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Authors: Thomas, Donald B., and León, Jesse H. De

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IS THE OLD WORLD FIG, *FICUS CARICA* L. (MORACEAE), AN ALTERNATIVE HOST FOR THE ASIAN CITRUS PSYLLID, *DIAPHORINA CITRI* (KUWAYAMA) (HOMOPTERA: PSYLLIDAE)?

DONALD B. THOMAS AND JESSE H. DE LEÓN
USDA-ARS Kika de la Garza Subtropical Agriculture Research Center, 2413 East Bus Highway 83, Weslaco TX 78596

The Asian citrus psyllid, *Diaphorina citri* (Kuwayama) is the vector of Huanglongbing, the most serious disease of citrus worldwide (Bové 2006). Although most psyllids, including *D. citri*, confine breeding to a narrow range of usually related host plants, the adults will feed and obtain nutrition from a wider array of plants (Hodkinson 1974). The Asian citrus psyllid prefers *Citrus* spp. and all breeding host plants are in the family Rutaceae (reviewed by Halbert & Manjunath 2004). Feeding could spread the disease agent to non-citrus plants that might then serve as reservoirs of the disease in nature. A mitigating factor is that only the immature stages of the psyllid can effectively acquire and host the bacteria (Pelz-Stelinski et al. 2010), and thus, any plant species that serve as breeding hosts are of primary concern in that regard.

On 23 Jul 2010 while surveying dooryard citrus trees for infestations of *D. citri*, we noted that adults of this insect were abundant on a fig tree, *Ficus carica* (L.) adjacent (~2m) to a heavily infested key lime tree, *Citrus limon* (L.) (Fig. 1). On further inspection we noted immature psyllids, including fifth instars, on the new flush of this fig tree. A second fig tree on the same property was not infested, possibly because it was not in flush. Some of the nymphs were brought to the laboratory where we confirmed their identity as *D. citri*. There are at least 7 genera of psyllids, most in the tribe Homotomini, known to infest *Ficus* spp. in the Old World with a record of one species, *Homotoma ficus* (L.) infesting figs in California in 1969. Nymphs of the Asian citrus psyllid can be distinguished from all of these by its 3 - segmented antenna, narrow lateral thoracic sclerites, and a se-



Fig. 1. Fig tree next to an infested lime tree in a dooryard in Weslaco, TX found to be infested with Asian Citrus psyllid in the summer of 2010.

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ries of lanceolate setae on the margin of the abdomen (Fig. 2). Although The nymphs on the fig tree keyed out to *D. citri* (White & Hodkinson 1985), and were clearly not one of the known fig psyllids. Yet, even though they were identical in all respects to known nymphs of Asian citrus psyllid, and adult Asian citrus psyllids were abundant on the tree, there remained a possibility that the nymphs could be of another, perhaps native psyllid species, for which the nymphal stages are unknown. We therefore subjected 7 specimens of the nymphs from the fig to DNA analysis.

DNA was isolated by a rapid crude DNA extraction procedure as described in previous works (de León et al. 2006; de León & Morgan 2007). The mitochondrial cytochrome oxidase subunit I gene (COI) from 11 species of Psyllidae were sequenced, aligned, and a *Diaphorina citri*-specific molecular diagnostic marker was developed that generated a 183-bp COI DNA fragment. The sequences of the markers were as follows: ACP-183-F (forward: CAT-ACG-AGC-AAA-TTT-ATA-TAA-C) and ACP-183-R (reverse: GTA-TAA-GAT-TGG-GTC-TCC-A) with the current assay conditions Tm 62 °C; 1.4 mM $MgCl_2$; 10X ThermoPol Buffer (New England BioLabs, Beverly, Massachusetts); and 30 cycles (PTC-200 Peltier Thermal Cycler, Bio-Rad Laboratories, Hercules, California). The species of native Psyllidae collected locally in Hidalgo County, Texas were: *Aphalaroida pithecolobia* (Crawford), *Bactericera cockerelli* (Sulc), *Cacopsylla alba* (Crawford), *Heteropsylla texana* (Crawford), *Heteropsylla mimosae* (Crawford), *Pachypsylla celtidismamma* (Riley), *Pachypsylla celtidisgemma* (Riley), *Leuronota maculata* (Crawford), *Trioza diospyri* (Ashmead), and *Tetragonocephala flava* (Crawford). Representative specimens of each species were slide mounted and are maintained as vouchers at the USDA-ARS lab in Weslaco, Texas. The results demonstrated that the molecular marker was highly specific for *D. citri* as cross-reactivity was not observed with any of the native species (Fig. 3).

On subsequent visits in Jul and Aug the fig tree continued to be infested with immature *D. citri.* However, the latter months of 2010 beginning with Sep, were exceptionally dry in south Texas and the trees (fig and lime) stopped producing flush and nymphal psyllids were no longer observed on them. However on 13 Jul 2011, following heavy rains at the end of Jun, the same tree was found to be reinfested with a small number of early instars.

Although many of the nymphs observed in 2010 were late instars exhibiting wing pads, and there were numerous adults of *D. citri* present, we could not be certain that any of the adults found on the fig had originated from nymphs that completed development on this same tree, as opposed to the nearby lime tree. In order to determine whether *D. citri* nymphs can complete development to adults on fig, we therefore initiated a greenhouse experiment on 14 Sep wherein 2 potted fig trees (cv. 'Alma') were enclosed in a tent with an infested lime tree. Two tents, (dimensions 2 × 2 × 3m) were used with the fig trees at opposite corners, not touching. There was no intervention in this free-choice experiment. To keep disturbance of the insects to a minimum we did not remove any adults or nymphs from any of the trees except for vouchers as noted. For maintenance, all trees were watered and fertilized under warm temperatures (25 °C) and high relative humidity (75-80%) to induce and maintain flush. One of the

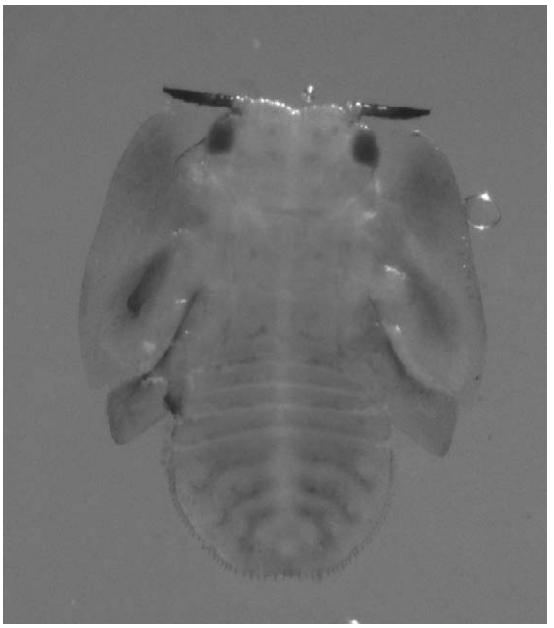


Fig. 2. Dorsal view of nymph developed on fig tree.

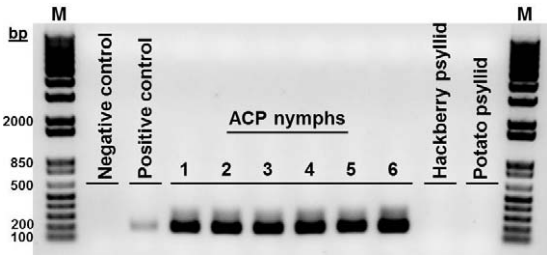


Fig. 3. Screening of ACP nymphs collected on fig trees with a *Diaphorina citri*-specific COI molecular diagnostic marker. The expected size of the DNA band is 183 base pairs. The negative control is a reaction with sterile water instead of template DNA. The positive control is an adult ACP. Lanes 1-6 are individual ACP nymphs. For comparison, the hackberry psyllid (*Leuronota maculata*) and potato psyllid (*Bactericera cockerelli*) are included.

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'Alma' fig trees flushed heavily and on 1 Nov we observed on it the first *D. citri* nymphs. On 29 Nov we removed 3 fifth instar nymphal exuviae from this fig each with an eclosion slit in the dorsum. These were mounted on slides as vouchers.

The greenhouse experiment continued with many adults observed on the fig trees and on the sides of the tent. But during the next several months only early instars were seen on the apical meristems of the figs. On 28 Apr we noted that a colony of middle instars had established on a stem of one tented fig tree accompanied by copious amounts of waxy exudate. One late instar nymph was removed and slide-mounted as a voucher at that time. As it was 1.6 mm in length it was judged to be a fifth instar based on size and other diagnostic criteria (EPPO 2005). The EPPO standard gives the dimensions of a fifth instar as 1.45-1.58 mm length. On 4 May we noted teneral adults on the fig. We removed 12 eclosed fifth-instar exuviae and slide-mounted them as vouchers. All exuviae were 2.0 mm in length.

For a field experiment 3 potted fig trees (cv. 'Black Mission') were placed in the USDA-ARS experimental citrus grove where insecticide use is eschewed and *D. citri* populations were active. Beginning on 5 Oct 2011, each potted fig was intercalated among grapefruit trees on the outer row of the grove. On 3 Jan 2011, one of these potted figs was found to be infested with late instars of *D. citri* of which 1 fifth instar was taken and mounted as a voucher. However, the onset of cold temperatures prevented further development. Drought conditions then prevailed over the first half of 2011 and populations of *D. citri* on *Citrus* collapsed, preventing further field experiments.

Outside of the family Rutaceae, *D. citri* has been reported breeding on jackfruit, *Artocarpus heterophylus* Lamerek, in the same plant family as the fig, Moraceae (Shivankar et al. 2000). However, in follow-up experiments Peña et al. (2006) were unable to substantiate the status of jackfruit as a breeding host. But, more recently, Fan et al. (2011) discovered an instance of *D. citri* breeding on a legume tree, *Pithecellobium lucidum* Benth., in China, an observation that was subsequently confirmed with greenhouse experiments. These reports, along with the present observations on fig trees, suggest that while *D. citri* breeds mainly on a narrow range of host plants within the family Rutaceae, some non-preferred plants outside of that family may be acceptable alternatives.

Summary

Adults and immatures of the Asian citrus psyllid were found infesting a dooryard fig tree, *Ficus carica* (L). When potted fig trees were placed in an infested citrus grove, one of the fig trees (cv 'Mission black') also became infested. In a greenhouse experiment wherein fig trees were exposed to infested lime trees, one of the fig trees (cv 'Alma') became infested. In one generation at least 3 adults eclosed from an infestation and in a second generation at least 12 adults eclosed from an infestation on the fig. Although the usual host plants for the Asian citrus psyllid are in the family Rutaceae, the old world fig may be an adventitious breeding host.

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