Annual Report of the LaMer Project FY 2023

1. Title of the research project:

Insecticide resistance status and future insecticide choices for vector control strategies in Bangladesh.

2. Names and affiliation of members:

1. Prof. Kozo Watanabe, Full Professor of Molecular Ecology and Health (MEcoH) Laboratory at the Center for Marine Environmental Studies (CMES), Ehime University, Japan.

2. Dr. Ashekul Islam, Assistant Professor and Principal Investigator of Vector Borne Diseases Laboratory at the Department of Biochemistry and Molecular Biology, Mawlana Bhashani Science and Technology University, Tangail 1902, Bangladesh.

3. Mohammad Mosleh Uddin, PhD student, Molecular Ecology and Health (MEcoH) Laboratory at the Center for Marine Environmental Studies (CMES), Ehime University, Japan.

4. Md. Sujan Islam, MS thesis student, Vector Borne Diseases Laboratory at the Department of Biochemistry and Molecular Biology, Mawlana Bhashani Science and Technology University, Tangail 1902, Bangladesh.

5. Sadia Afrin, MS thesis student, Vector Borne Diseases Laboratory at the Department of Biochemistry and Molecular Biology, Mawlana Bhashani Science and Technology University, Tangail 1902, Bangladesh.

6. Maria Mehjabin Anki, MS thesis student, Vector Borne Diseases Laboratory at the Department of Biochemistry and Molecular Biology, Mawlana Bhashani Science and Technology University, Tangail 1902, Bangladesh.

3. Purposes:

Monitor and analyze trends in insecticide resistance among local mosquito populations, particularly in *Culex spp.* and *Aedes aegypti*, to inform and optimize vector control strategies. This approach should include periodic surveillance, resistance mechanism studies, and the evaluation of alternative control methods to ensure effective and sustainable vector management.

4. Methods:

Mosquito larvae collection and rearing:

Mosquito larvae were collected from several locations in Tangail Sadar and Dhaka, Bangladesh, and reared into adults at the Vector Borne Disease Laboratory (VBDL) of Mawlana Bhashani Science and Technology University (MBSTU). The colonies were maintained independently, fed with either a 10% sugar solution or anesthetized mice blood, following the methods previously described by Min-Lin Zheng et al. [1].

Adulticide action under laboratory conditions:

The mosquito populations were assessed for susceptibility to Deltamethrin and Dimefluthrin by measuring mortality rates at 30 minutes. According to WHO and CDC guidelines, populations with mortality rates below 90% were classified as resistant, those with rates of 98% or higher were deemed susceptible, and mortality rates between 90% and 97% suggested developing resistance [2, 3].

Results:

Adulticide action using Deltamethrin:

In our study on the effectiveness of Deltamethrin and Dimefluthrin, we found that both *Ae. aegypti* and *Culex spp.* demonstrated resistance to Deltamethrin. At a diagnostic time (DT) of 30 minutes and using the CDC-recommended diagnostic dose of 10µg/ml per 250 ml bottle, the laboratory-reared *Ae. aegypti* population had a mortality rate of 61.5% at 1× concentration and 72.727% at 5× concentration (Figure 01).

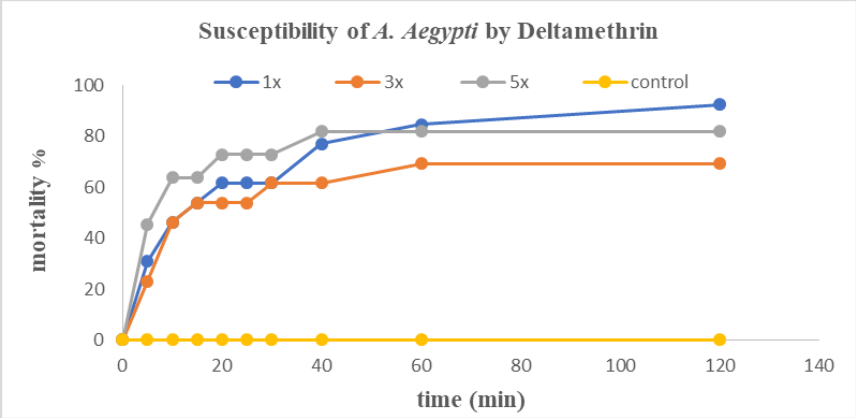

Figure 01: Susceptibility of *A. aegypti* by Deltamethrin

Mosquito survival past the diagnostic time (DT) suggests resistance. To assess resistance strength, we extended observations beyond the DT. According to CDC guidelines, the failure to achieve 100% mortality at any dose (1×, 3×, or 5×) indicates significant resistance. Similar patterns were observed in wild-type *Culex spp.*; at a DT of 45 minutes with a dose of 10µg/ml per 250 ml bottle, mortality was only 26.0%, and no dose reached 100% mortality, underscoring the extensive resistance (Figure 02).

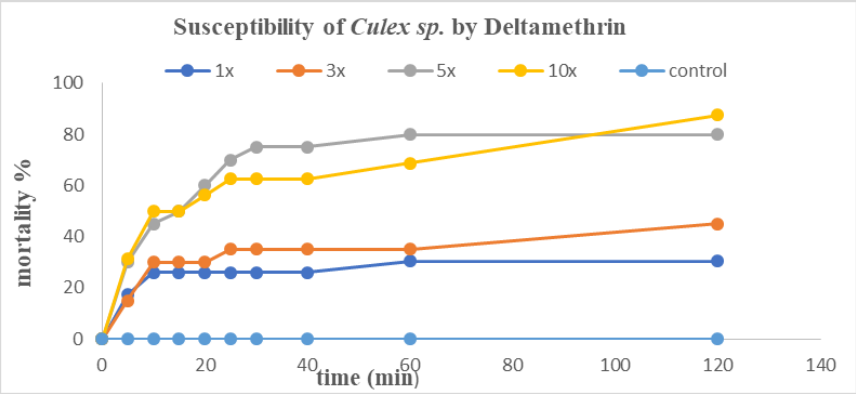

Figure 02: Susceptibility of *Culex spp.* by Deltamethrin

Adulticide action using Dimefluthrin:

We tested Dimefluthrin at different concentrations (2µg/ml, 4µg/ml, 6µg/ml, 10µg/ml) and at higher levels (100µg/ml, 200µg/ml, 300µg/ml) similar to those found in commercial mosquito coils. Our data indicate that both *Ae. aegypti* and *Culex spp.* are highly susceptible to Dimefluthrin. For *Ae. aegypti*, the maximum survival time was 25 minutes at the 2µg/ml dose, with knockdown occurring before 15 minutes at higher doses (Figure 03).

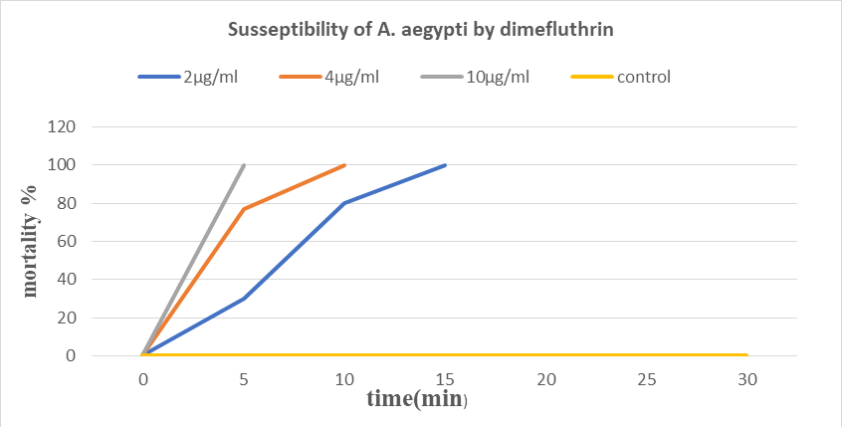

Figure 03: Susceptibility of *A. aegypti* by Dimefluthrin

Culex spp. showed a similar pattern, with the longest survival at 2µg/ml for 15 minutes and knockdown before 15 minutes at all higher doses (Figure 04).

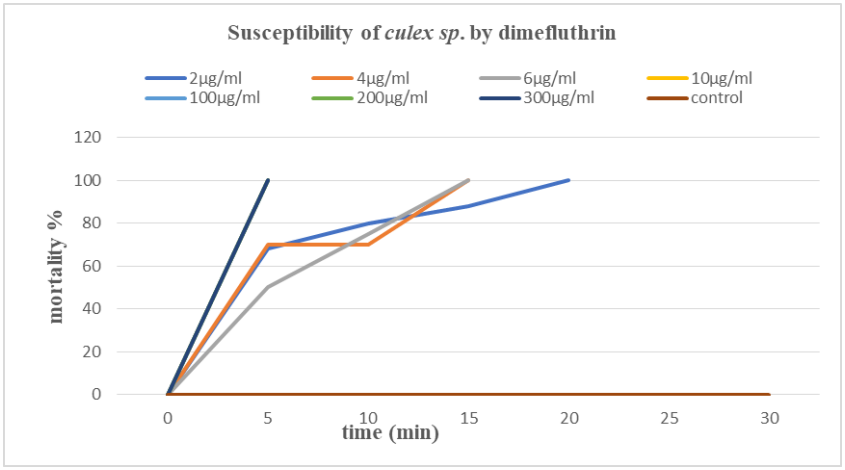

Figure 04: Susceptibility of *Culex spp.* by Dimefluthrin

Future Challenges:

Although laboratory bioassays provide growing data on insecticide resistance in mosquito vectors, they fall short of guiding stakeholders on effective vector control strategies. To address practical resistance, standardization at the local level is crucial, enabling national programs to assess the changing efficacy of control measures over time. Therefore, assessing the real-world impact of insecticide use requires comprehensive studies incorporating surveillance, disease prevalence, and resistance mechanisms.

References:

1. Zheng ML, Zhang DJ, Damiens DD, Lees RS, Gilles JRL. Standard operating procedures for standardized mass rearing of the dengue and chikungunya vectors Aedes aegypti and Aedes albopictus (Diptera: Culicidae) - II - Egg storage and hatching. Parasites and Vectors. 2015;8:1–7. doi:10.1186/S13071-015-0951-X/TABLES/3.

2. Al-Amin HM, Johora FT, Irish SR, Hossainey MRH, Vizcaino L, Paul KK, et al. Insecticide resistance status of Aedes aegypti in Bangladesh. Parasit Vectors. 2020;13. doi:10.1186/S13071-020-04503-6.

3. Al-Amin, H.M., Gyawali, N., Graham, M., Alam, M.S., Lenhart, A., Hugo, L.E., Rašić, G., Beebe, N.W. and Devine, G.J. (2023), Insecticide resistance compromises the control of Aedes aegypti in Bangladesh. Pest Manag Sci, 79: 2846-2861. https://doi.org/10.1002/ps.7462