**Research Project:** IPM TECHNOLOGIES FOR SUBTROPICAL INSECT PESTS

**Location:** Subtropical Insects Research

**Title:** Gene expression in midgut tissues of Diaphorina citri: application to biology and vector control

**Authors**
- Hunter, Wayne
- Hert, Mizuri
- Shelby, Kent
- Coudron, Thomas

**Submitted to:** International Research Conference on Huanglongbing

**Publication Type:** Proceedings/Symposium

**Publication Acceptance Date:** November 15, 2008

**Publication Date:** December 1, 2008


**Interpretive Summary:** The Asian citrus psyllid is an important citrus pest because it vectors the bacterium responsible for citrus greening disease, sometimes called huanglongbing. Genes expressed in the alimentary tract of adult Asian citrus psyllid, AsCP, were identified and characterized through sequencing and analysis, which produced a gene dataset of approximately 7,800 ESTs. Enzymes important to digestion and feeding on a phloem diet were identified including several serine proteases, hydrolases, and cathepsins. These and other transcripts with significant homology (E-value =10-20 or better) were identified through similarity searches to other known insect genes in the GenBank database. Use of genomics approaches enabled the identification of some of the genetic basis of psyllid digestion and pathogen interactions. These genomic analyses of psyllid gut enzymes in AsCP have advanced the understanding of feeding and digestion of psyllids. Many of the digestive enzymes have homology across insect species, but there is enough genetic variation to support the development of specialized designed RNAi strategies against specific enzymes within the AsCP. These sorts of novel management strategies for the psyllid are on the horizon. Continued investigations into these genetic
products should provide the information needed for advance studies to isolate AsCP specific genes, which would then be targeted to reduce the spread of HLB and to reduce psyllid populations using environmentally friendly, highly specific management strategies. The newly created dataset was published under accession numbers: FK254041-FK260232, in the National Center for Biotechnology Information (NCBI) public database.

**Technical Abstract:** We produced a gene expression dataset from the midgut tissues of the Asian citrus psyllid (AsCP), Diaphorina citri (Hemiptera: Psyllidae). The AsCP is the primary vector associated with the spread of a devastating citrus trees disease, huanglongbing (HLB). The occurrence and spread of the AsCP and HLB from Florida into other citrus producing states has cause for concern as the disease results in inedible fruit and tree death. HLB is currently a worldwide problem in citrus. There are no effective strategies available for managing the disease except through suppression of psyllids using insecticides (which provides mediocre control of the disease). Thus we are using a genomics approach to identify key genes within AsCP for the development of RNAi strategies to reduce/kill AsCP to reduce HLB spread. We have identified many different enzymes within D. citri. Biological Processes: distribution of Diaphorina citri transcripts from Blastx analysis using a summary with a minimum of 70 sequences per category resulted in these categories in descending order: response to stress 227; proton transport 167; response to chemical stimuli 157; glycolysis 139; instar/pupal development 137; larval development 129; mesoderm development 124; intracellular signaling cascade 118; proteolysis 117; oogenesis 113; behavior 111; amino acid metabolic process 111; proteins amino acid phosphorylation 109; regulation of cellular process 109; cytokinesis 107; DNA metabolic process 105; and monocarboxylic acid metabolism 104. While regulation of enzyme production and release appears to be influenced by feeding, it is likely to also involve a combination of hormonal release, paracrine activity and direct feeding mechanisms. This has been observed in the beetle C. zealandica, where addition of serine protease inhibitors to the diet caused trypsin and leucine aminopeptidase activities to increase. Research has also shown that protease activity can be affected by disease infection of C. zealandica larvae by the bacteria Serratia spp. When Serratia spp contain a specific plasmid, a rapid elimination of serine protease activity in the midgut occurs. Disruption of insect-specific physiological processes has been identified as a useful route for the development of novel management strategies against insect pests. One such area of disruption is to affect the specific enzymes used in digestion which can be inhibited or blocked at synthesis by RNAi approaches. This tactic is promising but has proven more difficult than first envisioned as insects generally produce a range of enzymes under control of multiple genes, thus in-depth genetic studies need to be completed to identify specific enzymes and their biological pathways. The availability of these sequences now enables investigations into these important questions regarding AsCP digestion and biology. The psyllid gene expression data set advances what is currently known about psyllid digestion. The enzymes identified further provide possible genetic targets to be used to alter AsCP digestive enzymes and physiological processes. Dataset was published under accession numbers: FK254041-FK260232, in the National Center for Biotechnology Information (NCBI) public database.