FIELD RELEASE IN FLORIDA OF *DIAPHORENCYRTUS ALIGARHENSIS* (HYMENOPTERA: ENCYRTIDAE), AN ENDOPARASITOID OF *DIAPHORINA CITRI* (HOMOPTERA: PSYLLIDAE), FROM MAINLAND CHINA

Eric A. Rohrig[1,2], David G. Hall[3], Jawwad A. Qureshi[4] and Philip A. Stansly[4]
[1]Entomology and Nematology Department, University of Florida, Gainesville, FL 32611

[2]USDA-ARS Center for Medical, Agricultural and Veterinary Entomology, Gainesville, FL 32608

[3]USDA-ARS Horticultural Research Laboratory, Fort Pierce, FL 34945

[4]University of Florida-IFAS, SWFREC, Immokalee FL 34142

The Asian citrus psyllid, *Diaphorina citri* Kuyawama (Hemiptera: Psyllidae), is a serious pest of citrus that vectors the phloem-limited nonculturable bacterium *Candidatus* Liberibacter asiaticus Jagoueix, Bové and Garnier (Jagoueix et al. 1994). The resulting disease is commonly referred to as citrus greening or huanglongbing (HLB) (Bové 2006). The psyllid was first reported from Florida in 1998 and citrus greening in 2005 (Halbert 1998, 2005).

Two species specific parasitic wasps, *Tamarixia radiata* (Waterston) (Hymenoptera: Eulophidae) and *Diaphorencyrtus aligarhensis* (Shafee, Alam and Argarwal) (Hymenoptera: Encyrtidae), were imported into Florida from Taiwan and Vietnam and released beginning in 1999 as biological control agents for *D. citri* (Hoy & Nguyen 1998). *Tamarixia radiata* has established in Florida (Qureshi et al. 2009), while *D. aligarhensis* has not.

An additional population of *D. aligarhensis* was collected in Guangdong province, China and sent to Gainesville, Florida in 2006. A colony was initiated in the Maximum Security Room of the Florida Biological Control Laboratory, Florida Department of Agriculture and Consumer Services, Division of Plant Industry. Parasitoids were held in quarantine through 20 generations before release permitting was complete and mass rearing began. Sixty-seven releases were made at 10 sites in 6 Florida counties (Alachua, Collier, DeSoto, Hendry, Orange and Polk) between 8/2007 and 10/2009, deploying a total of approximately 11,100 wasps (Table 1). Sites included commercial and private citrus groves as well as *Murraya paniculata* hedges. Wasps were either tapped out of holding tubes directly onto emerging plant shoots infested with immature psyllids or were released into cages consisting of fine mesh bags tied around new shoots infested with nymphs. Shoots were carefully inspected and predators removed prior to tying cages closed. Mesh size allowed parasitoid wasps to move freely in or out of the cage while preventing most predaceous insects from entering.

Released wasps successfully parasitized nymphs in the field and second generation wasps emerged. Evidence of this includes laboratory emergence of adult wasps from field clippings, *D. aligarhensis* parasitized mummies found on shoots covered with mesh cages, and several adult wasps sighted foraging on *D. citri* infested *M. paniculata* plants (Table 1).

All citrus groves used for wasp release, including certified organic groves, used pesticide sprays during the release period. The use of insecticides targeting *D. citri* likely reduced both psyllid and natural enemy populations (Qureshi & Stansly 2007). *Murraya paniculata* release sites were not subjected to any form of insect control, although occasional plant trimming by the property owners removed much of the new soft growth that *D. citri* uses for feeding and oviposition, consequently leaving few immature hosts for *D. aligarhensis* reproduction.

Adult psyllids were present at all locations throughout the release period although their incidence appeared to decline over the >2 year release period, likely from increased insecticide use to reduce the spread of citrus greening disease. During many trips to various locations for parasitoid release there was little to no host plant flush or immature psyllids present, particularly during late summer and fall.

Tamarixia radiata wasps were present at all release sites utilized in this study. Through the combined effect of host feeding and parasitism, a single female *T. radiata* can kill nearly twice as many psyllids in her lifetime as can a female *D. aligarhensis* (500 vs. 280, respectively) (Chien 1995). Additionally, *T. radiata* has a shorter generation time and a higher reproductive rate (Skelley & Hoy 2004). Furthermore, any hosts parasitized by *T. radiata* before or within 5 d following parasitism by *D. aligarhensis* will result in *T. radiata* offspring (E. R. unpublished data). Thus, competition with T. radiata could present another impediment to establishment of *D. aligarhensis*.

Coccinellid predators were seen at all release sites. Generalist predators can induce high levels of psyllid mortality (Qureshi & Stansly 2009). *Curinus coeruleus* Mulsant, *Cycloneda sanguinea* (L.) and *Harmonia axyridis* Pallas readily consumed *D. citri* nymphs parasitized by *D. aligarhensis* (E. R. unpublished data) causing wasp mortality.

TABLE 1. SUMMARY OF *DIAPHORENCYRTUS ALIGARHENSIS* RELEASED AND OF ADULT *DIAPHORINA CITRI* (ACP), AND *DIAPHORENCYRTUS ALIGARHENSIS* (*D.A.*) AND *TAMARIXIA RADIATA* (*T.R.*) RECOVERED FROM FIELD PLANT CLIPPINGS.

Release			Recovery		
Site, City, County	Date mo/d/yr	D. a.	Adult ACP	D. a.	T. r.
Citrus sp.					
Commercial grove	08/21/07	300	—	—	—
Immokalee	09/11/07	500	33	0	1
Collier County	10/08/07	300	46	1 (+3)	0
	11/06/07	300	29	0	2
	12/04/07	350	12	0 (+3)	0
	05/28/08	150	21	0	3
	6/17/08	150	4	0	0
	07/11/08	300	33	0	0
	07/31/08	400	25	0	2
	03/23/08	250	—	—	—
	04/16/08	200	—	—	—
Research grove	08/21/07	200	37	0	0
Immokalee	10/08/07	200	22	0 (+3)	1
Collier County	11/06/07	100	17	1 (+2)	2
	12/04/07	150	12	0	1
	05/28/08	250	12	0	0
	06/17/08	150	7	0	1
	03/23/09	250	—	—	—
	04/16/09	200	2	0	0
Dooryard grove	04/30/08	50	—	—	—
Eustice	06/11/08	25	0	0	0
Collier County	06/26/08	25	12	0	1
	07/08/08	150	7	0	0
	07/17/08	75	4	0 (+4)	0
	04/30/09	200	—	—	—
	05/15/09	100	—	—	—
	06/05/09	200	2	0	1
	07/21/09	100	1	0	0
	09/01/09	100	0	0	0
	09/21/09	100	—	—	—
Organic *Citrus* sp.					
Commercial grove	05/10/09	150	32	0	2
Melrose	05/17/09	300	28	0	0
Alachua County	05/30/09	150	8	0 (+4)	4
	06/10/09	200	11	0	2
	06/20/09	200	16	0	3
	06/25/09	200	21	0 (+2)	0
	07/12/09	300	22	0	1
	07/21/09	200	17	0 (+6)	2
	08/03/09	250	12	0	2
	08/28/09	200	—	—	—
	09/18/09	200	9	0	3
	10/02/09	200	11	0	1
	10/09/09	200	7	0	2
	10/20/09	300	5	0	3
Commercial grove	03/23/09	250	12	0	2
Arcadia	04/16/09	300	9	0	1
DeSoto County					
Commercial grove	03/23/09	250	12	0	0
Lake Wales	04/16/09	300	17	0	3
Polk County					
Murraya paniculata					
Private dooryard	04/30/08	50	38	0	0

+ indicates number of parasitized mummies (with adult wasp emergence holes) observed in exclusion cages remaining on tree shoots from the previous release.

‡ indicates the number of adult wasps foraging on *Diaphorina citri* infested plants.

TABLE 1. (CONTINUED) SUMMARY OF *DIAPHORENCYRTUS ALIGARHENSIS* RELEASED AND OF ADULT *DIAPHORINA CITRI* (ACP), AND *DIAPHORENCYRTUS ALIGARHENSIS* (*D.A.*) AND *TAMARIXIA RADIATA* (*T.R.*) RECOVERED FROM FIELD PLANT CLIPPINGS.

Release			Recovery		
Site, City, County	Date mo/d/yr	D. a.	Adult ACP	D. a.	T. r.
Apopka	05/21/08	100	31	3 (‡3)	1
Orange County	06/11/08	50	27	9 (‡2)	0
	06/26/08	50	11	0	3
	09/08/08	30	12	0	2
	04/30/09	50	8	0	4
	05/15/09	50	—	—	—
	06/05/09	100	7	0	0
	06/25/09	50	—	—	—
	07/12/09	25	—	—	—
	09/01/09	50	8	0	1
	09/21/09	50	6	0	0
Chain restaurant	05/29/08	100	17	0	1
Marco Island	06/18/08	75	8	0	2
Collier County	03/24/09	50	—	—	—
Government office	06/17/08	50	0	0	0
Clewiston	07/11/08	50	2	0	1
Hendry County					
Private office	06/17/08	50	1	0	3
Clewiston	07/11/08	50	0	0	0
Hendry County					

+ indicates number of parasitized mummies (with adult wasp emergence holes) observed in exclusion cages remaining on tree shoots from the previous release.

‡ indicates the number of adult wasps foraging on *Diaphorina citri* infested plants.

SUMMARY

Despite the release of thousands of *D. aligarhensis* in 2000-2002 (Tawain population) (Skelley & Hoy 2004) and over 11,000 in 2007-2009 (mainland China population), in numerous counties throughout the citrus growing regions of Florida, *D. aligarhenisis* appears not to have established to date. Factors contributing to this apparent failure include intense pesticide use to control *D. citri*, low and variable populations of immature psyllids, competition with *T. radiata*, and predation of parasitized hosts by generalist predators. Nevertheless, the 2 parasitoids are known to co-exist in many parts of Asia, and further efforts to establish *D. aligarhensis* in Florida are still warranted.

ACKNOWLEDGMENTS

Funding for this work was provided by the Florida Citrus Production Research Advisory Committee.

REFERENCES CITED

BOVÉ, J. M. 2006. Huanglongbing: A destructive, newly-emerging, century-old disease of citrus. J. Plant Pathol. 88(1): 7-37.

CHIEN, C. C. 1995. The role of parasitoids in the pest management of citrus psyllid, pp. 245-261 *In* Proc. Symp. Research and Development of Citrus in Taiwan, Taichung, Taiwan.

HALBERT, S. E. 1998. Entomology section. Tri-ology. http://www.doacs.state.fl.us/pi/enpp/triology/archive/98-may-jun.htm (14 VI 10).

HALBERT, S. E. 2005. Pest Alert: Citrus Greening/Huanglongbing. Florida Dept Agric. Consumer Serv., Division of Plant Industry. http://www.doacs.state.fl.us/pi/chrp/greening/citrus greeningalert.html.

HOY, M. A., AND NGUYEN, R. 1998. Citrus psylla: here in Florida - an action plan. Pest Alert. http://pestalert.ifas.ufl.edu/hoy-0615.htm (June 1998).

JAGOUEIX, S., BOVÉ, J. M., AND GARNIER, M. 1994. The phloem-limited bacterium of greening disease of citrus is a member of the α subdivision of the proteobacteria. Int. J. Syst. Bacteriol. 44: 379-386.

QURESHI, J. A., AND STANSLY, P. A. 2007. Integrated approaches for managing the Asian citrus psyllid *Diaphorina citri* (Homoptera: Psyllidae) in Florida. Proc. Florida State Hort. Soc. 120: 110-115.

QURESHI, J. A., AND STANSLY, P. A. 2009. Exclusion techniques reveal significant biotic mortality suffered by Asian citrus psyllid *Diaphorina citri* (Hemiptera: Psyllidae) populations in Florida citrus. Biol. Control 50(2): 129-136.

QURESHI, J. A., ROGERS, M. E., HALL, D. G., AND STANSLY, P. A. 2009. Incidence of invasive *Diaphorina citri* (Hemiptera: Psyllidae) and its introduced parasitoid *Tamarixia radiata* (Hymenoptera: Eulophidae) in Florida citrus. J. Econ. Entomol. 102: 247-256.

SKELLEY, L. H., AND HOY, M. A. 2004. A synchronous rearing method for the Asian citrus psyllid and its parasitoids in quarantine. Biol. Control 29(1): 14-23.