

Habitat variation and larval mosquito abundance in San Isidro River, Laguna, Philippines

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ABSTRACT

This study aims to assess the mosquito abundance in San Isidro River (Laguna, Philippines) in varying breeding habitat types and water quality conditions. The habitat variation of the mosquitoes was assessed in terms of the water body's upstream, midstream, and downstream areas. The variations in the habitat were determined by considering the water quality parameters (dissolved oxygen, temperature, pH, total dissolved solids, and conductivity) and the breeding habitat conditions (intensity of light, water current, water depth, and vegetation). Mosquito larvae surveys were conducted in all areas of the San Isidro River. Results showed that the *Culex annulirostris* mosquito larvae inhabited the entire San Isidro River. All water samples obtained were within water quality standards, except total dissolved solids and dissolved oxygen. Results of the multiple regression analysis suggest that temperature and dissolved oxygen are the best predictor variable associated with the abundance of the mosquito larvae ($r = 0.709$, $P = 0.000$). Monitoring of freshwater environment is needed to ascertain the possible threats that may affect human health.

KEYWORDS: biomonitor, water quality, mosquito larvae

INTRODUCTION

Mosquitoes are recognized to be important vectors in disease transmission. They proliferate in our environment and contribute to the occurrence of morbidities and mortalities. Their occurrence has been primarily associated with the availability of breeding habitats. However, studies have shown that their abundance and distribution are affected by a number of environmental conditions emanating in their breeding habitats, namely water chemistry, geographical location, size of water body, presence of vegetation, predators (Sanford et al., 2005; Schafer et al., 2004; Greenway et al., 2003), degree of eutrophication, sunlight, and presence of human and animal population distributions (Muturi et al., 2008; Okogun et al., 2003). Their occurrence has been indicated to vary from place to place, as they primarily depend on the biological and physicochemical conditions present (Kengluocha et al., 2005). The challenge of understanding the population dynamics of mosquitoes in varying habitats is important, as this will enable us to predict their occurrence and studies as such can be used to control these mosquitoes. In the Philippines, there are limited studies that investigate the environmental conditions of breeding habitats and the relation of these habitats to the mosquito larval distribution. Most studies focus more on larval surveillance, but not relating the habitat conditions to the occurrence of larval mosquitoes in the environment. This study aims to investigate the larval habitats and assess whether variation in the breeding habitat type and water quality parameters can affect the mosquito abundance in a freshwater body resource like a river. The result of this study is vital, as it enables us to monitor the larval mosquito occurrence for efficient mosquito control.

METHODS

The study was conducted in San Isidro River, Laguna, Philippines. Three study sites (upstream, midstream, and downstream) were identified to represent the entire stretch of the San Isidro River. The San Isidro River is a shallow river with slow water current that exits into Laguna Lake. There are five sampling points purposively selected on the three study sites identified. The upstream area of San Isidro River is less accessible to the communities, whereas the midstream and downstream areas are situated near the

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community settlements. The study site is located between the longitude of 14°21'50.6"N and the latitude of 121°3'14.5"E.

Sampling sites 1 and 2 are situated at the downstream of the San Isidro River, sampling sites 3 and 4 are situated at the midstream of the San Isidro River, and sampling site 5 is situated at the upstream area. The sites were assessed for their habitat environmental variables, particularly vegetation covering, and presence of algae and other biotic organisms, water depth, light/shade, and permanence. Grab water samples were obtained in all study sites. Water samples were assessed for the physicochemical parameters, particularly pH, total dissolved solids (TDS), temperature, salinity, conductance, and dissolved oxygen using the Cyberscan Series 600 (Eutech Instruments Pte. Ltd.) in situ. Water quality parameters were compared with standards set in the DENR Administrative Order of 2008 for Water Quality Standards and General Effluent Standards. The water depth was determined using a meter stick. The movement of larval habitat's water was assessed whether it's flowing or not flowing. Light/shade was assessed in terms of sites that are sun-exposed or shaded. Presence of vegetation was assessed in terms of sites that have vegetation or have no vegetation, and other biotic organisms were recorded in the area.

The standard dipping method (O'Malley, 1989) was used to collect the mosquito larvae. The field-collected fourth instar larvae from the study sites were examined morphologically using standard keys. Mosquito larvae that were not in their fourth instar stages were reared until they reached their fourth instar stages and examined morphologically to species level. Mosquito larvae were placed in hot water and preserved in 10% formalin solution.

The breeding index for each study site was calculated using the formula (Belkin, 1954): $BI = TLP/ND \times BP$, where BI = breeding index, TLP = total number of larvae and pupae taken, ND = number of dips, and BP = number of breeding places/sampling stations. Larval mosquito abundance per study site was recorded and compared.

An ANOVA was used to determine if significant differences exist for all the water quality parameters across all the study sites observed. Significant level was set at 0.05. A multiple regression analysis was used to determine the best predictor variables contributing to the abundance of the mosquito larvae in the study area. The analysis was done through a stepwise elimination method. The model equation was developed following a regression relationship: $Y = \alpha + \beta X$, where α is the intercept variable, β is the slope parameter, and X is the measurement of the abiotic factors (salinity, conductance, pH, dissolved oxygen, TDS, and temperature). A criterion was used for α : for entry set, $\alpha = 0.05$; for removal set, $\alpha = 0.10$. The linear model was developed, and all statistical analyses were performed using the GNU PSPP software.

RESULTS

The physicochemical quality of the water of San Isidro River is indicated in Table 1. The TDS and the dissolved oxygen were below the threshold limit of the Class D waters set by the DENR. All the other water quality parameters were within the threshold limit. Significant differences exist for all the water quality parameters examined across all the study sites of San Isidro River ($P < 0.05$). Among all the study sites, site 3 is a non-flowing river inlet, unlike the other flowing sites. Variations in the biotic organisms were also observed in the different study sites, as fish and macrophytes were evident in sites 1, 2, and 4 and vegetation is present in site 5. Sites 3 and 5 were partly shaded areas, while sites 1 and 2 were lighted areas.

Table 1. Mean \pm SEM of the water quality parameters of San Isidro River, Laguna, Philippines

Study sites	Temperature (°C)	TDS (ppt)	Dissolved oxygen (mg/L)	pH	Salinity (ppt)	Conductivity (mS)	Water depth (cm)
Site 1	29.38 \pm 0.55	1.17 \pm 0.26	0.82 \pm 0.01	7.02 \pm 0.01	1.173 \pm 0.000	142.83 \pm 0.01	9.75 \pm 0.50
Site 2	29.38 \pm 0.06	1.02 \pm 0.00	2.27 \pm 0.00	6.69 \pm 0.00	1.043 \pm 0.000	154.13 \pm 0.04	15.85 \pm 0.50
Site 3	29.95 \pm 0.07	1.12 \pm 0.00	1.43 \pm 0.00	6.73 \pm 0.00	1.124 \pm 0.000	163.53 \pm 0.01	22.48 \pm 0.80
Site 4	29.97 \pm 0.05	1.13 \pm 0.00	1.18 \pm 0.01	6.94 \pm 0.00	1.165 \pm 0.000	171.12 \pm 0.00	13.67 \pm 0.52
Site 5	29.97 \pm 0.05	1.14 \pm 0.00	2.88 \pm 0.00	6.76 \pm 0.00	1.171 \pm 0.000	164.38 \pm 0.02	15.77 \pm 0.64

A total of 776 mosquito larvae were obtained in all the study sites examined. Table 2 shows the abundance, distribution, and breeding index of the larvae mosquito obtained in San Isidro River. Among

the sites examined, the upstream area had the most larval mosquitoes and the highest breeding index whereas sites 1 and 2 representing the downstream area had the lowest abundance and breeding index.

Table 2. Larval mosquito abundance, distribution, and breeding index in San Isidro River

Study site	Abundance	Breeding index
Site 1	80	16.0
Site 2	97	19.0
Site 3	216	43.2
Site 4	122	24.4
Site 5	261	52.2

The mosquito larva obtained in all the study sites was the *Culex annulirostris*. The larva of the *C.annulirostris* is characterized morphologically based on the taxonomic key (Delfinado, 1966).

The results of the multiple linear regression analysis showed that, among the different water quality parameters examined, dissolved oxygen and temperature were the important factors in the abundance of the *C.annulirostris* mosquito larvae ($Y = -884.4 + 30.28$ [temperature] + 8.88 [dissolved oxygen], $r = 0.709$, $r^2 = 0.502$, $P < 0.05$). No significant correlation was established between salinity, conductance, pH, and TDS.

DISCUSSION

This was a cross-sectional study, and its scope was limited in determining the habitat of the mosquito larvae. This study was also limited in relation to the abiotic factors (pH, conductance, salinity, TDS, temperature, and dissolved oxygen) to the abundance of the mosquito larvae obtained in all the study sites in San Isidro River, Philippines. We observed heterogeneity in terms of the habitat particularly in the water quality parameters observed in the downstream, midstream, and upstream areas of the freshwater river. All the study sites were identified as shallow areas that provided habitats conducive for the survival of the mosquito larvae. The presence of other organisms in the water, like in sites 1, 2, and 4, may be contributory to the lower mosquito larvae abundance observed in the areas. The vegetation in site 5 may also be contributory to the presence of more mosquito larvae, as the presence of vegetation may have provided shelter for the larvae against its predators.

We likewise observed that, in spite of the habitat heterogeneity in the different areas of the freshwater river, only one mosquito larva was observed. The *C.annulirostris* larvae were evident in all study areas examined in the freshwater river. The occurrence of *C. annulirostris* in the area is possible, as a previous report indicated that such mosquito inhabits this particular region of the Philippines. Although, studies (Hall-Mendelin et al., 2012; Van Den Hurk et al., 2008) have indicated that this mosquito has the potential to transmit ailments like the Japanese encephalitis, no report in the Philippines has been indicated yet. This mosquito larva inhabits freshwater environments (Chapman et al., 2003). The *C. annulirostris* larval mosquito abundance in the river was observed to be influenced by two important water quality parameters, namely temperature and dissolved oxygen. Studies have presented that dissolved oxygen (Opoku et al., 2005) and temperature (Beck-Johnson et al., 2013) significantly play an important role in the larval mosquito growth and development. The dynamics of the larval mosquito development are temperature-dependent and dissolved oxygen-dependent. The fairly constant temperature observed in the sites may play an influence on the abundance of the mosquito larvae as temperatures from 20 to 30°C (Beck-Johnson et al., 2013) and low dissolved oxygen (Amala and Anuradha, 2012) in the waters showed the largest abundance of the *Culex* mosquitoes in the area. A study (Silberbush et al., 2015) supports our findings as they hypothesize that as the dissolved oxygen levels in the water decreases, the larvae had access to atmospheric oxygen responsible for increasing their development in the water. Temperature likewise plays an important role in the survival and development of the mosquito larvae as a study (Dodson et al., 2012) indicated that temperature plays a role in the pupation and emergence of the mosquito. The temperature we observed in the water contributed to the development of the adults in the larvae. It likewise provides an environment that facilitates the growth of the microorganisms providing the necessary food resources for the mosquito larvae's sustenance in the water (Teklu et al., 2010). The larval mosquito variation in the water, as we observed, appears to be contributed by an interplay of a variety of conditions emanating in the water body that includes biotic (aquatic life and vegetation) and abiotic (water quality parameters and

environmental conditions) factors (Opoku *et al.*, 2005; Sia Su *et al.*, 2012). Studies support our results indicating that the water quality parameters (Noori *et al.*, 2015) and the environmental conditions (Teklu *et al.*, 2010) are essential for the mosquito larvae as it provides the necessary conditions and resources for its survival and development.

Conclusion

In conclusion, this study highlights the abundance of *C. annulirostris* larval mosquitoes in the freshwater river as influenced by both biotic and abiotic factors that emanate in the water body. The study likewise shows that temperature and dissolved oxygen are the factors that significantly influence the abundance of the *C. annulirostris* larval in the river. Despite habitat heterogeneity, larvae of *C. annulirostris* inhabited the waters. Further mosquito surveillance and environmental monitoring of freshwater environments are needed to continuously monitor the occurrence of mosquito vectors that may have the capability to bring threats to human health.

REFERENCES

- Sanford, M.R., K. Chan, and W.E. Walton, 2005. Effects of inorganic nitrogen enrichment on mosquitoes (Diptera: Culicidae) and the associated aquatic community in a constructed treatment wetland. *J. Med. Entomol.*, 42(5): 766-76.
- Schafer, M.L., J.O. Lundstrom, M. Pfeffer, E. Lundkvist, and J. Landin, 2004. Biological diversity versus risk for mosquito nuisance and disease transmission in constructed wetlands in southern Sweden. *Med. Vet. Entomol.*, 18(3): 256-67.
- Greenway, M., P. Dale, and H. Chapman, 2003. An assessment of mosquito breeding and control in four surface flow wetlands in tropical-subtropical Australia. *Water Sci. Tech.*, 48(5): 249-56.
- Muturi, E.J., J. Mwangangi, J. Shililu, B.G. Jacob, C. Mbogo, J. Githure, and R.J. Novak, 2008. Environmental factors associated with the distribution of *Anopheles arabiensis* and *Culex quinquefasciatus* in a rice agro-ecosystem in Mwea, Kenya. *J. Vect. Ecol.*, 33: 56-63.
- Okogun, G.R.A., B.E.B. Nwoke, A.N. Okere, J.C. Anosike, and A.C. Esekhegbe, 2003. Epidemiological implications of preferences of breeding sites of mosquito species in Midwestern Nigeria. *Ann. Agric. Environ. Med.*, 10: 217-222.
- Kengluetcha, A., P. Singhasivanon, M. Tiensuwan, J. Jones, and R. Sithiprasasna, 2005. Water quality and breeding habitats of anopheline mosquito in northwestern Thailand. *Southeast Asian J. Trop. Med. Pub. Hlth.*, 36(1): 46-53.
- O'Malley, C., 1989. Guidelines for larval surveillance. *Proc. 76th Annual Meeting New Jersey Mosquito Control Assoc., Inc.*, pp. 45-55.
- Delfinado, M.D., 1966. The Culicine mosquitoes of the Philippines, Tribe Culicini (Diptera, Culicidae). *Mem. Amer. Entomol. Institute*, 7: 139-140.
- Belkin, J.N., 1954. Simple larval and adult mosquito indexes for routine mosquito control operations. *Mosq. News.*, 14: 127-131.
- Chapman, H.F., J. Hughes, S.A. Ritchie, and B.H. Kay, 2003. Population structure and dispersal of the freshwater mosquitoes *Culex annulirostris* and *Culex palpalis* (Diptera: Culicidae) in Papua New Guinea and Northern Australia. *J. Med. Entomol.*, 40(2): 165-9.
- Opoku, A.A., O.D. Ansa-Asare, and J. Amoako, 2005. The occurrences and habitat characteristics of mosquitoes in Accra, Ghana. CSIR-Water Research Institute, Ghana. Available from <http://www.wajae.org>
- Sia Su, G.L., A. Beronilla, and K.B. Yao, 2012. Water quality and *Aedes* larval mosquito abundance in Caloocan City, Philippines. *Dengue Bull.*, 36: 175-181.

- Beck-Johnson, L.M., W.A. Nelson, K.P. Paaijmans, A.F. Read, M.B. Thomas, and O.N. Bjørnstad, 2013. The effect of temperature on *Anopheles* mosquito population dynamics and the potential for malaria transmission. PLoS ONE, 8(11): e79276. doi:10.1371/journal.pone.0079276
- Amala, S., and V. Annuradha, 2012. Species composition and diversity of mosquitoes in selected areas of Vellimalai in Sirumalai Hills. Int. J. Biol. Med. Res., 3(1): 1281-1283.
- Silberbush, A., Z. Abramsky, and I. Tsurim, 2015. Dissolved oxygen levels affect the survival and development period of the mosquito *Culex pipiens*. J. Vect. Ecol., 40(2): 425-427.
- Dodson, B.L., L.D. Kramer, and J.L. Rasgon, 2012. Effects of larval rearing temperature on immature development and West Nile virus vector competence of *Culex tarsalis*. Paras. Vect., 5: 199.
- Noori, N., B.G. Lockaby, and L. Kalin, 2015. Larval development of *Culex quinquefasciatus* in water with low to moderate. J. Vect. Ecol., 40(2): 208-220.
- Hall-Mendelin, S., C.C. Jansen, W.Y. Cheah, B.L. Montgomery, R.A. Hall, S.A. Ritchie, and A.F. Van Den Hurk, 2012. *Culex annulirostris* (Diptera: Culicidae) Host feeding patterns and Japanese encephalitis virus ecology in Northern Australia. J. Med. Entomol., 49(2): 371-377.
- Van Den Hurk, A.F., S.A. Ritchie, C.A. Johansen, J.S. Mackenzie, and G.A. Smith, 2008. Domestic pigs and Japanese encephalitis virus infection, Australia. Emerg. Infect. Dis., 14(11): 1736-38.
- Teklu, B.M., H. Tekie, M. McCartney, and S. Kibret, 2010. The effect of physical water quality and water level changes on the occurrence and density of *Anopheles* mosquito larvae around the shoreline of the Koka reservoir, central Ethiopia. Hydrol. Earth Syst. Sci., 14: 2595-2603.