Preparedness in biological control of priority biosecurity threats

Asian citrus psyllid, *Diaphorina citri* (Kuwayama)

Introduction

The Asia Citrus Psyllid, *Diaphorina citri* (Hemiptera: Psyllidae) is native to Asia. It has already invaded several countries in Asia, Africa and America. It is a quarantine pest for Europe. Climate conditions and abundance of host plants indicate that it could be established in many European Countries.There is a broad host range in the Rutaceae family but *D. citri* develops best on *Citrus* spp. and *Murraya* spp.

It is a pest of great importance of citrus because it is a vector of the devastating disease, citrus greening, known as Huanglongbing (HLB) caused by the bacterium *Candidatus* Liberibacter *asiaticus.*

Apart from the spread of the HLB, the pest can cause serious damage to citrus trees. Nymphal feeding can eliminate young leaves and even whole young shoots. Additionally, nymphal feeding can prevent new buds to emerge.

History of classical biological control against *D. citri*

Studies have been conducted about the native natural enemies of Asian Citrus Psyllid that occur in citrus orchards and could contribute to the biological control of the psyllid in introduced areas. Among the natural enemies that have been found to be associated with *D. citri*, there are two parasitoid species that have been used in Classical Biological control Programs around the world and they seem to be the most promising Biological Control Agents.

1) Tamarixia radiata Waterston (Hymenoptera, Eulophidae), it is native in South East Asia. It has already been used in Classical Biological Control in many countries around the world where *D. citri* has invaded. It is a solitary, ectoparasitic parasitoid. It is probably monophagous. Until now there no other reports that it would parasitize any other psyllid species. It parasitizes the 4th and 5th instar of *D. citri.*

It has been introduced in Reunion Island, Guadeloupe Mauritius, Florida & California U.S.A, Taiwan, Saudi Arabia, East Java, Indonesia, the Philippines. Different levels of parasitism have been observed, depending on the country, the number of the releases and the other measures that are being applied in each country. In Taiwan, Guadeloupe, Reunion Island, and California T. *radiata* had a satisfying natural distribution and establishment due to the absence of hyperparasitoids. In California, a few years after the releases, it was recovered in 10 Km away of the release sites in i.e. 87% of the releasing sites. In Taiwan after 2 years from the releases, it had a good dispersal but different percentage of parasitism ranging from 0 – 80% depending on the site. Additionally, In Reunion Island and Guadalupe, it took two and one year respectively until a great reduction of pest population took place. In Florida, the rate of parasitism varied during the seasons (a greater percentage was recorded during fall comparing to spring). In general, among the factors that are considered important for establishing of *T. radiata* are hyperparasitism, the use of insecticides, the time of the year, and the intraguild interactions with other natural enemies.

Biological control in Reunion, Parasitoids of *T.radiata* were collected from India and Imported in Reunion island for propagation in the laboratory. Releases took place from May to December 1978, with 4.600 parasitoids released in total. It took two years to reduce the population of *D. citri*. Studies that took place a few years after the releases indicated that the pest was almost absent from the island.

Biological control in Taiwan. During 1983-1986 adults of *T.radiata* were imported in Taiwan, propagated and then released in citrus orchards. First releases took place in 1984. Several releases occurred, with a total number 0f 2.100 parasitoids. In 1986 the first recovery took place.

Then, initially parasitoids recovered were used for further mass production and releases. Within 1-5 months (depending the release site) after the releases, the first mummies of *D. citri* appeared in citrus orchards, and the parasitoid was reported in different locations on the island. During1984-1987 a total number of 21.164 parasitoids was released. In certain areas, 80-100% parasitism was reported whereas in others, much less occurrence of the parasitoid was reported (0-70%). *T. radiata* was dominant over the native parasitoid species *Diaphorencytrus aligarensis*. The percentage of hyperparasitisation of *T. radiata* was 1%, comparing to the percentage of hyperparasitisation of *D. aligarensis* which was much higher.

Biological control in Guadalupe

In Guadalupe *T. radiata* also established and distributed well. The parasitoid was imported from Reunion in 1999, reared in the laboratory and released. About 1000 *T. radiata* were released. A short time after the releases, the parasitoid was present in almost every citrus orchard and caused great reduction of the pest. It was also found in region where it was not released, which indicates that it could spread naturally.

California In California, a Classical Biological Control Programme was developed during 2010-2012, where the parasitoid was collected from its native region in Punjab, Pakistan, imported and finally released in infested areas. The establishment of the parasitoid did take place but it was considered weak (20%) the first two years. A few years later (from 2015 until 2018), field monitoring indicated successful establishment (establishment in 90% of the release areas with a reduction of *D. citri* of 75% in some cases, depending on the pest-parasitoid densities).

Florida *T. radiata* was imported in 1999-2001. *T. radiata* established and distributed but depending on the season, the rate of parasitism vary (20% during spring and 39-56% during fall). A few surveys indicate that due to jntraguild predation, the parasitoid was not the main factor for suppressing *D. citri.*

2) Diaphorencytrus aligarhensis (Shafee, Alam & Agarwal) (Hymenoptera, Encyrtidae). It is native in East Asia. The females are parasitizing the 2nd, 3rd& 4th instar of the psyllid. It is a solitary endoparasitic parasitoid. It is also highly specific to *D. citri.* It has been imported, reared and released in several areas through Classical Biological Control Program such as Reunion Island, Taiwan and Florida USA. It has not been successfully established in Florida. It has established in Taiwan and Reunion Island. In Florida, the parasitoid was not established mostly due to the predominance of *T. radiata.*

Most promising natural enemies

T. radiata seems to be more promising biological control agent than *D. aligarhensis* in classical biological control. In the countries where the two species have been imported and released (together or separately), *T. radiata* was successfully established in most of them, and achieved reduction of *D. citri*.

The greater establishment of *T. radiata* occurred due to the absence of hyperparasitism. The absence of hyperparasitism (either native or accidentally imported and released hyperparasitoids) is one of the most important factors for the successful establishment for these two species. In Taiwan, where *D. aligarhensis* is native species, the percentage of hyperparasitism for *D. aligarhensis* was about 40%, whereas, the hyperparasitism of *T. radiata* (imported species for Taiwan) was less than 1%. In Philippines, a great percentage of hyperparasitism was reported for both species. Furthermore, there are more hyperparasitiod species in East Asia that are known to attack *D. aligarhensis.*

As far as the biological traits, a female of *T. radiata* can kill up to 500 numphs of *D. citri* both by host feeding and parasitism (20% and 80% mortality respectively). The female of *D. aligarhensis*

parasitizes fewer hosts per unit time than *T. radiate* (280 of *D. citri* per female). It seems that interspecific competition occurs between the two species. Further, findings from California and Florida indicate that *T. radiata* is dominant to the *D. aligarhensis*.

Other natural enemies

Apart from the two parasitoids that have been applied in BCP, which are considered the most promising biocontrol agents, there are a few predators species that have been found on citrus fields preying on the pest.

Olla v-nigrum Muslant (Coleoptera, Coccinelidae). Adults and larvae of the coccinellid predator were found on citrus preying on the psyllid in Florida when it invaded the region. Additionally, it has a strong numerical response (both in field and in laboratory experiments). In Florida, wherever the pest exists, the population of the predator has increased, indicating that it could be an important factor suppressing the population of the pest.

Harmonia haxyridis Pallas, and *Curinus coeruleus* Mulsant (Coleoptera, Coccinelidae) are two coccinellid species that have been found in citrus orchards in Florida preying on the Asian citrus psyllid. In laboratory studies, it has been found that is a good diet for these coccinellid predators.

Among other BCA, there are a few fungal entomopathogens which have caused mortality of the pest under specific environmental conditions. Laboratory and field studies indicated that, *Isaria fumosorosea* (Hypocreales: Cordycipitaceae),*Hirsutella citriformis*(Ascomycota: Cordycipitaceae), *Cladosporium sp. nr. oxysporum* Berk & M.A. Curtis and other species could be applied in the citrus orchards for suppressing the pest.

References

1. Chen, X., & Stansly, P. A. (2014). *Biology of Tamarixia radiata (Hymenoptera: Eulophidae), Parasitoid of the Citrus Greening Disease Vector Diaphorina citri(Hemiptera: Psylloidea): A Mini Review. Florida Entomologist, 97(4), 1404–1413.*

2. Chong, J.-H., Roda, A. L., & Mannion, C. M. (2010). *Density and Natural Enemies of the Asian Citrus Psyllid, Diaphorina citri (Hemiptera: Psyllidae), in the Residential Landscape of Southern Florida. Journal of Agricultural and Urban Entomology, 27(1), 33–49.* doi:10.3954/11-05.1

3. Elizabeth E. Grafton-Cardwell, E., E., Stelinski, L., L., and Stansly, P., A. 2013. Biology and Management of Asian Citrus Psyllid, Vector of the Huanglongbing Pathogens. Annu. Rev. Entomol..58:413-432.

4.Michaud, J., Olsen, L. & Olsen, L. Suitability of Asian citrus psyllid, *Diaphorina citri*, as prey for ladybeetles. *BioControl* 49, 417–431 (2004). https://doi.org/10.1023/B:BICO.0000034605.53030.db

5. Milosavljevi´c, I.; Morgan, D.J.; Massie, R.E.; Hoddle, M.S. Density dependent mortality, climate, and Argentine ants affect population dynamics of an invasive citrus pest, *Diaphorina citri,* and its specialist parasitoid, *Tamarixia radiata*, in Southern California, USA. Biol. Control 2021, 159, 104627.

6. Gavarra M.R., Mercado B.G., Progress report on studies dealing with the psyllid vector (*Diaphorina citri* Kuwayama) of greening disease in the Philippines, in: Aubert B., Ke Chung, Gonzales C. (Eds), Proc. 2nd FAOUNDP Regional Workshop, Asian / Pacific Citrus Greening, 1988, pp. 23–28.