THE DISTRIBUTION AND SPECIES DIVERSITY OF BLACK FLIES (DIPTERA: SIMULIIDAE) IN SELECTED AREAS IN RANAU, SABAH

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ABSTRACT

Blackfly play an important role in the freshwater ecosystem as a biological indicator, as the blackfly prefers clean water area to breed. Certain blackfly species are categorized as vector as it can transmit disease known as Onchocerciasis (river blindness). This study is an entomology research which focuses on the species diversity of blackfly (Diptera: Simuliidae) in Ranau District of Sabah. Eight sampling stations located the highlands and lowlands were chosen. Larvae and pupae were manually collected directly from the substrate in the streams and river. A total of 12 data sets were collected over a period of six months, which started from August 2015 to January 2016. The data collection was done once every forth night. A preliminary
result from this study has so far recorded fourteen species of blackfly. Among the fourteen species recorded, *Simulium (Gomphostilbia) alienigenum* was the first specimen recorded in Malaysia especially in Sabah. All the fourteen species of black flies recorded in this study belongs to three subgenus which are; *Simulium, Nevermania* and *Gomphostilbia*. The highest individual being recorded comes from the subgenus of *Simulium*. The diversity indices result shows a ranged from 1.13 to 1.58, and this indicates low diversity in the eight sampling stations.

**Keywords:** species diversity, aquatic Insect, Simuliidae, Ranau district

**ABSTRAK**

Black flies are aquatic insect, from the order Diptera, family Simuliidae (Crosskey, 1990). The immature stages of the black fly are very important components in freshwater ecosystem, as they play an important role in aquatic food chain. The larvae and pupae feed on the organic particles in fast flowing water ecosystem (Zhang et. al., 1998). The environmental characterization in freshwater ecosystem may affect the distribution species and community of the black flies (Kazanci, 2006).

To date, there is a total 2189 black flies species recorded worldwide (Adler & Crosskey, 2015). While in Malaysia, there is more than 69 species (Adler & Crosskey, 2011; Takaoka et al., 2011a, 2011b), six species in Sarawak (Takaoka, 2001) and more than 24 species recorded in Sabah.

So far, diversity studies of black flies in Sabah have not been documented yet. This family can be locally restricted to stream areas where proper conditions such as fast flowing water, high dissolved oxygen and others that allow the development of their larvae (Lake & Burger, 1983).
Previous study states that altitude and current velocity had influence on the distribution of the black flies species (Gallardo-Mayenco, A. & Prenda, 2002). This study focused on the blackfly distribution in selected streams located at Ranau District of Sabah. The aim of this preliminary study was to determine the distribution and species diversity of black flies in Ranau district.

**MATERIALS AND METHODS**

**Study Sites**
The study was conducted in Ranau district (Figure 1), situated 108 km from Kota Kinabalu city which covers a total area of 3,555.51 km². Ranau District is situated between 5°30’N to 6°25’N and 116°30’E to 117°5’E. The sampling stations were located in a range of altitude between 370 to 1427 meters above sea level.

A total of eight sampling stations were chosen. Four sampling stations located in highland were Nikgold, Ranau 3, Ranau 4 and Ranau 5, while the Libang, Poring, Lohan and Ranau 1 were in lowland area. The eight sampling station chosen also had different in habitat characteristics (Table 1).

![Figure 1](image.jpg)  
**Figure 1**  
Location of Ranau on map
<table>
<thead>
<tr>
<th>Sites</th>
<th>Water pH</th>
<th>Temperature (°C)</th>
<th>Altitude (m)</th>
<th>Width of streambed (m)</th>
<th>Depth of Riverbed (cm)</th>
<th>Turbidity</th>
<th>Dominant Substrate</th>
<th>Canopy Cover</th>
<th>Riparian Vegetation</th>
<th>Stream Flow</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lohan</td>
<td>7.38</td>
<td>27.2</td>
<td>370</td>
<td>7</td>
<td>50</td>
<td>C</td>
<td>Leaves</td>
<td>Open</td>
<td>Bamboo Tress, Fruit Tress</td>
<td>F</td>
</tr>
<tr>
<td>Ranau 1</td>
<td>7.37</td>
<td>22.3</td>
<td>528</td>
<td>1.3</td>
<td>45</td>
<td>C</td>
<td>Roots</td>
<td>Close</td>
<td>Shrub</td>
<td>F</td>
</tr>
<tr>
<td>Poring</td>
<td>7.2</td>
<td>24.4</td>
<td>534</td>
<td>1.5</td>
<td>150</td>
<td>C</td>
<td>Roots</td>
<td>Open</td>
<td>Shrub</td>
<td>S</td>
</tr>
<tr>
<td>Libang</td>
<td>7.07</td>
<td>28.2</td>
<td>541</td>
<td>5</td>
<td>60</td>
<td>C</td>
<td>Leaves</td>
<td>Open</td>
<td>Bamboo Tress</td>
<td>M</td>
</tr>
<tr>
<td>Ranau 4</td>
<td>7.33</td>
<td>21.3</td>
<td>752</td>
<td>2</td>
<td>50</td>
<td>C</td>
<td>Leaves</td>
<td>Open</td>
<td>Shrub, Fruit Trees</td>
<td>F</td>
</tr>
<tr>
<td>Ranau 5</td>
<td>7.35</td>
<td>21.4</td>
<td>754</td>
<td>3</td>
<td>40</td>
<td>C</td>
<td>Rocks, Leaves</td>
<td>Close</td>
<td>Fruit Tress</td>
<td>F</td>
</tr>
<tr>
<td>Ranau 3</td>
<td>7.13</td>
<td>20.5</td>
<td>759</td>
<td>1.2</td>
<td>20</td>
<td>C</td>
<td>Roots</td>
<td>Open</td>
<td>Shrub</td>
<td>S</td>
</tr>
<tr>
<td>Nikgold</td>
<td>7.31</td>
<td>18.9</td>
<td>1427</td>
<td>2</td>
<td>35</td>
<td>T</td>
<td>Roots</td>
<td>Open</td>
<td>Shrub</td>
<td>S</td>
</tr>
</tbody>
</table>

T= Turbid, C = Clear, S = Slow, M = Moderate F = Fast
Sampling Methods
The sampling method in this study consists of 4 phases. There are, sample collection, sorting of specimens, rearing of pupa and lastly was species identification. The larvae and pupae were manually collected directly from the substrate under the water. A total 12 data sets were collected over a period of six months, which is started from August 2015 to January 2016. The data collection was done once every forth night. In this study the environmental parameter that were measured are, the altitude, water pH, water temperature, width and depth of streambed, types of substratum, turbidity, canopy cover, riparian vegetation and stream flow.

After collecting the sample from a river or stream, pupa were placed into a test tube and brought to the laboratory for rearing. After the adult blackfly emerged, it is preserved along with the pupa skin in an effendorf tube containing 80% ethanol, for further species identification. While the black fly larvae were preserved in Carnoy’s solution.

Data Analysis
The data were analyzed based on diversity indices using the PAST software (Paleontological Statistic Software Package for Education and Data Analysis). The Diversity indices include the Margalef Index, Shanon Index, Evenness Index and Beta Diversity Index. While the similarities among the sites were analyze by using cluster analysis based on Jaccard’s Coefficient index. These analyses were runs by using the PAST software.
RESULT AND DISCUSSION

Results of this study recorded a total of 14 Simuliidae species from the eight selected areas in Ranau from August 2015-January 2016 (Table 2). The 14 species of black flies came from three subgenera *Gomphostilbia*, *Nevermania* and *Simulium*. Among the 14 species, *Simulium alienigenum* and *S. nobile* were only found in Lohan sites, which is a highland area. While the *S. keningauense*, *S. sabahense* and *S. sp.* were widely distributed in almost all the eight sampling stations.

The previous study suggests that, *S. keningauense* and *S. sabahense* are considered common species, as both of this species can be found in various environmental conditions (Smart & Clifford, 1968; Takaoka, 2001). This shows that both *S. keningauense* and *S. sabahense* can be tolerate with the fast, moderate and slow water velocity and different substrates.

While, the *S. (Nevermania) aureohirtum* was only found in highland area, Nikgold and Ranau 3. According to previous study, it was reported that most of the subgenus *Nevermania* species inhabit highland area (Thaijarern *et. al*, 2013). In this study, *S. aureohirtum* can only be found in the streams with the average water temperature ranging from 18.9 °C to 20.5 °C.

There were overlapping species which occurred between several study sites and this is also supported by the Beta Diversity index score of 1.7, as shown in Table 3. Among the overlapping species that occurred were *S. sabahense*, *S. keningauense* and *S.sp.* These species were found in Libang, Poring, Lohan, Ranau 1, Ranau 3, Ranau 4, and Ranau 5. The overlapping species may occur due to the similar features habitat in the eight study areas.
<table>
<thead>
<tr>
<th>Subgenus</th>
<th>Species</th>
<th>Nikgold</th>
<th>Libang</th>
<th>Poring</th>
<th>Lohan</th>
<th>Ranau 1</th>
<th>Ranau 3</th>
<th>Ranau 4</th>
<th>Ranau 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gomphostilbia</td>
<td><em>S. alienigenum</em></td>
<td></td>
<td></td>
<td>●</td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>S. sarawakense</em></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>S. sheilae</em></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td>●</td>
<td>●</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>S. (nr) trangense</em></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td>●</td>
<td>●</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>S. rayohense</em></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td>●</td>
<td>●</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nevermania</td>
<td><em>S. aureohirtum</em></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>S. borneonse</em></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Simulium</td>
<td><em>S. alberti</em></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td>●</td>
<td>●</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>S. beludense</em></td>
<td>●</td>
<td></td>
<td>●</td>
<td></td>
<td>●</td>
<td>●</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>S. keningauense</em></td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
</tr>
<tr>
<td></td>
<td><em>S. laterale</em></td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
</tr>
<tr>
<td></td>
<td><em>S. nobile</em></td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td>●</td>
<td>●</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>S. sabahense</em></td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
</tr>
<tr>
<td></td>
<td><em>S. sp</em></td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
</tr>
</tbody>
</table>
The Margalef species richness index values in all sites were less than 1, which indicates very low species richness. However, Poring recorded the highest species richness index value (D.mg) with the score 0.80 with total of eight species, which is the highest among all sites.

While the species diversity index $H'$, ranged between 1.13 to 1.58 which indicates low diversity of black flies in all sites. However, Ranau 3 shows the highest value of $H'$ (1.58), with a total of seven species recorded. Nikgold has the lowest value of $H'$ index, (0.51) as there were only two species of black flies recorded.

Despite the poor diversity, the evenness value index $E$, scored almost 1, indicating an even distribution of black flies communities (Table 3). The common shared species which occurred across all the sites were $S. keningauense$, $S. sabahense$, $S. sp.$ and $S. laterale$. The occurrence of overlapped species may be due to the similar habitat characterizations among the sites that support development of some blackfly species (Basoren & Kazanci, 2011). The $\beta$ diversity score which was greater than the value ‘1’, indicates that there are shared species among all the study sites.
Table 3  The Diversity Indices Score for 8 selected areas in Ranau.

<table>
<thead>
<tr>
<th>Sites</th>
<th>Libang</th>
<th>Poring</th>
<th>Lohan</th>
<th>Ranau 1</th>
<th>Ranau 3</th>
<th>Ranau 4</th>
<th>Ranau 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>985</td>
<td>1760</td>
<td>282</td>
<td>1168</td>
<td>928</td>
<td>916</td>
<td>1023</td>
</tr>
<tr>
<td>$D_{mg}$</td>
<td>0.44</td>
<td>0.80</td>
<td>0.72</td>
<td>0.57</td>
<td>0.73</td>
<td>0.73</td>
<td>0.43</td>
</tr>
<tr>
<td>$H'$</td>
<td>1.13</td>
<td>1.47</td>
<td>1.36</td>
<td>1.25</td>
<td>1.58</td>
<td>1.39</td>
<td>1.28</td>
</tr>
<tr>
<td>$E$</td>
<td>0.77</td>
<td>0.62</td>
<td>0.78</td>
<td>0.69</td>
<td>0.81</td>
<td>0.67</td>
<td>0.9</td>
</tr>
<tr>
<td>$\beta$</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.7</td>
</tr>
</tbody>
</table>

$N =$ sample size, $D_{mg} =$ Margalef’s Index, $H' =$ Shanon’s Weiner Index, $E =$ Evenness Index, $\beta =$ Beta Diversity Index

Figure 2  Cluster analysis showing the similarities among the 8 study sites (Jaccard’s Coefficient)
Figure 2, it shows the cluster analysis results of the eight sampling stations. A result shows that there is similarity of black flies species in Ranau 1 and Ranau 5 is the highest. There were four species recorded in Ranau 1 and Ranau 5, which were S. keningauense, S. laterale, S. sabahense and S. sp. Ranau 1 and Ranau 5 have similar environmental parameters whereas both of these sites having fast flowing water, close canopy area and almost same water temperature.

While Nikgold and the other seven study area showed a poor similarity value. As shown in Table 2, the presence of S. aureohirtum and S. borneoense were only found in Nikgold. Furthermore, the microhabitat characteristic in Nikgold was different compared to the other seven study sites. The water temperature in Nikgold was 18.9 °C which was the lowest water temperature average compared to the other study area. Nikgold is also located at an elevation of 1427 m above sea level which categorize it as highland area. Previous study recorded that the Nevermania species group in the highland area, with lower water temperature and fast flowing water (Takaoka, 2001).

CONCLUSION

In this study, Simulium (Gomphostilbia) alienigenum and Simulium (Simulium) nobile were recorded from Sabah areas for the first time. Besides, the biodiversity index score shows that, the species diversity in the eight selected sites in Ranau is still low, with only 14 species recorded. However, it is expected that more species will be found in Ranau District, because there are many more unexplored areas that could potentially be the habitat of black flies.

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REFERENCES


