

## **UTILIZING MOSQUITOES IN HUMAN IDENTIFICATION**

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Mosquitoes are often considered as pests and vectors of diseases such as malaria, dengue, zika, and West Nile viruses. However, female mosquitoes, particularly *Anopheles stephensi*, can be useful in obtaining human DNA due to the nature of their food: human blood. Nucleated white blood cells found in the blood in the midgut of these insects can be used to identify the donor on which the female mosquito has fed.

Generation of short tandem repeat (STR) DNA profile is useful for identification of the donor of the blood on which the mosquito has fed by using direct amplification and capillary electrophoresis. Commercially available direct amplification kits allow for detection of the donor of a body fluid within a very short period of time as it bypasses the labor intensive and time consuming steps needed for extraction and quantification of DNA.

Previous research in this laboratory using another amplification kit indicated that DNA starts to degrade after 8 hours post-feed and is completely degraded 72 hours post-feed. Based on those results, mosquitoes were fed on warm blood meal and euthanized at 0 hours, 4 hours and 8 hours in this current project.

COPAN microFLOQ® direct collection devices contain a Nylon-fiber swab with lysis agent at the tip which allows the direct amplification process. These devices were used to puncture the midgut of each mosquito and a minute quantity of blood from the midgut was collected on the tip. The tips, containing the bloodstains, were broken off and put into the PCR amplification tubes containing direct amplification reagents. They remained inside the tubes during the thermal cycling process. Some of these samples were single source and others were fed on a 1:1 mixture of blood of two donors. Blood samples from each donor were used as reference.

This research indicated that DNA profiles obtained from the mosquitoes were consistent between and within each donor. In addition, both donors were identified in the mixture profiles for all euthanization time periods. It was concluded that DNA contained in the abdomen of the mosquitoes can be used to determine the donors' identity using direct amplification. This is a robust method to establish the presence of one or more individuals at a location of interest within a specific time window.