



Speaker:

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Date: Monday, November 28, 2022

Time: 4:00 pm - 4:50 pm

Format: In-Person Seminar & Virtual Access

Location: Genomics Auditorium 1102A

Zoom: 938 1040 4405

Passcode: 833289

Title:

“CRISPR-mediated gene editing in *Homalodisca vitripennis* for the genetic control of Pierce’s disease”

Abstract:

Homalodisca vitripennis (glassy-winged sharpshooter, GWSS) is an important agricultural pest that infests over 100 different plant species. GWSS is an efficient vector of the bacterium *Xylella fastidiosa*, the causal agent of Pierce’s disease of grapevine, Citrus Variegated Chlorosis, and other important diseases. As resistance of GWSS to chemical insecticides emerges, genetic approaches to the control of GWSS are needed. We established parameters for efficient genome editing by CRISPR gene-editing technology using the white (*w*) and cinnabar (*cn*) genes as targets. Knock-out mutants with mosaicism of eye color were obtained at high frequency at the cinnabar and white loci. Stable lines were established and maintained to the G8 (*cn*) and G10 (*w*) generations. To restore the wild-type phenotype, oligonucleotides containing wild-type and barcode sequences were microinjected into *w* and *cn* mutant embryos with Cas9 protein and specific sgRNAs. Restoration of wildtype eye color was obtained. Amplicon sequencing confirmed recombination of the oligonucleotides into the GWSS genome via homologous directed recombination. A CRISPaint strategy (utilizing non-homologous end-joining DNA repair) was used to integrate a reporter gene, mCherry, under the control of a whitefly promoter into the *cn* and *w* genes. Evidence of integration mCherry was obtained by PCR. Our results illustrate the potential of GWSS to become a model organism within the Hemiptera and for the harnessing of CRISPR technology for the development genetic control strategies for Pierce’s disease.

Refreshments will be served in the Entomology Building Courtyard at 3:30pm