Heredity (2008) 101, 473–474

www.nature.com/hdy

NEWS AND COMMENTARY

Insights beyond *Wolbachia-Drosophila* interactions

Never completely trust a model: insights from cytoplasmic incompatibility beyond *Wolbachia–Drosophila* interactions

O Duron

Heredity (2008) **101,** 473–474; doi:10.1038/hdy.2008.113; published online 22 October 2008

Recent years have witnessed the revelation of inherited microorganisms as major features of arthropod biology. Among them, the α-Proteobacteria *Wolbachia* has experienced a remarkable explosion of interest, fascinating biologists by its capacity to drive the evolution of their hosts, for instance inducing postzygotic sterility, altering sex ratios or affecting the host's genome organization (Bandi *et al*., 2001; Dunning Hotopp *et al*., 2007). The lifestyle of *Wolbachia* is a true success story since these endosymbionts became among the most abundant on Earth—they are four or five times as common as any other inherited bacterium (Duron *et al*., 2008)—by manipulating the reproduction of arthropods. How this manipulation takes place is an intriguing quest for Science but very little is actually known. A key step forward was achieved by studying the behaviour of a variety of *Wolbachia* strains in the *melanogaster* group of *Drosophila* fruit flies. However, in a recent issue of Heredity, Clark *et al*. (2008) present new insights on *Wolbachia* biology through the use of understudied host model systems. Although *Drosophila* reigns supreme as a laboratory system, Clark *et al*. (2008) establish that conclusions from the *Wolbachia–Drosophila* interactions may not hold true across all *Wolbachia*–insect interactions.

The ultimate cause of the reproductive parasitism exerted by *Wolbachia* can be found at the cytoplasmic level, where there is inheritance only through the female hosts, therefore making males a 'dead end' (Bandi *et al*., 2001). In some cases, this asymmetry has selected for an increased production of daughters—but not of sons—by infected females. More commonly, *Wolbachia* exerts a form of conditional male sterility termed cytoplasmic incompatibility (CI). Through CI, *Wolbachia* hamper the reproduction of the uninfected females mated with infected males indirectly. Zygotes produced in this cross are killed in diploid species, and killed or forced into male development in haplodiploid species. This loss of uninfected female progeny confers a reproductive advantage to the infection, and can drive *Wolbachia* through natural populations to high prevalence. Although the phenotype and population biology of CI are well known, the knowledge of proximate mechanism remains desperately limited. CI is classically considered to result from two bacterial components: a *mod*—for modification—function that affects sperm, and a *resc*—for rescue—function provided by the *Wolbachia* present in the egg that restores compatibility (Werren, 1997). When the spermatozoon enters the egg, CI is expressed by the failure of paternal chromosomes to properly condense, whereas the maternal chromosomes enter normally into mitosis, leading to haploid or aneuploid conditions.

A key requirement for understanding how *Wolbachia* induce CI is to establish when and how *Wolbachia* interact with developing sperm. Studies of the interaction of diverse *Wolbachia* strains during *Drosophila* spermatogenesis have been the major focus of study to date. *Wolbachia* are found in the *Drosophila* testes but are absent from mature sperm, being removed from developing cysts with cytoplasm and most other organelles. Variation of *Wolbachia* density within cysts for different *Wolbachia* strains, and across different *Drosophila* species, correlates with the variation in CI levels between these species, suggesting that an abundance of *Wolbachia* in the testes is necessary (although not sufficient) to induce CI (Clark *et al*., 2003; Veneti *et al*., 2003). Furthermore, *Wolbachia* density in testes has been shown to decrease with male ageing in *Drosophila*, which correlates with the reduced strength of CI found in aged males (Clark *et al*., 2003). These observations form the basis of the *Wolbachia*-infected spermatocyte/spermatid hypothesis (WISSH), which argues that infected cysts represent the cellular basis of CI (Clark *et al*., 2003). According to the *Drosophila* data, *Wolbachia* infection of spermatocytes is then required for CI expression.

Clark *et al*. (2008) challenge this hypothesis using two non-*Drosophila* models: the haplodiploid wasp *Nasonia vitripennis* and the diploid beetle *Chelymorpha alternans*. Both species show high levels of CI, resulting in all-male progeny in *N. vitripennis* and the death of almost all embryos in *C. alternans*. High CI levels in both host species suggest that all spermatozoa have been successfully modified by *Wolbachia*. According to WISSH, most of the spermatocytes should be infected by *Wolbachia* to explain the high CI level observed but a very different pattern of infection is actually observed: only a few developing sperm are seen to be infected in *N. vitripennis* and none in *C. alternans*. The convergence of these observations demonstrates that *Wolbachia* do not modify the male chromosomes during late spermatogenesis *in situ*. The sperm modification by *Wolbachia* takes place either across tissue membranes from somatic infected cells or very early in the development of the host before spermatogenesis. The WISSH's scope therefore appears limited to *Wolbachia–Drosophila* interactions, and cannot be generalized across *Wolbachia*–host interactions.

Many studies that review *Wolbachia* biology have speculated on how CI is induced on the basis of *Drosophila* results. We should now raise the question of the adequacy of the other *Wolbachia–Drosophila* results. With respect to the *mod* function, CI intensity has been shown to decrease with male ageing in *Drosophila*, but no effect has been found in numerous other hosts, as documented in *N. vitripennis* by Clark *et al*., suggesting that *Drosophila* is a non-typical *Wolbachia*–host. With respect to the *resc* function, it is sometimes argued that CI embryos exhibit the same defects as in *Drosophila*, suggesting a conserved mechanism, but even this assertion must be considered carefully. Embryos blocked at a variety of developmental stages (from recently fertilized to quite well developed) are usually observed in

all incompatible cross types in *Drosophila*, whether females are uninfected or infected by a *Wolbachia* strain incompatible with the one present in males (Callaini *et al*., 1996). By contrast, CI crosses in the mosquito *Culex pipiens* produced embryos that consistently failed to develop beyond a very early stage when mothers are uninfected but, when males and females are infected by incompatible *Wolbachia* strains, a large number of embryos are blocked at later developmental stages (Duron and Weill, 2006). Maternal *Wolbachia* present in the eggs allow some morphogenesis in *C. pipiens*, notwithstanding the infection being different to that in the male, in contrast to the *Drosophila* case. Such observations highlight another level at which there is variation in *Wolbachia*–host interactions, and that study beyond *Drosophila* is essential to gain a complete analysis. There is now no doubt that it is wise to conduct more investigations on a wide range of host–*Wolbachia* associations before constructing a general model of CI. Furthermore, the relatively recent identification of the Bacteroidetes bacterium *Cardinium* as another causative agent of CI in arthropods represents a fascinating model for further comparative studies (Hunter *et al*., 2003).

Dr O Duron is at the Department of Genetics, Evolution and Environment, University College London, London NW1 2EH, UK or Institut des Sciences de l'Evolution—Montpellier, Université Montpellier 2, CC 065, 34095 Montepllier Cedex 5, France.

e-mail: oduron@univ-montp2.fr

Bandi C, Dunn AM, Hurst GDD, Rigaud T (2001). Inherited microorganisms, sex-specific virulence and reproductive parasitism. *Trends Parasitol* **17**: 88–94.

Callaini G, Riparbelli MG, Giordano R, Dallai R (1996). Mitotic defects associated with cytoplasmic incompatibility in *Drosophila simulans*. *J Invertebr Pathol* **67**: 55–64.

Clark ME, Bailey-Jourdain C, Ferree PM, England SJ, Sullivan W, Windsor DM, Werren JH (2008). *Wolbachia* modification of sperm does not always require residence within developing sperm. *Heredity* **101**: 420–428.

Clark ME, Veneti Z, Bourtzis K, Karr TL (2003). *Wolbachia* distribution and cytoplasmic incompatiblity during sperm development: the cyst as the basic cellular unit of CI expression. *Mech Dev* **120**: 185–198.

Dunning Hotopp JC, Clark ME, Oliveira DSG, Foster JM, Fischer P, Torres MC. *et al*. (2007). Widespread lateral gene transfer from intracellular bacteria to multicellular eukaryotes. *Science* **317**: 1753–1756.

Duron O, Bouchon D, Boutin S, Bellamy L, Zhou L, Engelstädter J, Hurst GDD (2008). The diversity of reproductive parasites among arthropods: *Wolbachia* do not walk alone. *BMC Biol* **6**: 27.

Duron O, Weill M (2006). *Wolbachia* infection influences the development of *Culex pipiens* embryos in incompatible crosses. *Heredity* **96**: 493–500.

Hunter MS, Perlman SJ, Kelly SE (2003). A Bacteroidetes-group bacterial symbiont induces cytoplasmic incompatibility in the parasitoid wasp *Encarsia pergandiella*. *Proc R Soc Lond B* **270**: 2185–2190.

Veneti Z, Clark ME, Zabalou S, Karr TL, Savakis C, Bourtzis K (2003). Cytoplasmic incompatiblity and sperm cyst infection in different *Drosophila–Wolbachia* association. *Genetics* **164**: 545–552.

Werren JH (1997). Biology of *Wolbachia*. *Ann Rev Entomol* **42**: 587–609.

Editor's suggested reading

Bordenstein SR, Werren JH (2007). Bidirectional incompatibility among divergent *Wolbachia* and incompatibility level differences among closely related *Wolbachia* in *Nasonia*. *Heredity* **99**: 278–287.

Gotoh T, Noda H, Ito S (2007). *Cardinium* symbionts cause cytoplasmic incompatibility in spider mites. *Heredity* **98**: 13–20.

Pool JE, Wong A, Aquadro CF (2006). Finding of male-killing *Spiroplasma* infecting *Drosophila melanogaster* in Africa implies transatlantic migration of this endosymbiont. *Heredity* **96**: 100–105.

Yusa Y (2006). Genetics of sex-ratio variation inferred from parent–offspring regressions and sib correlations in the apple snail *Pomacea canaliculata*. *Heredity* **96**: 100–105.