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# Influence of water physicochemical characteristics on Simuliidae (Diptera) prevalence in some streams of Meghalaya, India

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## ABSTRACT

*Background & objectives:* Simuliids (Diptera) are one of the medically important biting insects group and have worldwide distribution. Their immature stages proliferate in fast flowing river or stream water and have been considered as important ecological indicator.

*Methods:* Aquatic stages of simuliids were collected and speciated from 16 different fresh water rivers and streams. Water flow rate was determined and water samples were analysed for various water variables such as water temperature, pH, dissolved oxygen concentration (DOC), dissolved oxygen saturation (DOS), conductivity, total dissolved solute (TDS), turbidity, resistivity and salinity. Linear regression was used to determine relationship between simuliid density and water variables, whereas multiple regression was used to determine the fitness for the presence of simuliid species. Principal component analysis (PCA) was used to determine the water parameters association with simuliid distribution.

*Results:* Total 565 specimens comprising of three species namely, *Simulium (S) barraudi* Puri, *S. (S) striatum* Brunetti, and *S. (S) himalayense* Puri were recorded in the present study. *Simulium barraudi* was the most abundant (56.8%) and its density was high ($\chi^2 = 289.3$; df = 2; $p < 0.0001$) as compared to the others. The average population size of each species was 188.3, whereas Simpson and Shannon-Wiener diversity indices were 0.4466 and 1.306 respectively. Linear regression showed that simuliid density was associated with the water flow rate. Principal component analysis indicated that the water parameters accounted for 42.25% variation along D1 axis, while 24.1% variation along D2 axis. Atleast two principal components have eigenvalue >1 and accounted for 32.6% of variation.

*Interpretation & conclusion:* Our study provides new information on simuliid species association with breeding water parameters in a little studied region of high biological interest. Turbidity, water flow and pH are important water parameters affecting the simuliid species prevalence. Each simuliid species preferred different sets of physicochemical parameters of breeding habitat, which are specific to that particular species. Therefore, simuliid species community as a whole cannot be considered as a suitable indicator of the streams water quality. In addition to describing simuliids, the information provided herein will be useful for the conservation of aquatic ecology and environment in Meghalaya state of India.

**Key words** Density; immature stages; Meghalaya; simuliid; water parameters

## INTRODUCTION

Simuliids (blackflies) have a worldwide distribution and are important components of aquatic ecosystem. The immature stages of blackflies often predominate the fresh water invertebrate fauna of rivers and streams[1]. Larvae exhibit dynamic breeding ecology and have been reported from various watercourses ranging from small water channels to large streams. Simuliids are found attached to various substrates in the fresh water streams and exhibit peculiar breeding habitat selection. The data on prevalence and diversity of simuliids in India are sparse and about 74 species have been recorded, of which most of the species have been collected from Arunachal Pradesh and Asom states[2, 3].

Many biotic and abiotic factors have been found associated with the richness and variation in simuliids species composition at alpha and beta level[4, 5]. The variation in diversity might originate from variation in environmental factors among sites and from niche differences between the different species[6, 7]. Many environmental factors, such as water temperature, pH, dissolved oxygen concentration and turbidity have been known to affect blackflies diversity[1, 8] and many of these variables are used as predictors of species distribution[9, 10]. Blackfly larvae exploit the fine particulate organic matter in water for food requirement, therefore, enrichment of lotic systems has been shown to have significant effects on change in species composition, larval density and developmental rate[7].

Simuliids are medically important and transmit onchocerciasis (river blindness) in many countries of Africa and America, however, in India, simuliids have not been incriminated to be a vector of onchocerciasis. These flies have wide distribution in the northeastern states of India and known as serious pests causing biting nuisance and local allergic reactions on human[11, 12]. The study on various physicochemical characteristics of simuliids breeding water is imperative for comprehensive understanding of breeding ecology and prevalence of simuliid flies. Present study investigates the distribution and density of blackflies species in 16 different streams and rivers of Meghalaya state of northeastern India. The study also determined the relationship of different biotic and abiotic parameters with blackfly species density to obtain information about their environmental preferences at micro level.

## MATERIAL & METHODS

*Study area and simuliids identification*

The aquatic stages of simuliids were sampled from 16 different streams and rivers of Meghalaya state of northeastern India during January–March 2012. Meghalaya state covers a total land area of 10,435 km$^2$ (25°5′– 26°10′ N and 90°45′–92°15′ E) and has an altitude range of 400–1600 m above sea level. The region has sub-tropical monsoonic climate with moderately high mean temperature and very high mean relative humidity.

Table 1. Altitude and GPS position of study locations

| Location | Source | Altitude (m) | GPS position |
|---|---|---|---|
| Elephant falls | Stream | 1710 | N25°32′17.3″ E91°49′19.3″ |
| Umtyngngar | River | 1678 | N25°27′ 58.6″ E91°49′38.9″ |
| Sohra | Stream | 1467 | N25°17′34.2″ E91°42′56.3″ |
| Mawsmai | Stream | 1199 | N25°14′17.7″ E91°43′57.2″ |
| Mawjrong | River | 1742 | N25°27′04.1″ E91°49′05.4″ |
| Dawki | River | 52 | N25°11′23.6″ E92°01′09.7″ |
| Jorain | River | 1211 | N25°20′55.6″ E92°08′30.0″ |
| Sonapur | River | 99 | N25°06′25.2″ E92°21′40.5″ |
| Umtrew | River | 818 | N25°43′10.0″ E91°53′22.0″ |
| Umran | River | 727 | N25°46′22.5″ E91°52′26.9″ |
| Umsamlem | River | 661 | N25°47′48.4″ E91°52′26.0″ |
| Byrnihat | River | 154 | N26°02′22.8″ E91°52′02.6″ |
| Rari | Stream | 99 | N25°47′18.7″ E90°25′44.4″ |
| Chiokgre | River | 538 | N25°40′30.1″ E90°20′25.2″ |
| Warebok | River | 657 | N25°36′46.5″ E90°19′09.1″ |
| Rongkhon | River | 376 | N25°32′16.6″ E90°13′44.9″ |

The rainfall is excessive and distributed throughout most of the year. The details of collecting locations, type of breeding habitats, altitude and GPS positions have been depicted in Table 1. Most of the land is either under rain forest cover or mineral (coal and lime stone) rich barren and uncultivated (Fig. 1). The sampling locations were selected in accordance with their accessibility by roads and sampled from atleast three different places for 15 min each. Larvae and pupae were collected from all the available substrates present in the habitat. Meghalaya state has annual temperature ranging from 4 to 35°C. The state experiences high rainfall and therefore characterized by the presence of numerous annual rivers and streams (Fig. 1). Larvae and pupae collected were stored in 1:3

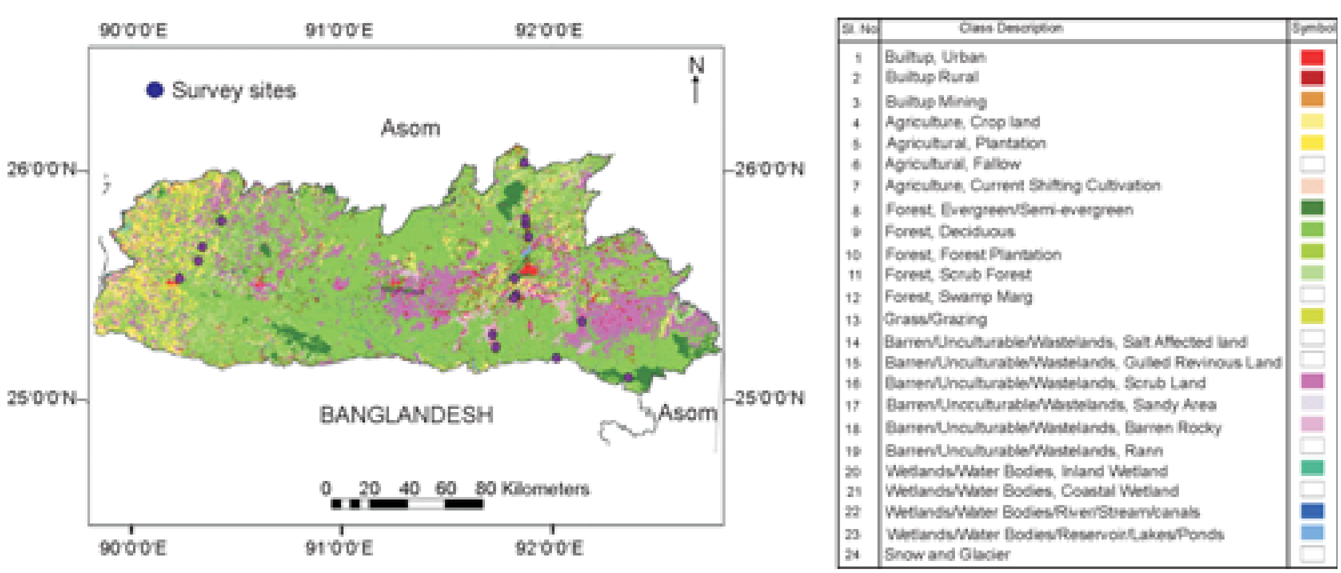


*Fig. 1:* Topographic map showing study sites and type of land use in Meghalaya state of northeastern India.

acetic acid and ethanol solution and identified morphologically[13–19].

*Determination of physicochemical parameters*

Water samples were collected in one litre bottles and analysed for nine different physicochemical parameters using Orion® 5-Star portable multi-parameter analyser (Thermo Scientific) following manufacturer's instructions. Water samples from atleast three different places of each collecting location were taken for expressing various parameters, viz. water temperature, pH, dissolved oxygen concentration (DOC), dissolved oxygen saturation (DOS), conductivity, total dissolved solute (TDS), turbidity, resistivity and salinity. Water flow rate was determined using rod float method[20] and expressed in $cm^{-1}$. These parameters are important predictors of blackflies distribution and density in the river ecosystems[21].

*Data analysis*

All the water variables and blackfly density at each place have been expressed as mean ± standard error of mean (SEM). Water variables of the locations where simuliids were recorded and compared with the locations where simuliids were not recorded using unpaired Student's *t*-test. Water flow rates and simuliids density among the various locations were compared using one-way ANOVA, whereas water flow rate between the locations where simuliids were recorded and not recorded were compared using unpaired Student's *t*-test. Two diversity indices, viz. Shannon-Wiener index (H) and Simpson Index (D) were calculated to understand the $\alpha$-diversity of simuliids, whereas cumulative distribution of population has been carried out using Lorenz method. The relationship between simuliids density and each water variable was determined using linear description of regression, whereas multiple regression was used to determine the fitness for the presence of simuliid species. Principal component analysis (PCA) followed by Varimax rotation was used to collapse water parameters into a smaller number of statistically independent principal components to determine their association with the simuliids distribution. All the data were tested for normality using Kolmorov-Smirnov test before statistical analysis[22]. The datasets were analysed using GraphPad InStat, XLSTAT and SPSS-19 computer softwares.

## RESULTS

*Simuliid species density and richness*

The pre-adult stages of simuliid species occurrence and density among the nine locations are shown in Table 2. A of total of 565 specimens (aquatic stages) comprising of the three species, namely *Simulium (S) barraudi* Puri, *S. (S) striatum* Brunetti, and *S. (S) himalayense* Puri, were collected during the study. Each of the 16 surveyed locations recorded only one type of simuliid species in the present investigation. *Simulium barraudi* was collected from five locations whereas *S. striatum* and *S. himalayense* could be collected from three and one locations respectively. *S. barraudi* was the most common (56.8%) and its density was significantly high ($\chi^2$ = 289.3; df = 2; $p$ <0.0001) as compared to *S. striatum* (34.2%) and *S. himalayense* (9%). Umsamlem river recorded the highest density (47 ± 2.6) of simuliids, whereas Mawsmai stream recorded the least density (9 ± 2.3). The difference in the density of simuliids among the collecting locations was statistically significant (F = 20.6; df = 25; $p$ <0.001). Diversity analysis revealed that average population size of each species was 188.3 and diversity indices D & H (Simpson & Shannon-Wiener) were 0.4466 and 1.306, respectively. The cumulative percentage of simuliid species population is shown in Fig. 2.

*Physicochemical parameters*

Various physicochemical parameters of water associated with the breeding ecology of simuliids among all

Table 2. Water flow rate and Simuliid species composition and abundance among the study sites

| Location | Water flow rate (Mean ± SEM)) | Substrate | Species recorded | Number recorded (Mean ± SEM) | Relative abundance |
|---|---|---|---|---|---|
| Elephant falls | 31 ± 2.1 | Stone | *Simulium barraudi* Puri | 18.7 ± 3 | 0.10 |
| Umtyngngar | 38 ± 6.3 | Stone | *Simulium himalayense* Puri | 17 ± 3.5 | 0.09 |
| Mawsmai | 22.3 ± 2.3 | Leaf | *Simulium barraudi* Puri | 9 ± 2.3 | 0.05 |
| Umtrew | 21 ± 1.2 | Leaf & Wood | *Simulium striatum* Brunetti | 7.7 ± 2.4 | 0.04 |
| Umran | 19.3 ± 1.2 | Leaf | *Simulium striatum* Brunetti | 9.7 ± 2 | 0.05 |
| Umsamlem | 55.3 ± 4.1 | Leaf | *Simulium striatum* Brunetti | 47 ± 2.6 | 0.25 |
| Rari | 33.3 ± 2.2 | Leaf | *Simulium barraudi* Puri | 19 ± 2.3 | 0.10 |
| Chiokgre | 43 ± 1.7 | Leaf | *Simulium barraudi* Puri | 26 ± 3.2 | 0.14 |
| Warebok | 40 ± 1.2 | Wood | *Simulium barraudi* Puri | 34.3 ± 4.1 | 0.18 |

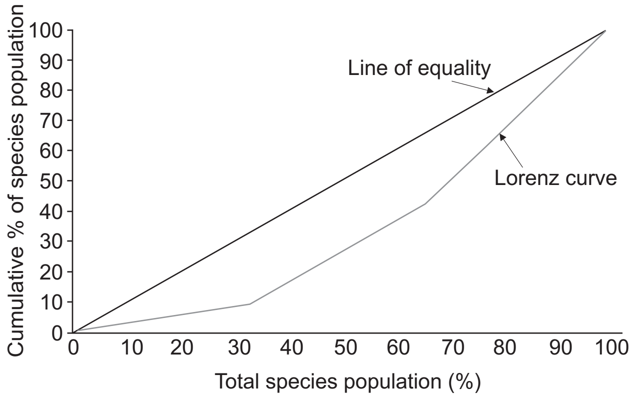


*Fig. 2:* Lorenz curve for cumulative distribution (%) of Simuliid species in Meghalaya.

the sixteen study locations are shown in Table 3. Among the nine physicochemical parameters, turbidity and pH among the locations where simuliids were recorded and not recorded differ significantly ($p$ = 0.03; df = 14; $t$ = 2.37 for turbidity and $p$ <0.01; df = 14; $t$ = 3.01 for pH). The pH and turbidity (mean±SEM) among the locations where simuliids were recorded were 7.21 ± 0.1 (95% CI = 6.98–7.45) and 5.5 ± 1.1 (2.96–7.99) respectively, while among the locations where simuliids were not recorded 6.02 ± 0.4 (95% CI = 4.96–7.09) and 2.25 ± 0.7 (0.61–3.89). Similarly, the water flow rate was higher among the locations where simuliids were recorded ($p$ = 0.004; $t$ = 3.3; df = 14). All the water parameters differ among all the study locations ($p$ <0.0001). The results of linear regression showed that simuliids density was found to be associated with water flow rate (Table 4). Multiple regression model for the presence of simuliids species is—Simuliids species present = –5.007 (Coefficient) + 0.007245 (Temperature) + 0.601 (pH) + 1.076 (DO concentration) + 0.1049 (DO saturation) + 0.02350 (Conductivity) + 0.03127 (Total dissolved solutes) + 0.03516 (Turbidity) + 4.824 (Resistivity) + 11.065 (Salinity).

The PCA post Varimax rotation indicated that water parameters along D2 axis accounted for 24.10%, whereas on D1 axis accounted for 42.25% variability among the sampling locations (Fig. 3). Atleast two principal components have eigenvalue >1 and accounted for 32.68% of variation. Simuliid density was associated

Table 4. Regression for simuliid density and water parameters

| Water parameter | Regression (r) | Squared regression ($r^2$) | 95% CI | $p$-value |
|---|---|---|---|---|
| Water flow | 0.94 | 0.88 | 0.72–0.99 | 0 |
| Water temperature | 0.20 | 0.04 | 0.53–0.76 | 0.60 |
| pH | 0.06 | 0 | 0.63–0.70 | 0.88 |
| DO concentration | 0.60 | 0.34 | 0.13–0.90 | 0.10 |
| DO saturation | 0.66 | 0.43 | 0.19–0.92 | 0.06 |
| Conductivity | –0.31 | 0.09 | 0.81–0.45 | 0.42 |
| Total dissolved solutes (TDS) | –0.31 | 0.10 | 0.81–0.45 | 0.41 |
| Turbidity | –0.13 | 0.02 | 0.73–0.58 | 0.73 |
| Resistivity | –0.05 | 0 | 0.69–0.64 | 0.90 |
| Salinity | –0.31 | 0.10 | 0.81–0.45 | 0.42 |

Table 3. Physicochemical parameters at the water at collection locations

| Location | Sample temperature (°C) | pH[1] | DO[2] concentration (mg/l) | DO[2] saturation (%) | Conductivity (µS/cm) | TDS[3] (mg/l) | Turbidity (NTU) | Resistivity (Ω-cm) | Salinity (ppt) | Species recorded |
|---|---|---|---|---|---|---|---|---|---|---|
| Elephant Falls | 11.5 ± 0.3 | 7.4 ± 0 | 4.5 ± 0.2 | 59.2 ± 0.3 | 104.3 ± 0.4 | 52 ± 0.6 | 1.8 ± 0 | 0.01 ± 0 | 0.1 ± 0 | Yes |
| Umtyngngar | 9 ± 0.2 | 7.7 ± 0 | 5.4 ± 0 | 66.9 ± 0.5 | 35.8 ± 0.1 | 17.7 ± 0.7 | 11.9 ± 0.4 | 0.03 ± 0 | 0 ± 0 | Yes |
| Sohra | 16.9 ± 0.1 | 3.8 ± 0.1 | 5.6 ± 0.1 | 69.1 ± 0.2 | 306 ± 3.1 | 157.7 ± 2.9 | 2.8 ± 0.1 | 0 ± 0 | 0.2 ± 0 | No |
| Mawsmai | 18.9 ± 0.1 | 6.6 ± 0 | 5.2 ± 0.1 | 63.5 ± 0.4 | 213.2 ± 1.7 | 102.3 ± 2 | 3.1 ± 0 | 0 ± 0 | 0.1 ± 0.1 | Yes |
| Mawjrong | 11.8 ± 0.3 | 7.7 ± 0 | 6.9 ± 0.5 | 75.8 ± 0.6 | 18.4 ± 0.4 | 11.3 ± 1.2 | 2.3 ± 0 | 0.05 ± 0 | 0 ± 0 | No |
| Dawki | 19.1 ± 0.1 | 7.4 ± 0 | 4.3 ± 0.1 | 55.5 ± 0.4 | 51.7 ± 0.7 | 24.3 ± 1.8 | 3.3 ± 0.2 | 0.02 ± 0 | 0 ± 0 | No |
| Jorain | 16.8 ± 0 | 6.1 ± 0.1 | 5.8 ± 0.1 | 71 ± 0.6 | 40.5 ± 0.4 | 20.7 ± 1.2 | 0.2 ± 0 | 0.02 ± 0 | 0 ± 0 | No |
| Sonapur | 20.8 ± 0.1 | 6.2 ± 0 | 5.5 ± 0.1 | 62.9 ± 0 | 325.3 ± 2.6 | 164.7 ± 1.5 | 0.6 ± 0 | 0 ± 0 | 0.3 ± 0 | No |
| Umtrew | 18.8 ± 0.1 | 7.4 ± 0.1 | 5.2 ± 0.1 | 60.9 ± 0.1 | 16.7 ± 0.3 | 7.3 ± 0.3 | 4.8 ± 0.1 | 0.06 ± 0 | 0 ± 0 | Yes |
| Umran | 18.5 ± 0.1 | 7.2 ± 0 | 5.7 ± 0.1 | 68.3 ± 0.2 | 27.8 ± 0.1 | 15 ± 1 | 9.5 ± 0.2 | 0.04 ± 0 | 0 ± 0 | Yes |
| Umsamlem | 18.8 ± 0.1 | 7.1 ± 0 | 6.8 ± 0 | 78.1 ± 0.2 | 28.1 ± 0.7 | 13.7 ± 1.2 | 6.5 ± 0.2 | 0.03 ± 0 | 0 ± 0 | Yes |
| Byrnihat | 25.4 ± 0.1 | 7.6 ± 0.1 | 4.7 ± 0.1 | 56.9 ± 0.3 | 46.1 ± 0.6 | 22.3 ± 1.8 | 5.7 ± 0.1 | 0.03 ± 0 | 0 ± 0 | No |
| Rari | 18.7 ± 0.07 | 7.4 ± 0 | 5.6 ± 0.2 | 63.4 ± 0.4 | 73.6 ± 0.3 | 37.7 ± 2.2 | 5.7 ± 0.1 | 0.02 ± 0 | 0 ± 0 | Yes |
| Chiokgre | 21.7 ± 0.06 | 7.1 ± 0 | 6.4 ± 0.1 | 79 ± 0.3 | 65.7 ± 0.3 | 30 ± 2 | 4.7 ± 0.1 | 0.02 ± 0 | 0 ± 0 | Yes |
| Warebok | 20.57 ± 0.03 | 7.2 ± 0 | 5.5 ± 0 | 66.9 ± 0.4 | 45.8 ± 0.2 | 23.3 ± 1.3 | 1.9 ± 0 | 0.02 ± 0 | 0 ± 0 | Yes |
| Rongkhon | 20.7 ± 0.06 | 7.7 ± 0.1 | 5.3 ± 0.2 | 58.6 ± 0.2 | 74.2 ± 0.5 | 37.7 ± 0.7 | 1.7 ± 0 | 0.02 ± 0 | 0 ± 0 | No |

[1]Hydrogen ion concentration; [2]Dissolved oxygen; [3]Total dissolved solute.

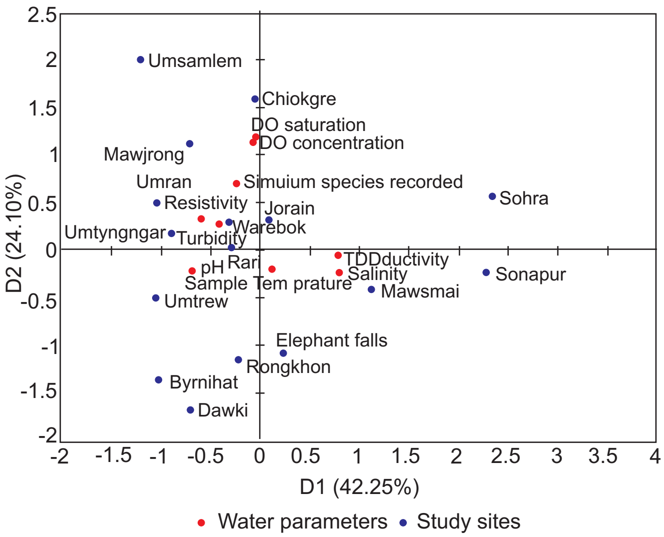


*Fig. 3:* Biplot (after Varimax rotation) of principal component analysis (PCA) for water parameters and Simuliid density (axes D1 and D2 = 66.35%).

with pH, water temperature and water flow in the study locations.

## DISCUSSION

The present study is the most detailed investigation for the prevalence of simuliid species and its association with various water parameters in 16 different river sites of Meghalaya. *Simulium barraudi* and *S. striatum* were the most common species observed in the present study, whereas *S. himalayense* was recorded from one sampling site only. A total of 10 species of simuliids have been reported from northeastern states of India, of which *S. himalayense* had wide distribution[13, 23–25]. The earlier studies have reported *S. aureohirtum*, *S. indicum*, *S. dentatum, S. grisescens* and *S. rufibasis* in various parts of Meghalaya and adjoining states[15, 16, 19, 26].

Rivers of Meghalaya have high water flow due to high rainfall, which support the breeding and distribution of simuliids. Pre-adult stages of the species are found to occur on trailing leaves, twigs, stone surfaces, stems, etc., which are about 5 cm below the water surface. Abundance and distribution of larvae and pupae are influenced by rainfall, which causes an increase in the water flow speed, nutritive status of the river and decolonising algae leading to an increase in larval density[27]. Water flow rate, pH and turbidity were the limiting parameters in the distribution of the pre-adult blackflies. A variety of factors (water temperature, velocity, streambed, dissolved oxygen concentration and vegetation) acting in concert have been found to influence the blackfly richness in the streams of central Amazonia[8]. We could record only three species and each river recorded only one species in a limited sampling time. Previous study has suggested that the number of species increases as the collecting time and efforts increase[28]. However, there are regions that support fewer simuliid species only and a large number of samples would not incur any increase in the species richness. Further, water variables form distinct ecoregions and have maximum contribution in simuliid species separation in different ecoregions. The overall river health has been important for the distribution and richness of the simuliid species, which have biotic interactions acting as primary determinants of species diversity[29, 30].

In the present study, the simuliid distribution was not associated with altitude, whereas in the earlier studies altitude has been reported as major component in species richness and distribution[6]. Freshwater ecologist believe that in addition to the certain measurable water variables, there are other parameters which either singly or in combination with each other limit the proliferation of pre-mature stages of simuliids in the breeding habitat[21]. Therefore, prevalence of aquatic stages in the rivers act as biological indicator of the river health, which is often considered analogous with human health[31, 32]. No simuliid specimen was recorded at seven river sites, two of which were near the cement factory, three had excavation sites and remaining two were near the open field coal mines. These rivers were receiving high effluents discharge from the nearby developmental activities. The industries effluent waste discharged into the river breeding habitat disturb the microhabitat, turning it unfit for simuliids breeding and growth. McCreadie *et al*[21] has reported that human related changes and discharge of waste water reduced the abundance of a non-anthropophilic simuliid species.

Our study provides new information on simuliid species distribution and its association with breeding water parameters in a little studied region of high biological interest. Turbidity, water flow and pH are important water parameters affecting the simuliid species prevalence and density in freshwater rivers. The study indicates that each simuliid species prefer different sets of physicochemical variables in breeding habitats, which are specific to that particular species. Therefore, simuliid species community as a whole cannot be considered as a suitable indicator of the streams water quality. In addition to describing simuliids, the information provided herein will be useful for the conservation of aquatic ecology and biodiversity in this environmentally important state of India.

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