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## SUITABILITY OF 'CLEOPATRA' MANDARIN AS A HOST PLANT FOR DIAPHORINA CITRI (HEMIPTERA: PSYLLIDAE)

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The Asian citrus psyllid, Diaphorina citri (Kuwayama) is currently the most significant insect pest of Florida citrus as the vector of *Candidatus* Liberibacter asiaticus, the phloem-limited bacterium associated with citrus greening disease, also known as huanglongbing (HLB). The biology of D. citri has been studied by Catling (1970); Pande (1971); Capoor et al. (1974); Yang (1989); Liu & Tsai (2000); Nakata (2006); and Nava et al. (2007). In these studies, the host range of D. citri is reported to comprise numerous species within the Rutaceae, including citrus and close citrus relatives. While there are many field observations regarding host-plant associations of D. citri (Halbert & Manjunath 2004), there are only 2 laboratory studies (Tsai & Liu 2000; Nava et al. 2007) in which factors including fecundity, adult longevity, sex ratio, and survival and development period of immature stages have been compared on different host plants. In both studies, significant differences were reported for adult fecundity and survival of nymphs when D. citri was reared on different host plants. However, despite these differall citrus species evaluated considered to be suitable host plants for *D. citri*.

During the course of our studies on the biology of *D. citri*, we observed a lack of successful development from egg to adult by *D. citri* on 'Cleopatra' mandarin (*Citrus reshni* Hort. ex Tan.) growing in both the field and greenhouse. Based on these observations, we compared the suitability of 'Cleopatra' mandarin versus 'sour orange' (*Citrus aurantium*) as host plants for *D. citri* by assessing oviposition rate, survival, and developmental period on each host.

Adult D. citri used in experiments were obtained from a colony reared on Murraya koenigii in a greenhouse at the Citrus Research and Education Center, Lake Alfred, Florida. Colony plants were maintained in polyester/nylon netting cages  $(61 \times 61 \times 91.5 \text{ cm}^3)$  (Bioquip, Rancho Dominguez, CA). Test plants of 'Cleopatra' mandarin and 'sour orange' (30-50 cm in height) were grown from seed in the greenhouse, pruned, and fertilized prior to use in experiments to ensure new leaf growth for psyllid oviposition. Clear acetate cylinders (34 cm height × 6 cm diameter) were used to cage adult psyllids on new leaf growth. One end of each cylinder was covered with extra fine muslin and the opposite end, in which the plant was inserted, was sealed with a sponge to prevent psyllids from escaping. To ensure proper ventilation, a small opening  $(5 \times 5 \text{ cm})$  covered with extra fine muslin was made in the side of each cage, 5 cm

from the top of the cage. All experiments were conducted under greenhouse conditions with an average temperature of  $28.8 \pm 6.79$ °C and  $70.9 \pm 27.31\%$  relative humidity.

Oviposition rate was assessed over a period of 15 d by caging 1 female *D. citri* on new leaf growth of 10 'Cleopatra' mandarin and 'sour orange' seedlings. Two male *D. citri* were confined with each female to allow multiple matings, which has been demonstrated to maximize reproductive output (Wenninger & Hall 2008). Female psyllids were moved every 3 d to new plants to provide suitable oviposition sites. Eggs laid on each plant were counted and remained caged until adult emergence. Survival from egg to adult was calculated as the number of the adults emerged divided by the number of eggs laid per plant. Sex ratio of the emerged adults was determined.

To compare the duration of the egg stage and each of the instars between the 2 host plants, female adult psyllids from the greenhouse colony were caged on new leaf growth of both citrus species under greenhouse conditions as described above. After oviposition, the adults were removed from the cages and eggs were counted with use of a stereomicroscope. The eggs were checked daily and when a first instar emerged, it was transferred with a fine paintbrush and caged on young leaf tissue of a seedling of the same species. Nymphs were checked daily for ecdysis until they reached the adult stage to determine developmental rate for each instar. All data collected were analyzed and compared by 2-sample *t*-test statistics (Analytical Software 2008).

During the 15-d caging period, significantly fewer eggs were laid on "Cleopatra' mandarin compared to 'sour orange' (2-sample t-test, t =6.34, df = 58, P < 0.0001). The mean number of eggs laid by female D. citri was  $24.1 \pm 2.7$  and 80.2± 8.5 on 'Cleopatra' mandarin and 'sour orange,' respectively. Survival to the adult stage on 'Cleopatra' mandarin was 4.38 ± 1.46%, and significantly lower compared to survival on 'sour orange'  $(62.21 \pm 2.86\%)$  (t = 18.03, df = 58, P < 0.0001). Survival on 'sour orange' was lower compared to results reported in Tsai & Liu (2000), who found a survival rate of 71% on 'sour orange'. Differences may be due to slight differences in environmental conditions under which experiments were conducted. Host plant did not have any effect on the sex ratio  $(0.50 \pm 0.04 \text{ on sour orange}, 0.50 \pm 0.09)$ on Cleopatra) (t = 0.16, df = 35, P = 0.8766).

The development rate of psyllid nymphs was significantly longer for 'Cleopatra' mandarin com-

TABLE 1. MEAN  $\pm$  SE DEVELOPMENTAL PERIODS (DAYS) OF IMMATURE STAGES OF D. CITRI ON 'SOUR ORANGE' AND 'CLEOPATRA' MANDARIN UNDER GREENHOUSE CONDITIONS.

Host	Egg	1st instar	2nd instar	3rd instar	4th instar	5th instar	Sum of all instars	Egg to adult
Sour orange Cleopatra F	$4.47 \pm 0.09 \mathrm{a}$ $4.56 \pm 0.11 \mathrm{a}$ 0.34	2.33 ± 0.10 a 2.61 ± 0.11 a 3.08	1.73 ± 0.13 a 1.78 ± 0.16 a 0.04	1.87 ± 0.16 a 1.94 ± 0.21 a 0.08	2.50 ± 0.12 a 2.67 ± 0.17 a 0.61	5.57 ± 0.14 a 6.11 ± 0.15 b 6.10	$14.00 \pm 0.24$ a $15.11 \pm 0.45$ b $5.58$	18.47 ± 0.14 a 19.67 ± 0.49 b 5.25
aı = 40 P	0.5608	0.0857	0.8339	0.7766	0.4382	0.0173	0.0224	0.0266

Means followed by the same letter within columns are not significantly different (P < 0.05).

pared to 'sour orange' with significant differences present in the duration of the 5th instar, the sum of all instars and from egg to adult (Table 1). Psyllid developmental rate on 'sour orange' was longer in duration than previously reported by Tsai & Liu (2000), but differences in environmental conditions for the 2 studies could explain the differences

Our results demonstrate that the fitness of the Asian citrus psyllid is greatly reduced on 'Cleopatra' mandarin as a direct result of high nymphal mortality due to some unknown cause. Further studies are needed to determine the factors responsible for reduced psyllid survival and whether this can be utilized in future citrus IPM programs as a tool for psyllid management. The authors thank Harry Anderson for providing capable technical assistance and Drs. Kirsten Pelz – Stelinski and Rajinder Mann (University of Florida) for improving a previous version of this manuscript. Funding for this research was provided by the Florida Citrus Production Research Advisory Council.

## SUMMARY

In this study we compared suitability of 'Cleopatra' mandarin and 'sour orange' as host plants for *D. citri*. Oviposition rate and survival of *D. citri* were significantly reduced on 'Cleopatra' mandarin. There was no effect of host plant on development rate of each immature stage or sex ratio, although development rate from egg to adult was significantly longer on 'Cleopatra'. The results of this study support our previous observations that 'Cleopatra' mandarin negatively affects fitness of *D. citri*. Additional studies are ongoing to determine the factors responsible for reduction in the survival of psyllid nymphs when reared on 'Cleopatra' mandarin.

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