ENTOMOLOGY

Incompatible mosquitoes

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Infection of mosquitoes by a particular bacterium has a fiendishly complicated influence on the success or failure of mosquito breeding. A window now opens on the molecular basis of this 'cytoplasmic incompatibility'.

Cytoplasmic incompatibility (CI) is a well-known phenomenon in many insects and mites[1]. It occurs when an agent inherited in the cell cytoplasm causes matings between strains of the same species to yield few or no offspring, and it is usually associated with infections of a bacterium known as *Wolbachia*. The bacterium lives inside the reproductive cells of its host, and was first associated with CI in a mosquito, *Culex pipiens*, when it was discovered that treating different strains of this species with an antibiotic restored compatibility between them. However, there are complex patterns of cross-compatibility between strains of *C. pipiens* from different populations[2], and these have never been explained.

On page 257 of this issue, Sinkins *et al.*[3] describe how they have used information from *Wolbachia* genome-sequencing projects to link this complexity to genes encoding specific protein motifs — members of the ankyrin family. This research opens up opportunities for understanding both the biological basis of CI and the potential for enlisting *Wolbachia* for mosquito control.

In its simplest form, CI is unidirectional — that is, with two strains of individuals involved in incompatibility, characterized by being either infected or uninfected by a single type of *Wolbachia*. Infected females are compatible with uninfected and infected males, but when uninfected females mate with infected males the embryo dies (Fig. 1). Because infected females mate successfully with any male and transmit the infection to all of their offspring, the *Wolbachia* infection should spread and occur at near 100% in populations. These dynamics apply to a *Wolbachia* infection in *C. pipiens* from California[4].

To generate other forms of incompatibility among strains, different types of *Wolbachia* are needed. CI can be bidirectional when strains carry different *Wolbachia* infections. There is incompatibility when males with one type of infection mate with females carrying a different infection, which means that these strains cannot reproduce successfully (Fig. 1). When the same individual carries two different *Wolbachia* infections, complex patterns of incompatibility can occur. Males carrying both infections can show unidirectional CI not only with uninfected females, but also with females carrying only one of the infections. Moreover, strains with only one type of *Wolbachia* can show bidirectional CI. Double infections that act in this manner are found in mosquitoes such as *Aedes albopictus*[5].

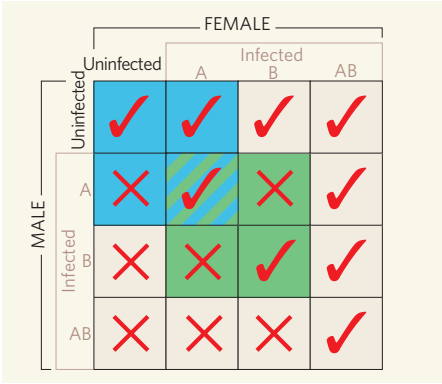

Figure 1 | Types of cytoplasmic incompatibility (CI). A and B are *Wolbachia* infections that occur singly (A, B) or together (AB) in cells. The blue area represents unidirectional CI, the green area bidirectional CI; the A × A mating is part of both unidirectional CI and bidirectional CI. Crosses indicate incompatibility, and ticks compatibility. A complex pattern of incompatibility results when strains with different *Wolbachia* status are intercrossed.

Although complex patterns of CI occur in crosses between strains of *C. pipiens*, attempts to isolate different types of *Wolbachia* from these strains have previously failed, even when using variation in genes known to distinguish *Wolbachia* infections in other insects[6]. Sinkins *et al.*[3] have now shown that *C. pipiens* strains that are bidirectionally incompatible differ in two ankyrin-encoding genes. Ankyrins are short, repeated amino-acid motifs thought to mediate interactions among proteins, as well as between proteins and the cytoskeleton, the cellular scaffolding. Genes with ankyrin sequences are particularly common in the *Wolbachia* genome relative to those of other bacteria[7], making them candidates for the expression of different patterns of CI, especially as they may control the way *Wolbachia* interacts with the cell cycle of its host.

Sinkins *et al.* found substantial genetic divergence in the two ankyrin-encoding genes of the two *C. pipiens* strains they examined; almost 10% of the nucleotides had diverged for one gene and around 3% for the other. The same genes also differed in another strain that exhibits a different incompatibility pattern. So these genes may be responsible for the varied expression of incompatibility, although the mechanism remains unknown.

Further players in events need introducing here. These are bacteriophages, phages for short, which are viruses that infect bacteria and can insert their genomes into that of their host. They enter the story because it turned out that both of the ankyrin-encoding genes are located in regions of the *Wolbachia* genome where phage chromosomes are inserted. Phages can excise from these regions and carry genes located within them to other places in the same genome or different genomes.

By using filters to separate the small phage particles from the larger *Wolbachia*, Sinkins *et al.*[3] found that the two ankyrin-encoding genes could be carried by phage particles independently of the *Wolbachia* genome. This is the first time that putative genes associated with CI expression have been located on phage particles, although it has previously been suspected[8] that phages may be involved in CI. Phage activity, along with the movement of transposable genetic elements, may help to explain the marked changes in the arrangement of ankyrin genes among *Wolbachia* genomes from related hosts[9]. Variation in one such element, known as Tr1, has also been associated with different incompatibility strains of *C. pipiens*[10].

Wolbachia have long been promoted as potential vehicles for transferring deleterious genes into mosquitoes, and other insect populations, for pest control. Among the obstacles to this are the absence of a system for genetically transforming *Wolbachia* and a way of generating new CI types capable of spreading transgenes. If CI genes are located within phage chromosomes, this should help in developing *Wolbachia* infections that can spread deleterious genes.

But there remains a danger that changes in the host's nuclear genome will decrease the efficacy of CI as a mechanism to drive transgenes into mosquito populations. Indeed, Sinkins *et al.*[3] used backcrosses of two strains of a mosquito species related to *C. pipiens* to show that nuclear genes modified the expression of incompatibility, highlighting the fact that these types of evolutionary changes are possible. So although we are now closer to understanding CI and using *Wolbachia* to introduce transgenes, host genome evolution may yet be a barrier to the aim of using *Wolbachia* for suppressing pest populations. ■

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