Characterizing bacteriophage proteins that hijack arthropod reproduction

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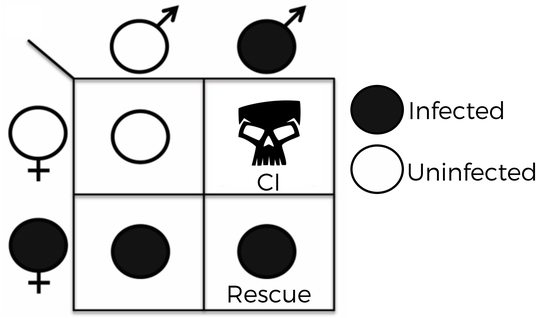
Introduction

Wolbachia are maternally transmitted intracellular bacteria that infect half of insect species.

Cytoplasmic incompatibility (CI) is a crossing incompatibility (shown below) that kills embryos and yields a fitness advantage to infected females



CI is caused by two phage WO genes (*cifA* and *cifB*) and rescue is caused by one gene (*cifA*). CI and rescue can be recapitulated with *cif* transgenes.

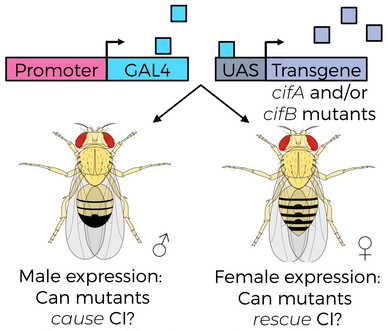
CifA and CifB proteins each have 3 putative domains, but their importance in CI and rescue is unclear.

Question:

What is the relative importance of Cif protein domains in CI and rescue?

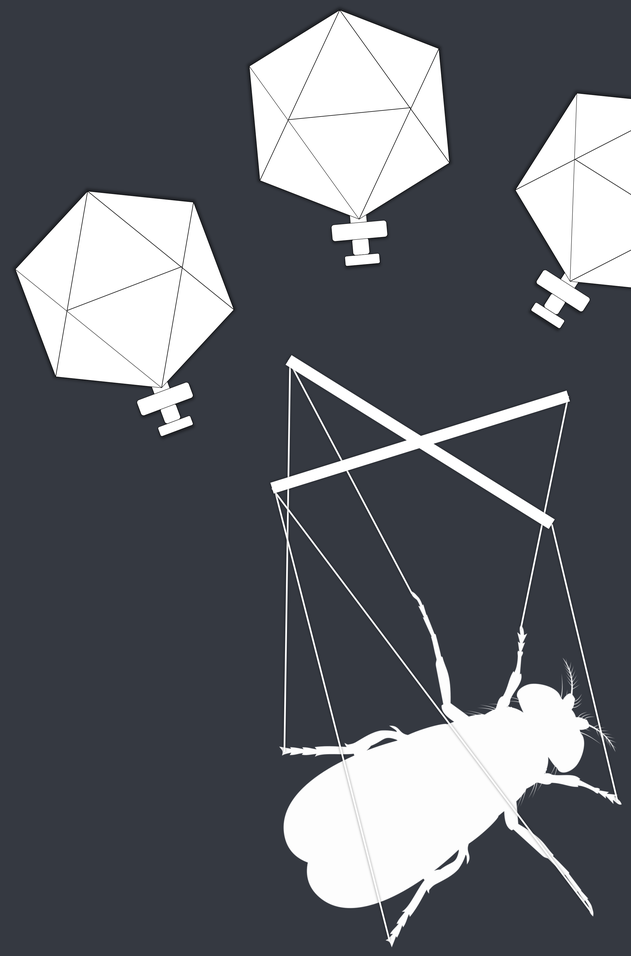
Methodology

Mutate conserved residues in each putative domain and transgenically express wild-type and mutant Cif transgenes and test for CI or rescue.



Assay: Embryo hatching

Site-directed mutagenesis reveals conserved sites in two bacteriophage proteins that are crucial for insect reproductive manipulation.



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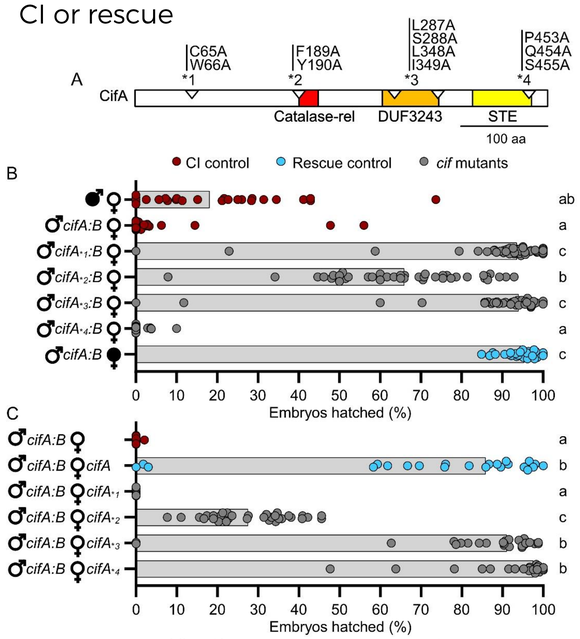
Results

4 mutant CifA lines were made by mutating conserved residues (below, A).

CI was ablated when CifA's N-terminal unannotated region, catalase-rel domain, and DUF were mutated (below, B).

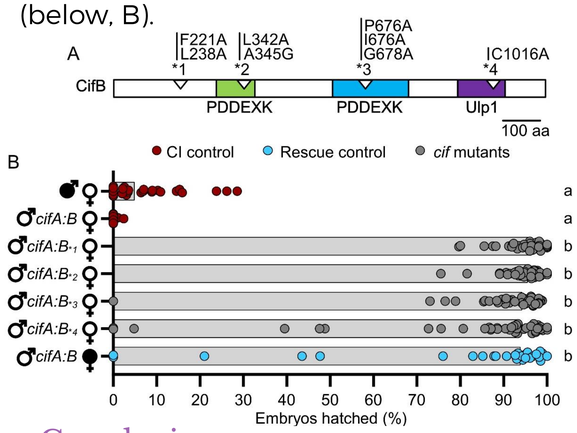
Rescue was ablated when CifA's N-terminal unannotated region and catalase-rel domain were mutated (below, C).

The STE domain does not impact either CI or rescue



4 mutant CifB lines were made by mutating conserved residues (below, A).

CI was ablated by all CifB mutations (below, B).



Conclusions

1) Part of CifA has a dual function in both CI and rescue.
2) Part of CifA is only necessary for CI.
3) Conserved sites in CifB are not amenable to change.