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PI : Smartt, Chelsea T
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The emergence of DDT and malathion resistance brought about the use of pyrethroid impregnated bed nets to control the spread of malaria. Now the emergence of pyrethroid resistant *Anopheles* has impacted malaria control efforts. Low levels of resistance to organophosphates in *Anopheles* from many parts of the world have been detected and most recently a population of *An. crucians* in Florida exhibited resistance to Dibrin. Dibrin resistant *An. crucians* were obtained from Manatee County Mosquito Control. RT-PCR on the RNA from the resistant mosquitoes was performed using primers that specifically amplify esterase alpha and beta and aldehyde oxidase (AO). Analysis of the PCR reaction revealed that the probable resistant mechanism for these mosquitoes was esterase alpha overexpression.

My laboratory has been determining the insecticide resistance mechanism used by Florida mosquitoes to detoxify organophosphates. Using lab *Culex nigripalpus* strains as models, we amplified esterase Beta and aldehyde oxidase (AO) from genomic DNA and esterase alpha from RNA. The above mentioned PCR fragments were cloned and sequence analysis performed. Sequence analysis of our putative esterase Beta did not reveal any significant matches to known sequences in GenBank. Further characterization on this clone will be forthcoming. Sequence analysis of our putative AO from *Cx. nigripalpus* genomic DNA showed similarity to a family of transposable elements containing endonuclease/exonuclease and phosphatase activity, which is indicative of enzymes active in signal transduction. Although our clone was not AO, its similarity to a transposable element is important because transposable elements have been found closely associated with the xenobiotic-metabolizing cytochrome P450 genes. In fact, the transposable elements have been found inserted in the 5'-, 3'-untranslated regions, in exons and in introns.

Wild female *Cx. nigripalpus* were collected in Indian River County and some used to isolate genomic DNA and RNA, the rest were frozen at -80C. Total RNA was isolated from field collected mosquitoes and RT-PCR to generate esterase and AO transcripts carried out. PCR amplified a putative esterase Beta and AO. Esterase Beta was isolated and cloned and will be further characterized by sequence analysis. The AO PCR fragment will be cloned and also subjected to sequence analysis.

During the summer of 2007, an undergraduate student from Bates College, with funding through a Hughes Summer Research Fellowship, worked in my lab and initiated the characterization of the the organophosphate resistant mechanism used by field collected

Anopheles crucians from Manatee County. Due to the lack of rain, we were not able to obtain wild OP-resistant *An. crucians* during her time in my lab. Because of this, genomic DNA from *An. crucians* was not analyzed for gene expression of esterase and AO. The student was able to use RNA previously isolated from the resistant *An. crucians*, to carry out RT-PCR analysis of the above mentioned genes. Esterase Alpha was amplified from *An. crucians* RNA and cloned. Sequence analysis of these clones revealed similarity to proteins involved in neuronal sensory and metabolic processes. Their implication in *An. crucians* insensitivity to OP will be investigate.