Test of an Exomic Capture Assay To Identify and Sequence Insecticide Resistance Genes in Non-Model Mosquitoes

Kaitlyn Fang, Nicole Wagner, Dina M. Fonseca
Cornell University and Rutgers, The State University of New Jersey

While yellow fever, dengue, and Zika are transmitted between humans by human biting mosquitoes, West Nile virus, eastern equine encephalitis and many other deadly human pathogens are zoonosis. The amplifying hosts of the viruses are birds and critical transmission among birds and spillovers to humans occurs by mosquitoes, such as *Culex restuans,* for which genomic information is very limited. We tested the use of a capture assay developed for *Culex pipiens* group mosquitoes, that among others targets all ~350 genes known to be associated with insecticide resistance in mosquitoes. We developed a side-by-side comparison by testing the bait capture in specimens from the *Cx. pipiens* group and *Cx. restuans.* We developed NextGen genomic libraries from existing DNA samples. After the bait hybridization with a temperature of 65℃ the average DNA concentration for *Cx. quinquefasciatus* was 6.98 ng/µL, for *Cx. pipiens f. molestus* was 5.48 ng/µL while the *Cx. restuans* was too low for readings to be measured even with a highly sensitive Qubit Fluorometer. To address this, we lowered the hybridization temperature to 60℃, therefore lowering the stringency and allowing slightly variable sequences to still anneal to the baits. The resulting DNA concentrations for the *Cx. restuans* were on average 2.76 ng/µL. In conclusion, the capture assay was successful although subsequent NextGen sequencing will determine the extent of the success. These results suggest that the gene-based capture assay is a cost-effective strategy to obtain information on insecticide resistance in mosquitoes, and ultimately help develop better strategies to limit its spread. Supported by R25ES020721.