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Identification of mosquito species in Cebu, Philippines

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Abstract
Mosquitoes are insects belonging to order Diptera, suborder Nematocera, and family Culicidae. Some species of which are important vectors of diseases. Identification of mosquito species based on morphological characteristics was conducted in Cebu, Philippines. Four municipalities of Cebu and two selected Barangays of Cebu City were chosen for the study. The municipalities include: Toledo, Argao, Naga, Lapu-Lapu and Liloan. The 2 selected Barangays of Cebu City, are; Lahug, and Toribio Padilla. Each of possible breeding places of mosquitoes were identified and examined for the presence of mosquito larvae in all sampling areas. Description of breeding sites included: type of breeding site (barrels, discarded tires, tin cans, etc.). There are four mosquito species identified during the 3 quarters of sampling period. The following mosquito species that identified are: *Aedes aegypti*, *Aedes albopictus*, *Culex pipiens pipiens* and *Culex pipiens quinquefasciatus*. *Aedes aegypti* species is present in all sampling areas. The breeding sites that were identified during the sampling areas were clean and stagnant water like metal drums and discarded tires which are favorable breeding sites of *Ae. aegypti*. However, *Aedes albopictus* and *Culex pipiens pipiens* are absent in Barangay Lahug and *Culex pipiens quinquefasciatus* is absent in Toledo and Naga. These mosquito species breed in dirty and polluted water like canals. In the said Barangays, there were canals found to be existing, however, the water present in the canals was flowing and for this reason, the mosquito failed to lay their eggs.

Keywords: *Aedes aegypti, Aedes albopictus*, *Culex pipiens pipiens, Culex pipiens quinquefasciatus,* Mosquitoes of Cebu, Philippines

Introduction
Mosquitoes (family Culicidae) are at the center of worldwide medical entomological research primarily because of their importance as vectors of dangerous diseases, such as malaria, dengue, yellow fever, encephalitis, lymphatic filariasis and chikungunya. They play a major role in life quality and health of humans. Mosquitoes are tiny insects but the damage they do on mankind is enormous. There are over 3,200 species of mosquitoes in the world. Of this figure, approximately 1,000 species are considered as vectors or carriers of pathogens and about 60 are considered dangerous because they are responsible for transmission of pathogens that can kill humans. Some species of mosquitoes that belong to the top 60 are; Aedes aegypti (main carrier of dengue and yellow fever viruses), Anopheles gambiae and An. funestus (vectors of malaria and filariasis), Culex pipiens complex (responsible for filariasis and arboviruses), and Aedes albopictus (vector of epidemic chikungunya).
In terms of morbidity and mortality caused by vector-borne diseases, mosquitoes are the most dangerous animals confronting mankind. They threaten more than two billion people in tropical and subtropical regions, and have substantially influenced the development of mankind, not only socio-economically but also politically. More than half of the world's population lives under the risk of becoming infected by the causative agents of these diseases. Estimates made by the World Health Organization (WHO), show that many hundreds of millions of people become ill, and some millions of people die annually (WHO, 2015). As human populations in developing countries continue to increase, breeding sites for urban mosquito species such as trash dump sites and stagnant drainage canals grow with them (WHO, 2014).
Dengue fever (DF) and dengue haemorrhagic fever (DHF) have emerged as major public health problems. They are caused by one of four closely related, but anti-genetically distinct virus serotypes (DEN1-, DEN-2, DEN-3, and DEN-4), of the genus Flavivirus (CDC, 2003). Since the first global appearance of DHF in 1953 in Manila, Philippines, 1958 in Bangkok, Thailand, and 1968 in Surabaya, Indonesia, it is presently considered as one of the leading

viral diseases causing hospitalization and death among children and adults in 100 tropical and subtropical countries throughout the Americas, South-East Asia, the Western Pacific Islands and Africa. About 500,000 people are hospitalized (95% of those affected are children) and about 24,000 fatalities are reported annually (WHO, 2015).

Dengue fever outbreaks continue to rise in South America and Asia. Among the affected countries in Asia, the Philippines is considered as one of the "high risk" zones. The resurgence of dengue, can be traced to rapid urbanization, poor sewage systems, and improper disposal of garbage. The persistence and numerical increase of *Aedes (Stegomyia) aegypti* (Linnaeus) as a primary vector and *Aedes (Stegomyia) albopictus* (Skuse) as a secondary vector is partly attributed to sanitary and hygienic practices. Urbanization has left many households with inadequate water supplies and has hastened the spread of the virus. Unfortunately, the majority of the people do not realize the seriousness of the situation until they become infected. Many people dispose of their garbage and waste just outside their door step, onto the streets, and in vacant lots. Eventually, the improperly disposed waste accumulates water during rainfall, becoming potential breeding sites for these vector mosquitoes.

An increase of population growth in Cebu, Philippines has somehow contributed to the rising mosquito diseases like dengue. As based on National Statistics Office (NSO, 2010), in year 2000, there were 2,160,569; 2007-2,440,120; and in 2010-2,619,362 with a population density of 780.1 persons per Km^2. In regards to the dengue incidences, the Department of Health Regional Epidemiology and Surveillance Unit in Cebu (DOH-RESU, 2014) reported 17,974 of January to October 15, 2016 resulting to 144 deaths. As the number of population increases in this area, dengue incidence likewise increases.

Although there were already published studies on the identification of mosquitoes in the Philippines but still present data is lacking on the mosquito species present in Cebu Province.

This study aims to identify the mosquito species of Cebu, Philippines. Through the identification of mosquito species, a listing of probable diseases that these mosquito species will bring to the inhabitants will be documented. Furthermore, once the mosquito species and their respective breeding places will be identified, proper actions or measures could be implemented for reducing their numbers in order to lessen the morbidity and mortality rates of infections (especially dengue) to a level wherein it will no longer be a public health problem. Lastly, results of this study will serve as a major tool for developing key program strategies for an integrated vector control approach.

Methodology

A. Research Areas

The study was conducted in Cebu, Philippines. Four municipalities of Cebu and two selected Barangays of Cebu City were chosen for the study. The municipalities are: Toledo, Argao, Naga, Lapu-Lapu and Liloan. In addition, two selected Barangays of Cebu City namely; Lahug, and T. Padilla were included (Figure 1). These areas were chosen due to their high incidences of dengue fever (DOH RESU, 2016) and were representing different geographical location.

Fig 1: Map of Cebu Province showing 6 areas as sampling places.

Legend: T. Padilla Toledo City Argao Naga City Lahug Liloan Lapu-lapu.

B. Data Collection and Analyses

Identification of mosquito breeding places

Every possible breeding place of mosquitoes were identified and examined in all sampling areas (Figure 1). Description of breeding sites included: type of breeding site (barrels, discarded tires, tin cans, etc.).

Collection of mosquito larvae, Preservations, Identification

Sampling was conducted in every three months from August 2014 - April 2015 (representing both dry and rainy seasons). Third to fourth instar larvae of mosquitoes were collected from various breeding sites of the sampling areas. This was done by using a prescribe mosquito dipper from World Health Organization (WHO). The collected larvae were kept in glass bottles with their corresponding information on the collection. They were stored in small vials with 70% alcohol until identification of species was done.

Mosquito samples were identified until species level by using a stereoscope following the taxonomic key of Becker *et al.*, 2007.

Results

There are four mosquito species identified during the sampling period. The following species are: *Aedes aegypti*, *Aedes albopictus*, *Culex pipiens pipiens* and *Culex pipiens quinquefasciatus* (Figure 2). A total of 417 mosquito mosquito species were collected and identified. Out of the figure, there are 268 *Ae. aegypti*, 35 *Ae. albopictus*, 58 *Cx. pipiens pipiens*, and 56 *Cx. pipiens quinquefasciatus*. The species of *Aedes aegypti* were found to be present in all sampling areas. *Aedes albopictus* and *Culex pipiens pipiens* species were absent in Barangay Lahug. *Culex pipiens quinquefasciatus* was absent in Toledo and Naga. The various species of mosquito exhibit different morphological characteristics (Figures 3-6).

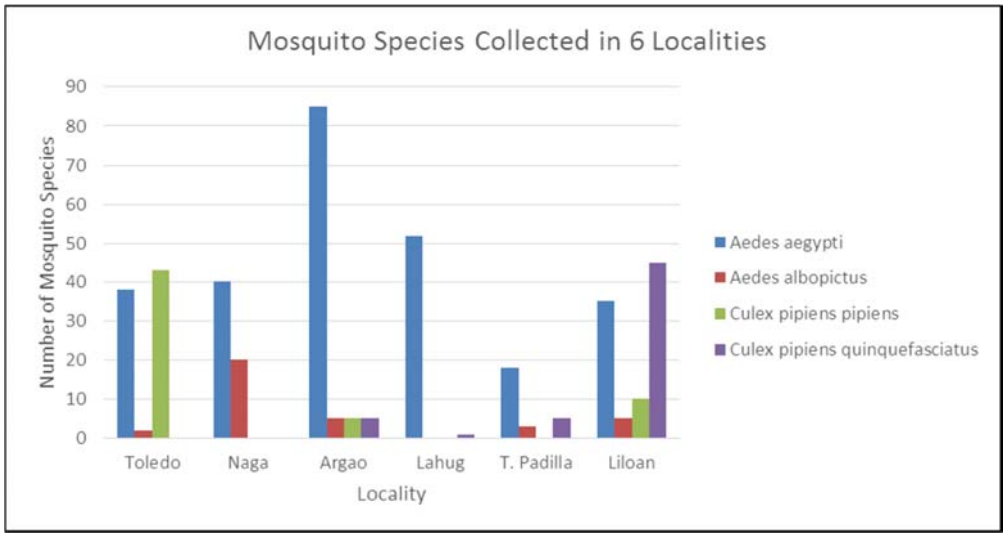

Fig 2: Mosquito species collected in 6 localities.

Key to the Fourth-Instar Mosquito Larvae Genera

1. Siphon with several pairs of siphonal tufts……..*Culex*

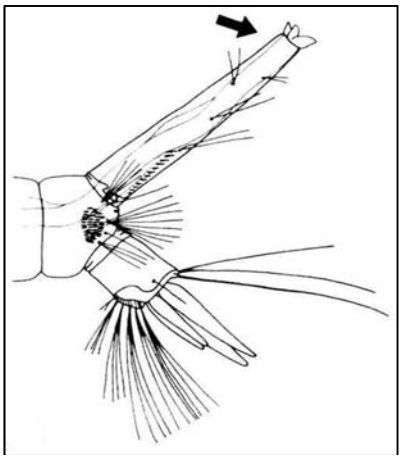

Fig 3: Siphon of *Culex pipiens pipiens*

2. Base of siphon without acus……*Aedes aegypti*

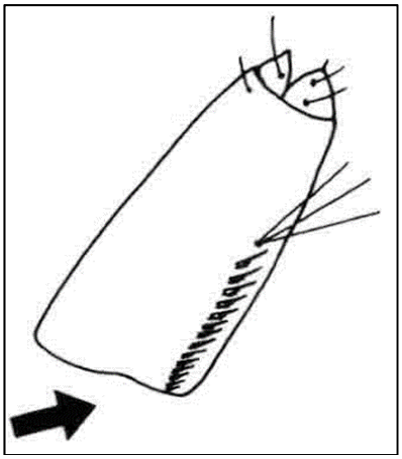

Fig 4: Siphon of *Aedes aegypti*

3. Antennal seta simple, Siphonal tuft inserted slightly beyond the middle of the siphon, distal to the pectin…….Aedes albopictus

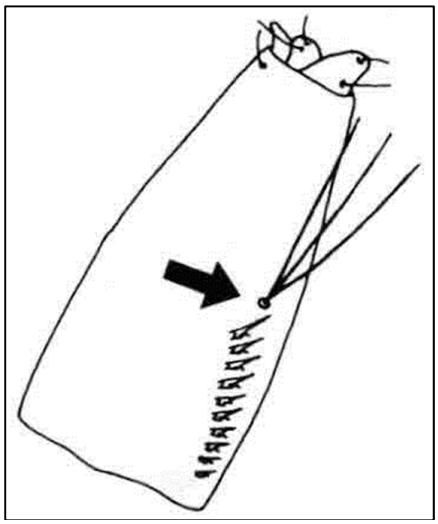

Fig 5: Siphon of *Aedes albopictus*

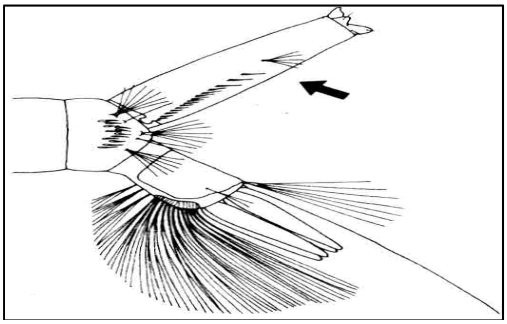

Fig 6: Abdominal segment VIII of *Aedes aegypti*

Discussion

Aedes aegypti species were present in all sampling areas could be explained that this species has a very wide range of habitat tolerance in both urban and rural areas. During the study, breeding sites that were found in the sampling areas were clean and stagnant water like metal drums, discarded tires, plastic containers, etc. that are

favorable breeding sites of *Ae. aegypti*. Another factor that contributed to the presence and abundance of this species is the irregular supply of water in the areas which enabled the residence to store water in drums or plastic container without putting lid on it. This practice is a contributing factor to the spread of *Ae. aegypti* mosquito.

Aedes albopictus was absent in Barangay Lahug can be explained that this species favors to grow in rural areas and hence Barangay Lahug is an urban area, this will support why this species was absent.

Culex pipiens quinquefasciatus was absent in Toledo and Naga could be explained that this species breeds in dirty and polluted water like canals, however in the said Barangays, there were canals existing but the water was flowing and the mosquito failed to lay their eggs.

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