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Aquatic insects as bio-indicator of water quality- A study on Bakuamari stream, Chakras hila Wildlife Sanctuary, Assam, North East India

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Abstract

The Chakrashila Wildlife Sanctuary (latitude 26° 15' - 26° 26' N and longitude 90° 15' - 90° 20' E) is located in western Assam. This study is a part of biomonitoring programme of the sanctuary using aquatic insects of Bakuamari stream during 2011- 2012. The study revealed presence of 21 species of aquatic insects belonging to 14 families and 7 orders. Different diversity indices were worked out in all the seasons. The environmental variables of water were estimated by standard methods. Pearson's Correlation Coefficient was computed to find out significant correlations between diversity and density of aquatic insects with the water variables. Biomonitoring Working Party score and the Average Score per Taxon were calculated to find the status of the water quality of the system. Canonical correspondence analysis (CCA) was used to infer the relationship between environmental variables and aquatic insect community structure. The influence of environmental variables in the diversity of aquatic insects has been discussed in the paper.

Keywords: Aquatic insect, Diversity, Dominance, Stream, Water quality

1. Introduction

Aquatic insects, an important component of aquatic ecosystems are very abundant and diverse group that inhabits a variety of aquatic environments ^[1]. They play an important role in ecosystem functioning ^[2, 3] and are used as bioindicator. These bioindicators have the advantage of monitoring anthropogenic stress of an ecosystem over a long period of time ^[4]. The north-east Indian biogeographic zone is most significant as it represents the transition zone between the Indian, Indo-Malay and Indo-Chinese biogeographic regions, as well as a meeting-place of Himalayan Mountains with those of Peninsular India ^[5]. In north east India studies on aquatic insect community are scanty in lotic systems and most of the studies were done in lentic systems ^[6-17]. Aquatic insect diversity of protected area and their role as bioindicator has not been studied so far. This study investigated the aquatic insect community, their distribution in different seasons and their role as bioindicator of water of the Bakuamari stream. The study of aquatic insect community and water quality of streams of Chakrashila Wildlife Sanctuary (CWS) is of great importance as the Wildlife Sanctuary falls on Eastern Himalayan region and in the biodiversity hotspot Himalaya ^[18] which is a data deficient region ^[19]. More over the Wildlife Sanctuary is introduced as a Key Biodiversity Area (KBA) ^[18].

2. Materials and Methods

Chakras hila Wildlife Sanctuary (CWS) (26° 15' to 26° 26' N and 90° 15' to 90° 20' E), located in the Dhubri and Kokrajhar district of Assam, North East India is the second home of Golden Langur (*Trachypithecus geei*) which is an endangered species, in the foothills of Himalaya. CWS has dry winter and hot summer followed by heavy rainfall. The diverse eco-systems of CWS support various mammalian species. Bakuamari (26° 21' N and 90° 19' E) is one of the perennial streams of the Wildlife Sanctuary (Fig. 1). Aquatic insects and water samples in three replicates were collected seasonally from different stretches of the stream during 2011-12 by three different methods such as "all out search" method, "a nylon pond net" method ^[20] and "1 minute kick" method ^[21] according to the habitat characteristics. Three such drags constituted a sample. The insects were sorted, counted and identified by using standard keys ^[22-34]. Relative abundance of insects in family level and species level were calculated.

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Rainfall (RF) data were collected from metrological station of Choibari tea estate, Kokrajhar, Assam. Water temperature (WT), discharge, pH, electrical conductivity (EC), dissolved oxygen (DO), total alkalinity (TA), free CO₂, nitrate, and phosphate were estimated by standard methods [35, 36]. Diversity indices like Margaleff Index (M), Shannon –Wiener Index (H'), Evenness Index (J), and Berger Parker Index of Dominance (d) for the insect community were worked out using the package Biodiversity Professional Version 2 for Windows 1997 (The Natural History Museum and Scottish Association for Marine Science). SIGNAL (Stream Invertebrate Grade Number- Average Level), a family-level water pollution index based on the known tolerances of aquatic macro-invertebrate families to various pollutants was worked out by the standard method [37]. BMWP (Biological Monitoring Working Party) and ASPT (Average Score Per Taxon) scores were computed by standard methods [38]. Statistical analyses, correlation coefficient analysis between environmental variables and species variables were done by using package SPSS 20.0 for Windows 7. Canonical Correspondence analysis (CCA) was done to see the relationship of insect abundance to environmental variables in the four seasons using CANOCO package for windows 4.5 [39].

3. Results

3.1 Distribution and Abundance of Insects

The study revealed presence of 21 species of aquatic insects belonging to 14 families and 7 orders (Table 1) (Plate 1). The orders are Hemiptera, Coleoptera, Trichoptera, Ephemeroptera, Odonata, Collembola and Diptera. In Pre-monsoon season 11 species belonging to four orders (Hemiptera, Coleoptera, Ephemeroptera and Trichoptera) were encountered while six species of two orders (Hemiptera and

Coleoptera) and seven species of three orders (Hemiptera, Coleoptera and Odonata) were recorded in Monsoon and Post-monsoon, respectively. In Winter highest number of species i.e., 12 species of six orders (Hemiptera, Coleoptera, Ephemeroptera, Trichoptera, Collembola, and Diptera) were recorded.

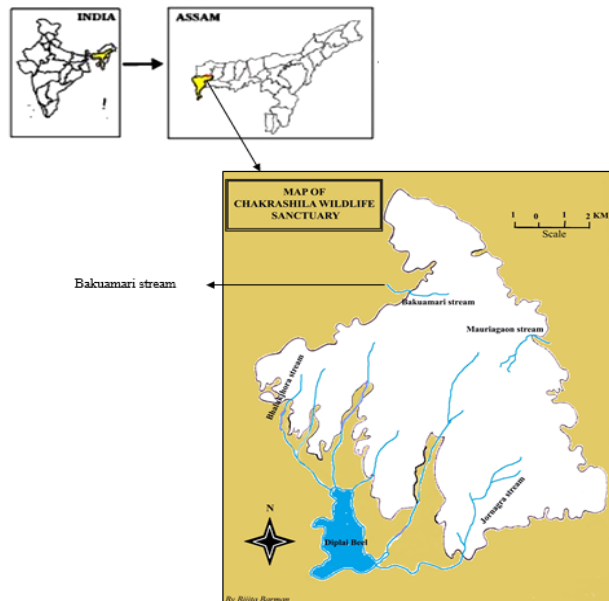


Fig 1: Map of India followed by Map of Assam showing the position of Chakrashila Wildlife Sanctuary followed by Chakrashila Wildlife Sanctuary Map showing the Bakuamari Stream

Table 1: Seasonal distribution of different species of aquatic insects in Bakuamari stream

z	Family	Taxa	Status			
			Pre- Monsoon	Monsoon	Post -Monsoon	Winter
Hemiptera	Gerridae	<i>Ptilomera assamensis</i>	+	+	+	+
		<i>Pleciobates expositus</i>	+	+	-	-
		<i>Metrocoris nigrofascioides</i>	+	+	+	+
		<i>Ovatametra gualeguay</i>	-	-	+	-
	Aradidae	<i>Notapictinus aurivilli</i>	+	-	-	-
	Mesoveliidae	<i>Mesovelia vittigera</i>	-	+	-	+
Coleoptera	Gyrinidae	<i>Rhagovelia obesa</i>	+	+	+	+
		<i>Orectogyrus sp.</i>	+	+	+	+
	Circulionidae	<i>Dineutus sp.</i>	+	-	-	-
		<i>Bagous affinis</i>	+	-	-	-
Trichoptera	Noteridae	<i>Hydrocanthus oblongus</i>	-	-	+	-
	Hydroptilidae	<i>Hydroptila sp.</i>	+	-	-	-
Ephemeroptera	Hydropsychidae	<i>Hydropsyche bidens</i>	-	-	-	+
		<i>Labeobaetis sp.</i>	+	-	-	-
	Baetidae	<i>Offadens sp.</i>	+	-	-	+
		<i>Stenonema sp.</i>	-	-	-	+
Odonata	Gomphidae	<i>Heptagenia sp.</i>	-	-	-	+
		<i>Erpetogomphus sp.</i>	-	-	+	-
Collembola	Entomobryidae	<i>Entomobrya nivalis</i>	-	-	-	+
		<i>E. sp.</i>	-	-	-	+
Diptera	Dixidae	<i>Nothodixa sp.</i>	-	-	-	+

In Pre-monsoon and Monsoon the species, *Ptilomera assamensis*, *Pleciobates expositus* and *Metrocoris nigrofascioides* belonging to the family Gerridae (Hemiptera) were found to be most abundant followed by species *Dineutus sp.* and *Orectogyrus sp.* belonging to the family Gyrinidae (Coleoptera). Again in Post-monsoon and winter the

Rhagovelia obesa belonging to Veliidae was found to be most abundant (Fig. 2 and Fig. 3). *Ptilomera assamensis*, *Metrocoris nigrofascioides*, *Rhagovelia obesa* and *Orectogyrus sp.* were found in all the seasons during study period.

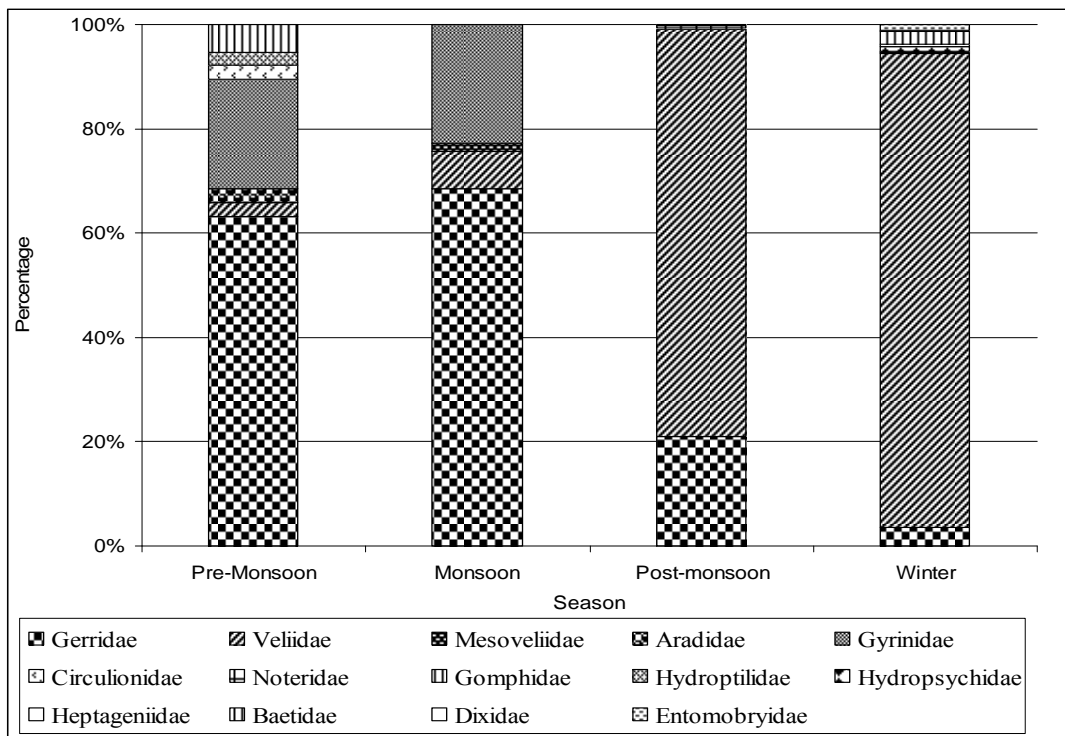


Fig 2: Seasonal variation in relative abundance of aquatic insect families in Bakuamari Stream

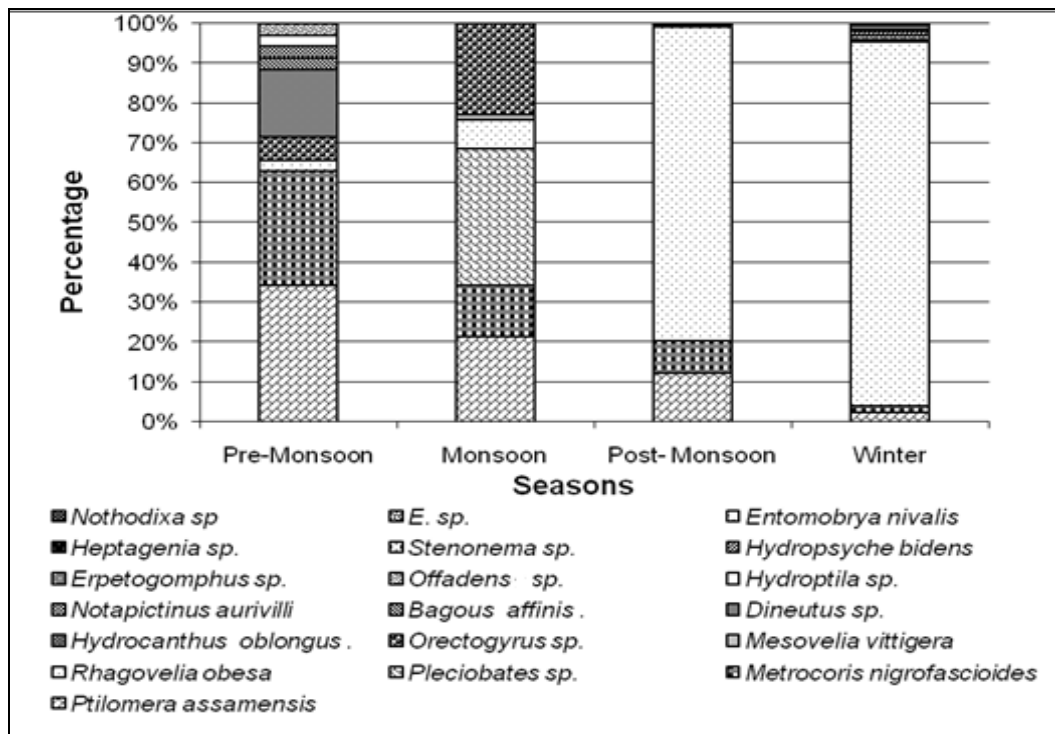


Fig 3: Seasonal variation in relative abundance of aquatic insect species in Bakuamari Stream

3.2 Diversity Indices

The Shannon-Wiener (H') (0.77) and Margaleff index (M) (8.35) values were found highest in Pre-monsoon with lowest Berger Parker Index of Dominance (d) (0.33). Lowest values of Shannon-Wiener (H') (0.3) and Evenness index (0.39) were recorded in Winter season with highest value of Berger Parker Index of Dominance (d) (0.79) (Table 2).

3.3 Biomonitoring Scores

The BMWP scores for the stream ranged between 25(Monsoon) to 44 (Winter) (Table 2). The ASPT score of the study site was found in the range of 5.83 (Pre-monsoon) to 7 (Post-monsoon). Highest SIGNAL value was found in Winter (4.22) and lowest was found in Monsoon season (3.67) (Table 2).

Table 2: Seasonal variation in diversity indices and Biomonitoring Scores of Bakuamari stream

Seasons	Margaleff M Base 10	Shannon H' Log Base 10.	Evenness'(J)	Berger-Parker Dominance (d)	BMWP score	ASPT score	SIGNAL Score
Pre-monsoon	8.353	0.772	0.772	0.333	35	5.83	3.83
Monsoon	7.046	0.672	0.864	0.343	25	6.25	3.67
Post- monsoon	5.13	0.302	0.388	0.789	35	7	3.69
Winter	5.625	0.227	0.204	0.903	44	6.29	4.22

BMWP score: 0-16=Poor water quality; 17-50=Moderate water quality; 51-100=Good water quality; 101-150=High water quality; 151+=Very high water quality [64].

ASPT score: >6= Clean water, 5-6= Doubtful quality, 4-5 = Probable moderate pollution, <4 = Probable severe pollution [57].

SIGNAL Score: Greater than 6= Healthy habitat, between 5 and 6= Mild pollution, between 4 and 5=Moderate Pollution, Less than 4=Severe pollution [65].

3.4 Physico-Chemical Properties of Water

The environmental variables of water were estimated in Pre-monsoon, Monsoon, Post-monsoon and Winter seasons (Table 3). The pH did not show much fluctuation ranging from 6.22 in Post-monsoon to 6.53 in Pre-monsoon. Water temperature fluctuated from 10.07 °C (Winter) to 24.29 °C (Pre-monsoon). Highest discharge was recorded in Pre-monsoon (27000 inch³.S⁻¹) and lowest in Monsoon (8356 inch³.S⁻¹). EC in all samples were observed in the range of 39.59 µS.Cm⁻¹ in Winter to 41.83 µS.Cm⁻¹ in Monsoon. The TA of stream water in the selected sites ranged from 25.8 in Winter to 42.56 mg.L⁻¹

¹ in Post-monsoon. The range of both phosphate and nitrate concentration in water samples of stream was 0.06 - 0.16 mg.L⁻¹ and 0.05-0.64 mg.L⁻¹, respectively. The DO values in different sites were good ranging from 6.63-12.17 mg.L⁻¹. Positive significant correlations of species richness of insects with DO in Pre-monsoon (r=0.85, p<0.01 with df=8) and Post-monsoon (r=0.91, p<0.01 with df=8) were found. Positive significant correlation was also found between species richness of insects and nitrate (r=0.78, p<0.01 with df=8). The density of insects also showed significant positive correlation with pH, EC, DO and nitrate (Table 4).

Table 3: Seasonal variations in environmental variables of water of stream Bakuamari (n=9)

Environ-mental Variables	Rainfall (cm)	AT (°C)	WT (°C)	Discharge (inch ³ .S ⁻¹)	DO (mg.L ⁻¹)	Free-CO ₂ (mg.L ⁻¹)	TA (mg.L ⁻¹)	pH	EC (µS.cm ⁻¹)	Phosphate (mg.L ⁻¹)	Nitrate (mg.L ⁻¹)
Pre-monsoon	7.94 ±0.053	26.50 ±0.6	24.29 ±0.4	26444.00 ±94.9	15.80 ±0.2	10.56 ±0.51	39.00 ±0.5	6.53 ±0.2	43.90 ±2.6	0.29 ±0.04	0.30 ±0.03
Monsoon	19.00 ±0.09	28.20 ±0.56	23.78 ±0.7	15825.67 ±73.82	13.61 ±1.57	8.11 ±0.19	35.44 ±4.03	6.44 ±0.15	39.17 ±3.32	0.02 ±0.003	0.19 ±0.02
Post-monsoon	6.98 ±0.01	20.70 ±0.65	19.00 ±1	15348.33 ±43.6	13.87 ±0.7	6.67 ±0.76	41.00 ±3.04	6.22 ±0.1	41.50 ±4.14	0.05 ±0.02	0.25 ±0.1
Winter	0.00	16.20 ±0.7	10.07 ±0.06	15225.00 ±87.08	12.73 ±0.05	5.83 ±0.3	26.17 ±0.58	6.39 ±0.04	44.50 ±5.33	0.17 ±0.06	0.23 ±0.1

(AT=Air temperature, WT=Water temperature, DO=Dissolved oxygen, TA=Total alkalinity, EC=Electrical conductivity)

Table 4: Significant Pearson’s correlations of species richness and density of insects with different environmental variables of water of the stream Bakuamari

Water parameters	Seasons	pH	Electrical Conductivity (µScm ⁻¹)	Dissolved oxygen (mgL ⁻¹)	Nitrate (mgL ⁻¹)
Correlation with Species richness	Pre-Monsoon	NS	NS	0.847**	NS
	Post-Monsoon	NS	NS	0.907**	0.783*
Correlation with Density	Pre-Monsoon	0.740*	NS	NS	NS
	Monsoon	-0.768*	0.766*	NS	NS
	Post-Monsoon	NS	0.724*	0.697*	0.755*

** , Correlation is significant at the 0.01 level (2-tailed). * , Correlation is significant at the 0.05 level (2-tailed)

Canonical Correspondence Analysis based on relative abundance of macroinvertebrates explained 63.6% of variance within the dataset (cumulative percentage variance of species data, first two CCA axes) (Table 5). The two axes were found significant (p<0.05, variable 1; F-ratio= 1.51; number of permutations= 9999) as revealed by Monte Carlo test (Table 6). In the present study the sum of all Canonical Eigenvalues was found 0.93. Association of insect species with the environmental variables in different seasons as revealed by CCA is displayed in Fig. 4.

Table 5: Eigen values from CCA of the aquatic insect community and environmental variables data

	Axis 1	Axis 2	Axis 3	Total
Eigen values	0.415	0.335	0.177	0.927
Cum. Percentage variance of species data	44.7	80.9	100	
of species-environment relation	44.7	80.9	100	
Spec.-env. correlations	1.000	1.000	1.000	

Table 6: Monte-Carlo test of significance of first canonical axis: Eigenvalue, test of significance of all canonical axes: Trace and P-value

Test of significance of first canonical axis: Eigenvalue	0.415
P-value	0.045
Test of significance of all canonical axes: Trace	0.927
P-value	0.293

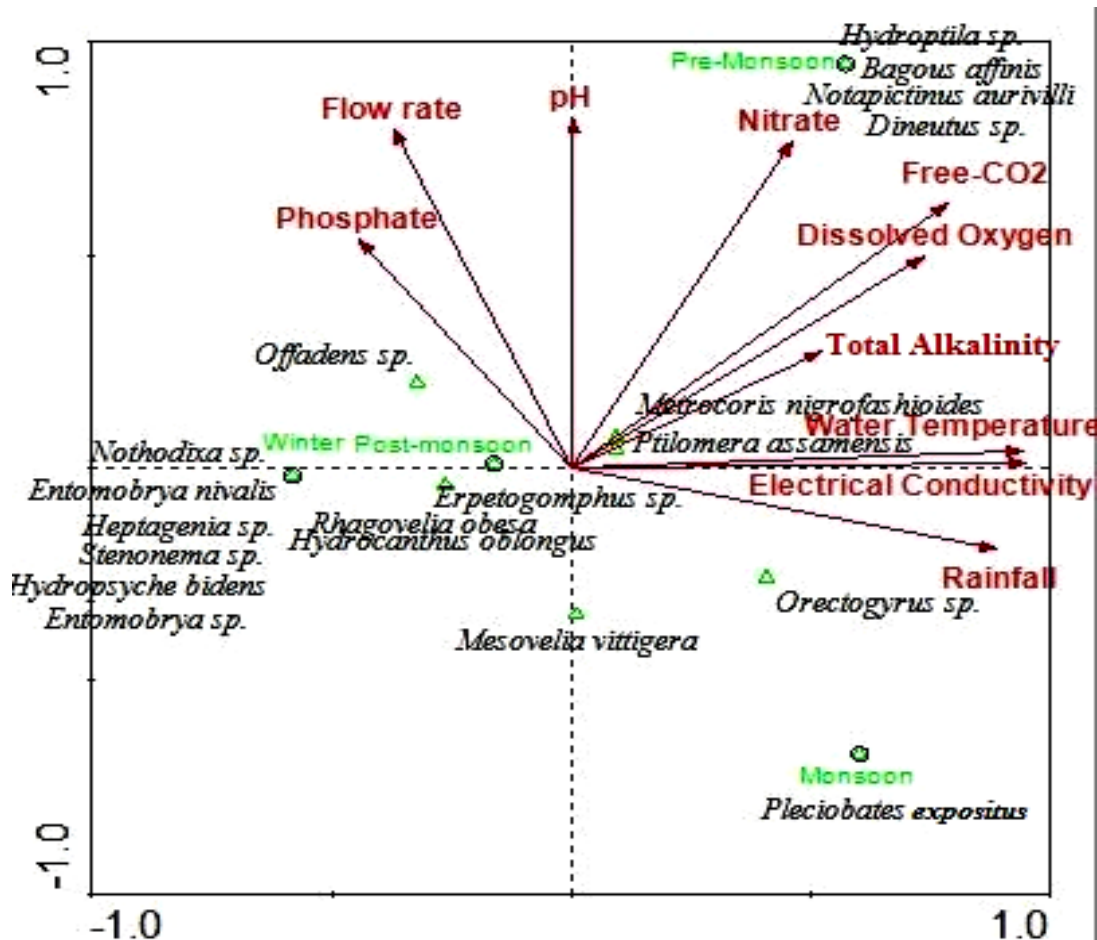


Fig 4: Triplot of a Canonical Correspondence Analysis (CCA) showing association among different species of aquatic insects (green triangles ▲), environmental variables (arrow) and four seasons (black circles O). Relationships between variables or similarity of samples are indicated by close clusters of points

4. Discussion

The use of living organisms for monitoring water quality originated in Europe and it is widely used throughout the world [40, 41]. Klemm *et al.* [42] developed methods and used macroinvertebrates as indicators of ecological conditions for streams in the Mid-Atlantic Highlands region. Experiences from USA and European programme have demonstrated that benthic macroinvertebrates are most useful in monitoring freshwater ecosystems [4, 43].

In the present study the order Hemiptera was found most diverse and relatively abundant in the stream. A study in the lower reach of Moirang River in Manipur, N.E. India also showed high Hemiptera diversity and density [44]. According to Huang *et al.* (2010) in Du river basin in northern Vietnam also Hemiptera was found to be the most diverse order [45]. Selvakumar *et al.* [46] suggested that change in community structure is mainly due to changes in the geomorphology and the associated destruction of in-stream physical habitats. The relatively more abundance or dominance of Gerridae and Veliidae of order Hemiptera in the stream could be due to their modified body structure. These known pond skaters who stay on the surface of water can walk on the surface of water and can utilize atmospheric variables without totally depending on water. The dominance of Gerridae and Veliidae also has been recorded by Naranjo *et al.* [47] in their study of aquatic and semiaquatic Heteroptera (Insecta) in high altitudinal stream systems of Cuba. The causes of fluctuations in insect abundance, dominance and distribution include macroclimatic and microclimatic changes and variation in the availability of

food resources [48-54].

Diversity index is a statistical method which is planned to evaluate the variety of a data group consisting of different types of components. Features of a population such as number of existing species (Richness), distribution of individuals equally (Evenness) and total number of existing individuals underlie the basis of diversity indices [55-56]. Thus, any changes in any of these three features will affect the whole population, so that the diversity indices depending upon these features are used effectively to determine the changes in a population [57]. Diversity index can also be used to measure environmental stress [58]. Shannon- Wiener diversity index (H') values were found to be less than 1 in all the seasons indicating polluted nature of stream water [59]. In Pre-monsoon the stream water was relatively good with more diverse taxa encountering highest Shannon H' (0.772), where as in winter the system was assembled by more dominant groups encountering highest Berger-Parker index of Dominance (0.903). Iwasaki [60] said that environmental stability rather than spatial heterogeneity has greater influence on H'. However, Margaleff's water quality index has no limit value and it shows a variation depending upon the number of species. Thus, it is used for comparison of the sites [61]. In the present study the values of Margaleff's water quality index were found more than 5. According to Lenat *et al.* [62] Margaleff's water quality index values greater than 3 indicate clean conditions; values less than 1 indicate severe pollution and intermediate value indicate moderate pollution.

The overall BMWP Score for a site is the sum of all of the scor

es of each family present at that site. Score values for individual families reflect their pollution tolerance based on the current knowledge of distribution and abundance. Pollution intolerant families have high BMWP scores, while pollution tolerant families have low scores [63]. The range of BMWP indicated a moderate water quality [64]. The Average Score Per Taxon (ASPT) represents the average tolerance score of all taxa within the community, and is calculated by dividing the BMWP by the number of families represented in the sample. A high ASPT usually characterizes clean sites with relatively large number of high scoring taxa i.e., approximate 6 ASPT values meant that the system supports some high scoring taxa, i.e., good quality of system [63].

The SIGNAL value between 4- 5 indicate moderate system [65]. Highest SIGNAL score (4.22) was recorded in Winter. Occurrence of Ephemeroptera group (pollution intolerant group) in Winter season might be the reason for moderate good score of BMWP and SIGNAL in that season as some taxa of Baetidae and Heptageniidae are the best bioindicators of agricultural impact and indicate pristine forest conditions with intact riparian vegetation and a high percentage of marginal vegetation [46]. These mayflies show a strong preference for a rocky substratum and marginal vegetation, and they use plant material for their food. Heptageniidae is filter-feeders/scrapers and generally prefer a 'clean' cobble substratum for attachment and high to turbulent flow to facilitate filtering [66]. Several factors are known to influence the distribution of aquatic macroinvertebrates, but the important factors likely to affect the diversity and abundance in an aquatic ecosystem, are water temperature, water velocity, nutrient availability, etc. Ward and Stanford also suggested that water flow, temperature and substrates are the major factors determining the composition and abundance of benthic invertebrates [67].

The range of pH between 6.5 and 8.5 is normally acceptable as per BIS [68] and WHO [69]. The water was found slightly acidic in Post-monsoon with pH 6.22. Popoola and Otalekor [70] also recorded pH range of 6.0-7.0 in Awba reservoir, Nigeria. Although pH usually has no direct impact on consumers, it is one of the most important operational water quality parameters [71]. The EC values are an index to represent the concentration of soluble salts in water. High flow events have been identified in many studies to greatly reduce the biomass and change the species composition of invertebrates in aquatic ecosystems. According to Minshall benthic invertebrates are particularly sensitive to different water velocities and bed sediment stability [72]. Alkalinity measures the various substances related to the basic property of water and high TA value is associated with poor quality of water. The range of TA was found within the desirable limit of drinking water according to Indian Standard Specifications for Drinking Water (IS: 10500-1992) [73]. Phosphorus and nitrogen are the basic nutrients which influence productivity of aquatic ecosystems. The different sources of phosphorus are fertilizers, pesticides, industry and cleaning compounds. Natural sources include PO_4^- containing rocks and solid or liquid wastes. Phosphate values in the present study were found high particularly during Pre-monsoon and Monsoon. The range of both PO_4^- and NO_3^- in the present study indicated that input of pollutants in the system is either very low. In general the natural stream water have DO in the range between 5-13 mgL^{-1} and DO within 7-11 mgL^{-1} is very good for most stream fishes [74, 75]. In the present study the concentration of DO was found more than 12 mgL^{-1} in different seasons. This justified the significant positive relationship of DO with species richness. A few more water variables like pH, EC, NO_3^- were found to have positive

significant correlations with species richness and density of insect. This conformed to the studies made by Zabbey and Hart in Woji creek and Arimoro *et al.* [76] in Ethiopie River in Niger delta [77].

Eigen values associated with each axis equal the correlation coefficient between species scores and site scores [78, 79]. Thus an Eigen value close to will represent a high degree of correspondence between species and sites, and an Eigenvalue close to zero will indicate very little correspondence [80]. In the present study the sum of all Canonical Eigen values was found 0.93 which indicated high degree of correspondence of species with seasons. The direction and length of each arrow in CCA ordination diagram indicate the direction and rate of maximum changes in each variable [79]. Except some species in Winter and Monsoon, most of the species were centered in the CCA ordination diagrams. Axis I of CCA can be described as a gradient of EC and WT. The CCA revealed a clear association of the aquatic insect species with EC/WT along Axis I. In Pre-monsoon season with high nitrate concentration, DO is located in the upper right quadrant, whereas Monsoon season with high rainfall and electrical conductivity is located in the lower right quadrant in Figure 4. The species like *Ptilomera assamensis*, *Metrocoris nigrofashioides* etc. were found strongly associated with WT, EC, TA, DO and F CO₂ of water. The increase of water temperature is highly correlated with development rate of the water striders [81]. The *Hydroptila* sp. (Trichoptera), *Bagous affinis*, *Dineutus* sp. (Coleoptera), *Notapictinus aurivilli* (Hemiptera) were found associated with Nitrate. Natural levels of nitrate are usually less than 1 mg L^{-1} and according to Sharon concentrations over 10 mgL^{-1} will have an effect on the freshwater aquatic environment [75]. In the present study concentration of nitrate was always less than 0.5 mgL^{-1} . So high DO and low nitrate might have favoured assemblage of Trichoptera which is known as pollution-sensitive group. Some species like *Pleciobates expositus*, *Orectogyrus* sp. were found positively correlated with high rainfall and EC in the lower right quadrant of CCA ordination graph.

5. Conclusions

The Bakuamari stream has been found to be a less diverse system as revealed by Shannon-Wiener diversity index and Evenness index whereas according to Margaleff index of diversity, water quality of the stream is good. This is confirmed by high DO of water and positive relationship of species richness with DO. The BMWP scores of the stream are in the moderate range (25-44) while ASPT and SIGNAL scores are in the range of moderate to good quality of water. According to Hilsenhoff [82] small streams typically have lower diversity values than larger streams in similar habitats with similar substrates, which may lead to erroneous conclusions about water quality if evaluated with diversity. Hence a long term monitoring programme and use of variety of diversity and biotic indices might throw light on the health of the stream and might influence government policy of conservation of the small streams of protected as well as unprotected areas.

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Plate 1: Images of aquatic insects found in Bakuamari stream during study; 1- *Ptilomera assamensis* Amyot & Serville 1843, 2- *Metrocoris* sp. Chen & Nieser 1993, 3- *Rhagovelia obesa* Uhler 1871, 4- *Orectogyrus* sp., 5- *Entomobrya nivalis* Linnaeus 1758, 6- *Offadens* sp., 7- *Hydropsyche* sp. Ross 1938, 8- *Nothodixa* sp. Edwards 1930

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