Selection and Enzyme Assays of Pyrethroid Resistance in *Anopheles minimus* Colony

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Abstract

This study was conducted to test susceptibilities of *Anopheles minimus* species A mosquitoes following exposures to deltamethrin, during each of 19 generations. The LD_{50} and LD_{90} (or LT_{50} and LT_{90}) values were determined for populations from each subsequent generation by probit analysis and significant increases occuring from one generation to the next. They were analyzed by chi-square test ($P<0.01$). Selection for resistance via the World Health Organization test protocol (was by exposing), sequential generations of *An. minimus* females to LD_{50} and LT_{50} values of deltamethrin. There was approximately a 26-fold increase in the LD_{50} and a 23-fold increase in LD_{90} when the F_{10} generation was compared to the parent colony (F_1). Similarly, the LT_{50} and LT_{90} values were also increased during selection experiments from generations 14-19. There was roughly a 3-fold increase in LT50 and LT90 values of F19 females compared to F_{14} females.

In addition, enzyme-based mechanisms of insecticide resistance were performed on susceptible and resistant colonies of *An. minimus* to deltamethrin using biochemical assay. Three enzyme assays, esterase, monooxygenases and glutatione S-transferases, were performed on 4 test populations (F_0, F_8, F_{12} and F_{18}). F_0 was found completely susceptible to deltamethrin, whereas F_8, F_{12} and F_{18} demonstrated levels of tolerance/resistance to deltamethrin. Monooxygenases (MFOs) activity was continuously elevated in resistant test populations (F_8, F_{12} and F_{18}) than those from the parent colony (F_0). There was a 5-fold increase in specific activity of MFOs in F_{18} compared to the control colony (F_0). Specific activities of alpha and beta-esterases as measured by the

hydrolysis of alpha and beta-naphthyl propionate to naphthol showed it was unclear whether it is responsible for pyrethroid resistance. Glutathione S-transferases (GSTs) were not elevated in the 4 resistant test populations. Based on our results, it is more likely that the development of physiological resistance to deltamethrin may be related to elevated MFOs activity.