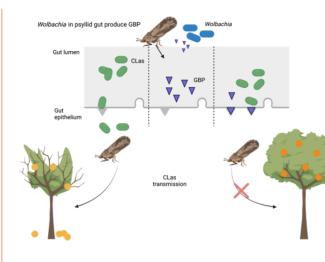
Paratransgenesis for Reducing Transmission of Vector-Borne Candidatus Liberibacter asiaticus



Novel strategy to block transmission of CLas pathogen by 'aummina up' the psyllid gut. A) CLas binds to the gut

surface and enters before it can enter into the psyllid and be transmitted. The psyllid becomes a vector. B) Gut binding peptides (GBP) were selected for binding to the ACP gut to outcompete the pathogen. C) GBP 'gums up' the surface of the gut and prevents CLas binding, thereby blocking the bacterium from entering ACP. GBP produced by Wolbachia in the gut of the psyllid will be tested for CLas transmission blocking. Created with BioRender.com

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Take Home Message:

- Gut binding peptides (GBPs) specifically interfere with CLas by preventing it from leaving the psyllid gut.
- Wolbachia, bacteria that live in the ACP, can be engineered with GBPs to create insects that can't transmit CLas.
- Development of resistance unlikely.

Effort Statement: Identified several promising gut binding peptides that interfere with CLas transmission in feeding assays.

Summary: The overall objective of this research is to discover a novel, biological control-based tool for decreasing pathogen transmission by harnessing native psyllid microbial

flora to reduce psyllid populations and/or pathogen prevalence. One strategy for stopping pathogen spread is to manipulate vector populations by replacing wild populations with altered populations that are unable to transmit a pathogen. The Asian citrus psyllid (ACP) harbor several symbiotic bacteria, including Wolbachia. Because these bacteria are present in all ACP and are not seemingly impacted by the presence of Liberibacter (Las), they provide good candidates for a bacterial symbiont strategy, paratransgenesis. Paratransgenesis involves genetic manipulation of symbiotic bacteria commonly found in pathogen-transmitting vectors to export anti-pathogen molecules into the host vector. This offers several

advantages over the alternative approach, i.e. modification of the vector genome, which involves insertion of novel genes into the heritable genetic material of the vector. In this project, we developed Wolbachia as a driver system for paratransgenesis. We demonstrated that Wolbachia can be used to introduce a phenotype-altering transgene into ACP. Wolbachia cell lines were developed that expressed several unique genes. Paratransgenic transformation of ACP with a gene needed for Las transmission reduced CLas plant infection, as well as acquisition, suggesting that the paratransgenic Wolbachia may be a useful tool for disrupting of insecttransmitted pathogens.

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