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Endosymbionts manipulation of the reproduction and development of spider mites

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The spider mite, *Tetranychus truncatus*, is a polyphagous agricultural pest harboring various endosymbionts, such as *Wolbachia* and *Spiroplasma*. How they interact with each other in the same host is not clear. How temperature impacts the host and their interaction with endosymbionts remains largely unknown. Furthermore, little is known about the bacterial communities of *Tetranychus truncatus* and the molecular mechanism underlying *Wolbachia* regulation of host genes. This keynote speech will review and reveal new developments and discoveries on endosymbionts from this spider mite in our laboratory.

The microbiota composition of *T. truncatus* varied across the different developmental stages. We found that Proteobacteria were the most dominant bacteria from all samples. Spider mites co-infected with *Wolbachia* and *Spiroplasma* had a significantly higher daily fecundity and juvenile survival rate than the singly infected or the uninfected ones (Yang *et al*., 2020). In addition, co-infection of *Wolbachia* and *Spiroplasma* changed the expression of many genes related to digestion, detoxification, reproduction, immunity and oxidation/reduction, conferring multiple effects on their hosts. Both *T. truncatus* infected and uninfected with *Wolbachia* and *Spiroplasma* induced similar levels of JA (Jasmonic acid) and SA (Salicylic acid) accumulation in tomato; whereas tomato plants damaged by *Wolbachia*(+) - *Spiroplasma*(+) spider mites showed lower expression levels of JA and SA responsive genes than those damaged by uninfected spider mites (Yang *et al*., 2021; Zhu *et al*., 2020a,b).

Thermal tolerance of mites was lower for mites that were singly infected with either *Wolbachia* or *Spiroplasma* than it was for co-infected or uninfected mites. Endosymbiont-mediated spider mite responses to temperature stress were complex (Zhu *et al*. 2021), involving a combination of changing endosymbiont infection patterns, altered thermoregulatory behavior, and transcription responses. Chorion protein S38-like and Rop facilitated *Wolbachia*-mediated changes in *T. truncatus* fertility. RNA interference (RNAi) of chorion protein S38-like and Rop in *Wolbachia*-uninfected *T. truncatus* decreased oviposition, which was consistent with *Wolbachia*-induced oviposition decrease. Interestingly, suppressing Rop in *Wolbachia*-infected *T. truncatus* led to increased *Wolbachia* titres in eggs; however, this did not occur after RNAi of chorion protein S38-like.

The strain *w*Ttru belongs to supergroup B. Two copies of the Type V $cif_{w\text{Ttru}}$ gene were found. The genome also contained a large number of mobile genetic elements which may contribute to sequence variation in $cif_{w\text{Ttru}}$ (Xia *et al*. 2021, 2022). Partial cytoplasmic incompatibility was induced by $cif_{w\text{Ttru}}$ in *T. truncatus*. The incomplete CI induced by $cif_{w\text{Ttru}}$ was shown to be related to $cif_{w\text{Ttru}}$ variation, especially variation in $cifA_{w\text{Ttru}}$.

In conclusion, *T. truncatus* can harbor two different endosymbionts which co-influence the host's development and reproduction in a complex way.

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Keywords: endosymbiont, spider mite, *Tetranychus truncatus*, *Wolbachia*, *Spiroplasma*, cytoplasmic incompatibility

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