which Viviparidae and Neritidae were found in only class 1 sites and rest of the taxa can be considered tolerant taxa as these were present in 4 classes of sites.

Although there are general similarities of macroinvertebrate families between temperate and tropical countries, some exceptions were observed in this context. No Plecoptera family was found in any sampling sites; conversely, in temperate countries it is an important indicator group of low level of pollution and an important measure of EPT richness. The same is true for Gammaridae and Asellidae. These families are abundant in temperate zone and act as reliable indicator animals (Thorne and Williams, 1997) but they are replaced by Alpheidae and Palaemonidae in Bangladesh. Polychaeta which is mostly a marine taxon was found in the Balu River. Indeed, Bangladesh fisheries research institute also reported the availability of Polychaeta in freshwater in Bangladesh (BFRI, 2005).

5.3 EVALUATION OF BIOLOGICAL INDICES

Taxa Richness

The richness of the community in terms of number of taxa indicates biodiversity of ecosystems and is commonly used as a quantitative measure of river water and habitat quality Taxa richness generally decreases as a stream ecosystem degrades (Resh and Grodhaus 1983). This is a very simple index to calculate the total number of taxa (families & higher levels), and was considered useful in discrimination of impacted and unimpacted sites by Resh & Jackson (1993).

In this study, the number of taxa declines with increasing pollution and disappearance of bank/riparian vegetation and thus performed well to distinguish the range of sites from class 1 to class 4. It is noteworthy that the number of taxa at the sampling site of the Norikhal, the severely polluted site was not too small as was expected before but all of them seemed to be tolerant taxa. The presence of marginal vegetation may be an explanatory factor playing an important role to support unexpectedly higher number of taxa.

EPT Richness

Many researchers have reported the effectiveness of EPT richness index to discriminating unimpacted sites of different habitat types and Resh (1995) recommended to apply this index in developing countries (as cited by Thorne and Williams, 1997). In Bangladesh context, the EPT richness index may be not equally effective because there was no Plecoptera taxon found in the whole gradient of Balu River. This is may be a rare family in other tropical countries as Durand & Leveque (1981) reported only one species of Plecoptera in the whole of west Africa (as cited in Thorne and Williams, 1997).

Biotic Indices

Both BMWP score and BMWP-ASPT showed the pollution gradient well and were largely able to separate the sites in 4 different water quality classes. Generally ASPT performs better than

BMWP. Because in the case of ASPT, the inherent weakness of the BMWP system resulting from the effect of sampling effort is overcome by dividing the BMWP Score by the number of taxa present in a particular site, but the converse applies in this study. BMWP-ASPT was less effective to distinguish class 3 from class 4 sites. Demraghat, a class 4 site, had the BMWP-ASPT index value of 3.57 where 7 out of 11 taxa belonged to BMWP taxa. On the hand, a total of 14 taxa were found in E.Chanpara, a class 3 site, of which BMWP taxa were 10 and BMWP-ASPT index value was 3.20. This discrepancy was due to assigning higher score to Unionidae family in BMWP score system. In this study, Unionidae was found in all sites except Norikhal. This suggests that BMWP score needs local modification as well as local taxa should be included in BMWP system in order to achieve better performance of the both biotic indices.

Diversity and Similarity/Loss Index

The sequential comparison index (SCI) could not demonstrate any clear relationship with the pollution gradient and assorted the index values of the whole sites belonging to 4 different water quality classes. The failure of this diversity index was largely because of large abundance of tolerant fauna across the gradient mostly 6 families of Gastropods, Oligochaeta, Chironomidae, Belostomatidae and Palaemonidae and contrarily, sensitive organisms were present in little abundance. de Zwart and Trivedi (1992) recommended for Sequential Comparison Index along with BMWP score for biological water quality evaluation in India. But according to this study, SCI seemed to be unsuitable for lowland rivers in Bangladesh.

Regarding similarity/loss index, community loss index values increased steadily along the so-called pollutional continuum i.e. unpolluted to grossly polluted sites and displayed a very stiff gradient and made explicit discriminations among the sampling sites and therefore, it met the test criteria significantly.

5.4 CORRELATION TESTS

Spearman's rank correlation results showed a significant correlation between biological indices and water chemistry of the river Balu except orthophosphate. All the biological indices are negatively correlated with nutrient concentrations (NH_4-N, NO_3-N and PO_4-P) as well as with electrical conductivity and positively with the dissolved oxygen of the water. These correlations are highly significant/ significant with all water chemistry variables except orthophosphate.

According to De Pauw and Roels (1988), the significant positive correlation between biotic indices and DO is quite normal because biotic indices have been developed principally in relation to organic pollution and the oxygen requirements of the macroinvertebrates. They also reported significant, but negative correlation between Belgium Biotic Index (BBI, Karr, 1981) with BOD, COD, NH_4 and PO_4.

With respect to correlation between PO_4 and biotic index, present study contradicts with the observation of De Pauw and Roels (1988). In the study of Miltner and Rankin (1998), Index of Biotic Integrity (IBI, Karr, 1988) was found to be negatively correlated with increasing concentration of total inorganic nitrogen and total phosphorus in headwaters and wadable streams, but no relationship was evident for large rivers.

5.5 DEVELOPMENT OF TAXA TOLERACE VALUES

This may be the first attempt to assign tolerance values to macroinvertebrates at family level (some cases in higher level) that occurring in the lowland floodplain river in Bangladesh. The tolerance values derived in this paper are very tentative, preliminary and nascent in nature because the level of adequacy of data was far less than the requirements to develop accurate and repeatable taxa tolerance values.

The principal component analysis (PCA) method was employed to determine taxa tolerance values. This PCA approach is dependent on existence or collection of large amounts of abiotic data (water chemistry variables, riparian vegetation, substrate type etc.) in order to effectively define the generalized stressor gradient existing across water bodies. These data need to be co-located with the biological data of interest (e.g. macroinvertebrate fauna). Consequently, principal component analysis approach may be not an effective method for Development of tolerance values for macroinvertebrate taxa at the very outset

However, when competent authority believes it has adequate data to effectively describe the general range of biological conditions found across a particular type of water resource, the PCA approach may be the most useful one (Blocksom and Winters, 2005).

A preliminary comparison between tolerance values of Bangladeshi taxa occurring in the Balu River/Norikhal and BMWP score is shown in Table 5.1. In order to correspond to BMWP scores tolerance values for macroinvertebrate taxa rescaled to a 1-10 range from 0-10 range and put in opposite order. Now that the greater the tolerance of taxa to pollution, the lower the tolerance scores.

Table 5.1 Pollution sensitivity grades for families (higher level in some cases) of river macroinvertebrates of Bangladesh and BMWP score.

Name of Taxa	TV of Bangladeshi Taxa	BMWP score
Baetidae, Beraeidae, Molannidae, Sericostomatidae	10	10
Philopotamidae	10	8
Hydrophilidae, Coleoptera larva	10	5
Viviparidae, Neritidae	10	6
Lepidostomatidae	9	10
Corduliidae	9	8
Gerridae	9	5
Coenagriidae	8	6
Alpheidae	8	-
Libellulidae	6	8
Hydrobiidae	6	3
Polychaeta	5	-
Unionidae	5	6
Thiaridae, Palaemonidae	4	-
Planorbidae, Lymnaeidae, Sphaeridae	3	3
Belostomatidae, Pilidae, Bithyniidae	3	-
Chironomidae	3	2
Oligochaeta	3	1
Hydrometridae	1	5
Glossiphoniidae	1	3

5.6 INDICATORS FOR ASSESSING PROGRESS TOWARDS THE MDG TARGET 9: BIOLOGICAL WATER QUALITY IN FRESHWATER ECOSYSTEMS

The Millennium Development Goal number 7 particularly aims to ensure environmental sustainability, and its Target 9, which calls for "integrating the principles of sustainable development into country policies and programs and reversing the loss of environmental resources" places the environment at the centre of development.

⊕Millennium Ecosystem Assessment

The over 1,000 scientists brought together by the Millennium Ecosystem Assessment (MA) have gathered the best available scientific knowledge to assess the state of the environment. Their mandate was to "assess the consequences of ecosystem change for human well-being and to establish the scientific basis for actions needed to enhance the conservation and sustainable use of ecosystems and their contributions to human well-being" (Millennium Ecosystem Assessment 2005a). The MA finds that "humans have changed ecosystems more rapidly and extensively than in any comparable period of time in human history…and this has resulted in a substantial and largely irreversible loss in the diversity of life on Earth." The MA's scenarios illustrate how environmental degradation may worsen over coming decades unless corrective action is taken.

⊕Fresh water ecosystems and Human Well-being

In the Millennium Assessment, fresh water is a ''provisioning'' service as it refers to the human use of fresh water for domestic use, irrigation, power generation, and transportation. However, fresh water and the hydrological cycle also sustain inland water ecosystems, including rivers, lakes, and wetlands. These ecosystems provide cultural, regulating, and supporting services that contribute directly and indirectly to human well-being through recreation, scenic values, and maintenance of fisheries. Fresh water also plays a role in sustaining freshwater-dependent ecosystems such as mangroves, inter-tidal zones, and estuaries, which provide another set of services to local communities and tourists alike.

A target of halving the number of people who cannot access safe drinking water is the only explicit water-related MDG. And it is implicit in the seventh MDG -Target 9. Still the full range of freshwater ecosystem services will come into play—directly or indirectly—in achieving these goals (UN World Assessment Program 2003).

⊕Measurable MDG Targets for Developing Environmental Strategies

An MDG-based strategy for sustainable development takes the MDG targets as the starting point. But MDG 7 does not provide a comprehensive set of environmental targets, countries need to operationalize environmental sustainability by establishing concrete, quantifiable objectives for reversing the loss of environmental resources. Schmidt-Traub and Cho (2005) suggest that as a first step for preparing *outcome-oriented* environmental strategies, the environmental working group will start by identifying environmental priorities, such as halting desertification and land degradation in certain parts of a country, protecting a specific freshwater ecosystem, or reducing particulate emissions in a major city. And for this priority area, countries will then develop quantified outcome targets to be achieved by 2015. To be useful, targets must be quantified, time bound, and refer to measurable *outcome* objectives, such as reductions in the pollution of freshwater resources.

⌖Biological Indicators of Water Quality for Assessing MDG Target

Water bodies with integrity, especially rivers, have persisted in, even modified, their regions'physical and chemical environments over millennia (Karr, 1999). As human populations increase and technology advances, land-scapes are altered in a variety of ways viz., modification of seasonal water flow, changes in the system's food base, changes in interactions among stream organisms and contamination of the water with chemicals and those changes alter the river's biota and thus the entire biological context of the river, causing it to diverge from integrity. By measuring biological condition and evaluation the result as divergence from baseline biological integrity, we can thus focus on the most integrative, biological endpoint (Karr, 1999).

The specific reaction of macroinvertebrate species to different types of environmental influences is used to measure the biological quality of water bodies. The relative abundance of indicator organisms, their community structure and function in the ecosystems give information on the effects of the influences: current velocity, substratum composition, organic pollution and nutrient enrichment, toxic chemicals etc. Biological indices incorporate those that illustrate changes in taxonomic richness, shifts in species composition reflecting human effects (sedimentation or nutrient enrichment), individual heath, food web organization, and other biological attributes that respond to human influence (Karr, 1999).

Protection and restoration of freshwater ecosystems is one of the priority areas in Bangladesh that has been brought under the PRSP thematic sectors with a linkage to MDGs 1 &7. In order to prepare MDG-based development strategies, Bangladesh government can use biological indicator to set out-come based objectives (e.g. return of fish). Biological water quality indices can be used to judge river health against defined objectives because they are more effective at diagnosing degradation, defining its cause(s), and suggesting treatment to halt or reverse the damage (Karr, 1999). With refer to measurable outcome objective, indices can be used to set quantifiable targets and to evaluate the success of management decision towards the attainment of targets.

Within the framework of biological monitoring programme, local community groups, landholders or other local interest groups can be involved for surveillance of water quality with the simple affordable technology like spot assessments of biological activities via macroinvertebrates in water, observations of birds or fish. WaterWatch in Australia is a good example of a successful community based biological assessment programme. With this communication technique, people will have the understanding of condition and trend in river health and the consequences of human activities for changes in biological systems. Through this community participation, freshwater restoration efforts can make a success.

In conclusion, Biological monitoring systems using macroinvertebrate communities identified at family level appear to have great potential for use in Bangladesh. Biological indices proposed to use for this monitoring system can be suitable indicators for assessing the progress towards achievement of MDG target 9 in protection and restoration of freshwater ecosystems in Bangladesh.

6. RECOMMENDATIONS

1. A database should be developed with at least 2 years sampling data for water chemistry, physical habitat, landuse and macroinvertebrate taxa for multiple rivers. First year data can be used for developing and calibrating indices and the second year would be used for evaluating the indices.

2. Selection and calibration of macroinvertebrate indices should be performed with respect to reference sites data. Reference or degraded site should be designated based on chemical and physical criteria that comprise a mixture of laboratory analytical chemistry, field chemistry, visual-based physical habitat and riparian conditions, and land use.

3. A distinction should be made between variability of benthic macroinvertebrate assemblage due to anthropogenic stresses and natural variability in community composition by employing different analytical techniques such as clustering and ordination.

4. Multiple indices should be used for biological water quality monitoring. Indices, which reflect environmental change of interest and do not supply duplicate information, should be included in this array of bioassessment indices.

5. With respect to biotic indices like BMWP score system, local taxa should be incorporated in this system as well as sensitivity score should be modified by taking into account the local conditions.

6. A water quality classification scheme should be developed by incorporating the results of selected indices and this scheme may be useful for understanding the conditions and trends in water quality by non-technical people and decision makers.

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Physicochemical Results **APPENDIX-1**

Table 1.1

Location	Distance Downstream (km)	Temperature (°C) Wet	Temperature (°C) Dry	pH Wet	pH Dry	Conductivity (µScm-1) Wet	Conductivity (µScm-1) Dry	DO (mgl-1) Wet	DO (mgl-1) Dry
Pubail	0	27.1	24.7	7.1	7.0	82	74	6.2	4.8
Udail	1.5	27.1	24.4	7.0	7.1	82	78	5.6	4.1
Raidah	4.5	26.5	24.8	7.2	7.1	71	74	5.1	4.2
Bindhain	6	26.5	24.4	7.1	7.1	71	74	5.1	4.0
Trimohoni	7	26.5	24.5	7.1	7.1	69	48	5.5	4.1
Fakirkhali	21	29.6	24.8	7.5	7.4	71	73	4.8	3.2
Kaikpara	22	29.6	24.8	7.2	7.4	82	83	4.8	3.8
Norikhal	24	29.6	24.8	7.2	7.1	208	119	0.6	0.1
West Chanpara	26	29.6	25.8	7.6	7.5	79	103	4.7	2.2
Noripur	28	30.6	25.5	7.5	7.5	116	104	3.7	2.5
East Chanpara	29	30.5	26.0	7.3	7.3	112	105	3.6	2.8
Demraghat	30	30.9	26.1	7.3	7.3	115	125	2.9	0.8

Table 1.2

	Location	Distance Downstream (km)	BOD5 (mgl-1) Wet	BOD5 (mgl-1) Dry	NH_4-N (mgl-1) Wet	NH_4-N (mgl-1) Dry	NO3-N (mgl-1) Wet	NO3-N (mgl-1) Dry	PO4-P (mgl-1) Wet	PO4-P (mgl-1) Dry
1	Pubail	0	1.48	1.5	0.03	0.01	<0.23	<0.23	0.91	0.78
2	Udail	1.5	1.5	1.5	0.03	0.02	<0.23	<0.23	0.25	0.35
3	Raidah	4.5	1.8	3.5	0.02	0.03	<0.23	<0.23	0.13	0.05
4	Bindhain	6	1.5	3.5	0.03	0.04	<0.23	<0.23	0.24	0.06
5	Trimohoni	7	1.8	5.5	0.02	0.12	<0.23	<0.23	0.25	0.72
6	Fakirkhali	21	2.9	7.5	0.07	0.15	<0.23	<0.23	0.05	0.69
7	Kaikpara	22	3.6	6.5	0.12	0.34	<0.23	<0.23	0.08	0.46
8	Norikhal	24	3.7	30.5	2.83	11.09	0.76	0.23	0.44	1.07
9	West Chanpara	26	3.4	10.5	0.13	2.18	<0.23	<0.23	0.38	0.37
10	Noripur	28	5.4	9.5	0.79	3.95	0.40	0.23	0.15	0.22
11	East Chanpara	29	2.9	8.5	0.97	3.88	<0.23	<0.23	0.16	0.29
12	Demraghat	30	1.7	9.5	0.61	3.90	0.30	<0.23	0.45	0.81

APPENDIX-2

Appendix 2A: Biological results

No.	Sampling site	No. of Taxa	BMWP score	BMWP-ASPT	SCI	CLIPubail as ref.	CLIRaidah as ref.
1	Pubail	24	109	5.45	0.76	0.00	0.13
2	Udail	22	78	4.88	0.73	0.23	0.23
3	Raidah	24	106	5.58	0.89	0.13	0.00
4	Bindhain	20	67	4.79	0.74	0.35	0.30
5	Trimohoni	17	57	4.75	0.79	0.59	0.41
6	Fakirkhali	16	40	4.00	0.73	0.69	0.56
7	Kaikpara	13	32	3.56	0.76	0.85	0.85
8	Norikhal	10	25	3.13	0.72	1.60	1.50
9	West Chanpara	14	36	3.60	0.75	1.00	0.93
10	Noripur	14	34	3.78	0.76	0.93	0.86
11	East Chanpara	14	32	3.20	0.77	0.86	0.79
12	Demraghat	11	25	3.57	0.72	1.36	1.27

Appendix 2B: PC1 axis values of the Principal component analysis

Sampling site		BOD_5	NH_4-N	PO_4-P	DO	Conductivity	PC1 axis
No.	Name	(Log_{10})	(Log_{10})	(Log_{10})	(Log_{10})	(Log_{10})	scores
1	Pubail	0.18	-2.00	-0.11	0.68	1.87	-1.032
2	Udail	0.18	-1.70	-0.46	0.61	1.89	-0.982
3	Raidah	0.54	-1.52	-1.30	0.62	1.87	-0.993
4	Bindhain	0.54	-1.40	-1.22	0.60	1.87	-0.923
5	Trimohoni	0.74	-0.92	-0.14	0.61	1.68	-0.693
6	Fakirkhali	0.88	-0.82	-0.16	0.51	1.86	-0.143
7	Kaikpara	0.81	-0.47	-0.34	0.58	1.92	-0.08
8	Norikhal	1.48	1.04	0.03	-1.00	2.08	2.119
9	W.Chanpara	1.02	0.34	-0.43	0.34	2.01	0.576
10	Noripur	0.98	0.60	-0.66	0.40	2.02	0.516
11	E.Chanpara	0.93	0.59	-0.54	0.45	2.02	0.499
12	Demraghat	0.98	0.59	-0.09	-0.10	2.10	1.135

APPENDIX-3

Field Protocol

1. Name of the Water body: Balu 2. Location:

3. Date:

4. Average depth (m):

1	2	3	4	5	6	Mean

5. Approx. width (m): 6. Cross-sectional area (m^2):

7. Length of stretch: (m):

Replicate	1	2	3	Mean flow
Travel time (s)				
Velocity (ms^{-1})				
Flow (m^3s^{-1})				

8. Substrate characteristics: Sand ○ Silt ○ Clay ○ Detritus ○ Plant ○

9. Surrounding land use: Urban ○ Arable ○ Grazing ○ Forest ○ Others ○

10. Macrophyte cover: ----------------%

11. Riparian vegetation:----------------------

12. Algae:

13. Shading: Nil ○ Moderate ○ Heavy ○

14. Type of Pollution: i. Point sources
    ii. Non-point sources

APPENDIX-3 (Continued)

LOCAL MEASUREMENTS

1. Sample location:
2. Sampling date/analysis date:

Parameter	Unit	Measured values			Mean values
		1	2	3	
Temperature	^oC				
PH					
DO	mgl^{-1}				
EC	$(mgl^{-1}$				
NH_4-N	mgl^{-1}				
NO_3-N	mgl^{-1}				
PO_4-P	mgl^{-1}				

Biological water Quality Datasheet **APPENDIX-3** (Continued)

1. Sampling location:
2. Sampling date:

Taxonomic group	BMWP score	Taxa found	BMWP taxa score	Taxonomic group	BMWP score	Taxa found	BMWP taxa score	Taxonomic group	BMWP score	Taxa found	BMWP taxa score	Taxonomic group	BMWP score	Taxa found	BMWP taxa score
Key1: Cased Caddisflies				**Key 2: Stoneflies**				**Key 3: Waterbugs**				**Key 4: Flatworms**			
Beraeidae	10			Leuctridae	10			Aphelocheridae	10			Planariidae	5		
Glossosomatidae	7			Nemouridae	7			Corixidae	5			Scirtidae	5		
Odontoceridae	10			Perlodidae	10			Gerridae	5			Dendrocoelidae	5		
Goeridae	10			Taeniopterygidae	10			Hydrometridae	5			**Key 4: Leeches**			
Hydroptillidae	6			Capniidae	10			Mesovellidae	5			Erpobdellidae	3		
Lepidostomatideae	10			Chloroperlidae	10			Naucoridae	5			Glossiphonidae	3		
Limnephillidae	7			**Key 2: Mayflies**				Nepidae	5			Hirudidae	3		
Molanmidae	10			Baetidae	4			Notonectidae	5			Piscicolidae	4		
Phryganeidae	10			Caenidae	7			Pleidae	5			**Key 4: Snails**			
Sericostomatidae	10			Ecdyonuridae	10			Belostomatidae	5			Ancylidae	6		
Key 1: Caseless Caddisflies				Ephemerellidae	10			**Key 3: Water beetles**				Hydrobiidae	3		
Hydropsychidae	5			Ephemeridae	10			Dryopidae	5			Lymnaeidae	3		
Philopotamiidae	8			Leptophlebiidae	10			Dytiscidae	5			Neritidae	6		
Polycentropidae	7			Potamanthidae	10			Elminthidae	5			Physidae	3		
Phycophilidae	7			Heptageniidae	10			Gyrinidae	5			Planorbidae	3		
Psychomydae	8			Siphlonuridae	10			Haliplidae	5			Valvatidae	3		
Key 1: Alderflies	4			**Key 2: Dragonflies**				Hydrophillidae	5			Viviparidae	6		
Key1: Shrimps	6			Aeshnidae	8			Hygrobiidae	5			Piliadae			
Key 1: Hoglice	3			Corduliidae	8			Beelte larvae	5			Bithynidae			
Palaemonidae				Gomphidae	8			Key4:Blackflies	5			Thiaridae			
Alpheidae				Libellulidae	8			Key4:Craneflies	5			**Key 4: Mussels**			
				Key 2: Damselflies				Key 4: Midges	2			Unionidae	6		
				Agriidae	8			Key 4: Worms	1			Sphaeridae	3		
				Coenagriidae	6			Bristle Worms				**Subtotal taxa**			
				Lestiidae	8							**Subtotal BMWP score**			
				Corophiidae	6							Total no. of taxa			
				Platycnemidae	6							Total no. of BMWP taxa (a)			
Subtotal taxa				**Subtotal taxa**				**Subtotal taxa**				Total BMWP score (b)			
Subtotal BMWP score				**Subtotal BMWP score**				**Subtotal BMWP score**				ASPT score (b/a)			

APPENDIX-3 (Continued)

Sequential Comparison Index Calculation Sheet

1. Sampling location:
2. Sampling date:

NUMBER	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	TOTAL RUNS	TOTAL ORGS	MOVING SCI
1																	15	
2																	30	
3																	45	
4																	60	
5																	75	
6																	90	
7																	105	
8																	120	
9																	135	
10																	150	

ANALYTICAL RESULT SEETS FOR BOD_5 (mgl^{-1})

1. Sample code:
2. Sampling location:
3. Sampling date:

Day	Date	Measured values (DO, mg/l)			Mean value
		1	2	3	
Day 0					D_1
Day 5					D_2

Bank Sample

Day	Date	Measured values (DO, mg/l)			Mean value
		1	2	3	
Day 0					B_1
Day 5					B_2

$$\text{BOD (mg/l)} = \frac{(D_1 - D_2) - (B_1 - B_2)}{\%\ \text{sample}} \text{X}100$$

APPENDIX-4

Results of paired *t*-test for physicochemical parameters (p = 0.05)

Appendix -4A

	Temperature		pH		Conductivity	
	Wet season	Dry season	Wet season	Dry season	Wet season	Dry season
Mean	28.675	25.05	7.258333333	7.241666667	96.5	88.33333333
Variance	3.142045455	0.39	0.035378788	0.031742424	1549	512.4242424
Observations	12	12	12	12	12	12
Pearson Correlation	0.793726057		0.870352694		0.72151873	
Hypothesized Mean Difference	0		0		0	
df	11		11		11	
t Stat	9.425996209		0.615881762		1.015696504	
P(T<=t) one-tail	6.65129E-07		0.275251858		0.165797002	
t Critical one-tail	1.795883691		1.795883691		1.795883691	
P(T<=t) two-tail	1.330E-06		0.550503716		0.331594004	
t Critical two-tail	2.200986273		2.200986273		2.200986273	

Appendix- 4B

	DO		BOD_5		NH_4-N	
	Wet season	Dry season	Wet season	Dry season	Wet season	Dry season
Mean	4.383333333	3.05	2.638333333	8.166666667	0.470833333	2.1425
Variance	2.281515152	2.084545455	1.50359697	59.15151515	0.665953788	10.7890204 5
Observations	12	12	12	12	12	12
Pearson Correlation	0.930014076		0.509098991		0.97955464	
Hypothesized Mean Difference	0		0		0	
df	11		11		11	
t Stat	8.299671085		-2.68025144		-2.324972828	
P(T<=t) one-tail	2.29847E-06		0.01069929		0.020111931	
t Critical one-tail	1.795883691		1.795883691		1.795883691	
P(T<=t) two-tail	4.59694E-06		0.02139858		0.040223862	
t Critical two-tail	2.200986273		2.200986273		2.200986273	

Appendix- 4C

	PO_4-P	
	Wet season	Dry season
Mean	0.290833333	0.489166667
Variance	0.055462879	0.103826515
Observations	12	12
Pearson Correlation	0.500289201	
Hypothesized Mean Difference	0	
df	11	
t Stat	-2.379610641	
P(T<=t) one-tail	0.018263116	
t Critical one-tail	1.795883691	
P(T<=t) two-tail	0.036526232	
Critical two-tail	2.200986273	