Introduction

Citrus is one of the most economically important crops in the U.S. with nearly 500,000 ha in citrus groves in California, Florida, Texas and Arizona. In Florida there are 342,105 ha of citrus groves with an annual earning of about $1.1 billion (Anonymous, 2000.) Like many other crops, citrus is plagued with a host of diseases caused by different etiological agents such as fungi, bacteria, viruses and phytoplasmas. Of all diseases of citrus described to date, citrus greening disease, also known as Huanglongbing or Likubin in Chinese, is considered probably the most destructive and lethal disease of citrus. Two forms of greening disease are currently known (Bové et al. 1974). One is a heat-sensitive type which is found in the southern part of Africa. When the temperature reaches above 30°C for several hours a day, the symptoms fail to develop. The other form is heat-tolerant and is predominantly distributed in Asia and Saudi Arabia. The heat-sensitive bacterium is transmitted by the African citrus psyllid, *Trioza erytreae* (del Guercio) whereas the heat-tolerant bacterium is transmitted by the Asian citrus psyllid, *Diaphorina citri* Kuwayana.

Bionomics of Asian citrus psyllid

![Fig. 1. Adult *D. citri* resting on a twig at 45° angle](http://ipmworld.umn.edu/chapters/TsaiGreening.htm)

The Asian citrus psyllid (*D. citri*) is a member of Sternorrhyncha: Psyllidae. It is considered a serious pest of citrus in the world due to its ability to efficiently transmit the greening agent. However, in the absence of citrus greening agent, this insect is usually a minor pest. This insect was first found in southern Florida in June 1998 and has since spread throughout the state (Halbert et al. 2002, Tsai and Liu 2000). The possible route of Florida introduction could be: 1) This insect has been in South America for many years; thus it could have moved through Central America and the Caribbean. From there it was carried northward by seasonal trade wind or hurricane storm similar to the introduction of the tropical corn pathogens and their insect vectors in the 1970s (Bradfute et al. 1981). 2) The other possibility is that *D. citri* could have been introduced with infested plants from Asia or South America.

*Diaphorina citri* is of known Far Eastern origin (Mead 1977). It is found in all of southeast Asia and the Indian subcontinent, the islands of Reunion and Mauritius, Saudi Arabia, Brazil, Iran, Venezuela, Argentina and Guadeloupe (Cermeli et al. 2000, da Graça 1991, Etienne et al. 1998, Halbert and Manjunath 2004, Mead 1977).
Liu and Tsai (2000) studied the development, survivorship, longevity, reproduction, and life table parameters of *D. citri* at 10ºC, 15ºC, 20ºC, 25ºC, 28ºC, 30ºC and 33ºC. The populations reared at 10ºC and 33ºC failed to develop. Between 15ºC and 30ºC, mean developmental period from egg to adult varied from 49.3 days at 15ºC to 14.1 days at 28ºC. The low-temperature developmental thresholds for 1st through 5th instars were estimated at 11.7ºC, 10.7ºC, 10.1ºC, 10.5ºC and 10.9C respectively. The survival of the 3rd through 5th nymphal instars at 15-28ºC was essentially the same. The mean longevity of females increased with decreasing temperature within 15-30ºC. The maximal longevity of individual females was recorded 117, 60, 56, 52 and 51 days at 15ºC, 20ºC, 25ºC, 28ºC and 30ºC, respectively. The average number of eggs produced per female significantly increased with increasing temperature and reached a maximum of 748.3 eggs at 28ºC (*P* <0.001). The population reared at 28ºC had the highest intrinsic rate of increase (0.199) and net reproductive rate (292.2); and the shortest population doubling time (3.5 days) and mean generation time (28.6 days) compared with populations reared at 15-25ºC. The optimum range of temperatures for *D. citri* population growth was 25-28ºC.

Life table parameters on three commonly grown citrus [rough lemon (*citrus jambhiri* Lush), sour orange (*C. aurantium* L.), grapefruit (*C. paradisi* Macfadyen)] and a non-citrus, orange Jessamine (*Murraya paniculata* (L.) Jack) have also been studied for Florida *D. citri*. The average egg incubation periods on orange Jessamine, grapefruit, rough lemon and sour orange varied very little (4.1-4.2 days). The average nymphal developmental periods on these four host plants were essentially the same except the fifth stadium. Survival of immatures on orange Jessamine, grapefruit, rough lemon and sour orange was 75.4, 84.6, 78.3 and 68.6% respectively. Female adults lived an average of 39.7, 39.7, 47.6 and 43.7 days on these respective host plants. The average number of eggs laid per female on grapefruit (856 eggs) was significantly more than those on other hosts (*P* <0.05). The intrinsic rate of natural increase (*r* *m*) for *D. citri* on grapefruit was highest. The mean population generation time on these hosts ranged from 31.6 to 34.1 days (Tsai and Liu 2000). Besides the size differences, the morphological characteristics of each nymphal stage and field detection were also described (Tsai and Liu 2000), (Figs. 1, 2, 3, 4, and 5). There are at least 56 species of plants including many of the close citrus relatives as hosts of *D. citri* (Halbert and Manjunath 2004). In southern Florida, psyllid populations were positively related to the availability of new shoot flushes which were in turn related to the weekly minimum temperature and rainfall (Tsai et al. 2002).

**Biology of Citrus greening agents**

The greening agents are fastidious phloem-restricted Gram-negative bacteria in the Genus Candidatus Liberibacter of

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12/15/2008
the Graciticutes. The Asian form, a heat tolerant, is named *L. asiaticus* and *L. africanum* for the heat sensitive (African) form on the basis of sequence homology (Garnier et al. 2000, Planet et al. 1995). The African citrus greening develops symptoms only under cool conditions (20-25°C) whereas the Asian greening develops symptoms under both cool and warm (up to 35°C) conditions. The Asian citrus greening is thought to have originated in southern China (Lin and Lin 1990) and was first reported in China in 1943 (Lin 1956), Tsai et al. 1988) and in Taiwan in 1951 (Su and Huang 1990, Su and Hung 2001).

Early symptoms on a greening infected citrus produce a leaf yellowing on a single shoot or branch which is descriptive for the Chinese name of yellow dragon (Fig 6). Infected leaves show a mottled or blotchy appearance (Fig. 7) at the initial stage of symptom development. The yellowing spreads to other parts of the tree and dieback (Fig. 8) and rapid decline follow. At the advanced stage, the leaves are small and often display symptoms similar to zinc or manganese deficiency. Fruits from the infected trees are underdeveloped, unevenly shaped and remain green, thus the name greening disease.

![Fig. 4. Waxy secretion from the nymphs is a good indication of *D. citri* infestation](image1)

![Fig. 5. Heavy *D. citri* feeding on the host can cause deformation of young leaves](image2)
In recent years several accurate diagnostic techniques have been developed. Strain specific DNA probes and PCR are now commonly used to detect the citrus greening agents both in psyllid vectors and in infected plants (Bové et al. 1993, Su and Hung 2001, Tian et al. 1996). The immunofluorescence technique using monoclonal antibodies has been

**Fig. 6.** Citrus infected by citrus greening agent showing typical "yellow shoot" symptoms.

**Fig. 7.** Leaf mottle is often found on the infected citrus

**Fig. 8.** ‘Dieback’ is another symptom of citrus greening infection

Infected trees die within 3-5 years. Infected orange (C. sinensis), mandarin (C. reticulata) and tangelo (C. reticulate x C. paradisi) produce the most severe symptoms. Most citrus cultivars (varieties) are susceptible to the Asian citrus greening. However, it is assumed that at least four isolates (strains) of Asian citrus greening agent have been noted in Taiwan (Su and Hung 2001). Citrus greening bacterium has been experimentally transmitted from the infected citrus to periwinkle (catharanthus roseus), a non-rutaceous host by means of dodder (Cuscuta campestris) (Garnier and Bové 1993, Ke et al. 1988).

The causal agents of citrus greening are transmitted by the psyllid vectors and graft inoculation. Graft transmission was first reported in China in the 1950s (Lin 1956). Although T. erytreae is the natural vector of African citrus greening and D. citri is the natural vector of Asian citrus greening, either psyllid vector can transmit both greening agents under experimental conditions (Lallemand et al. 1986, Massonie et al. 1976). However, it is not known if either vector can be infected simultaneously by both bacteria (Garnier et al. 1996). Vector transmission is the primary means of spread in the field. The minimum acquisition feeding period for D. citri to acquire the greening agent ranges from 30 min. to 5-7 hours and the minimum incubation period in the vector ranged from 1-25 days (Roistacher 1991, Xu et al. 1988). Graft transmission of the citrus greening agent varies with the plant part, the amount of tissue and the pathogen strain (Lin and Lin 1990, Van Vuuren 1993).

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developed for detection of different isolates (strains) of citrus greening agent (Garnier et al. 1987). At least 42 species of citrus and non-citrus plants are known as natural or experimental hosts of citrus greening agents (Halbert and Manjunath 2004).

Prior to finding the citrus greening agent in symptomatic citrus plants in Florida, the \textit{D. citri} vector had become well established throughout the state. Therefore, an eradication program was never initiated. Currently, only the movement of all potential hosts of citrus greening agent or \textit{D. citri} outside of the quarantine counties is prohibited.

Several reports have emphasized the importance of psyllid control with pesticides, especially during the flush period (Aubert 1987, Gonzales and Vinas 1981, Roistacher 1996, Su et al. 1986). With the exception of using neem extracts to control psyllid vector with a good result (Shivankar et al. 2000), there is no information on the availability of environmentally friendly pesticides for citrus psyllid control. In India, trunk injection of antibiotics has been used as part of an integrated management program (Nariani 1981). Treating budwood with tetracycline hydrochloride solution has been practiced in China (Zhao 1981).

Utilization of biocontrol agents yields promising results. Two fungal pathogens, \textit{Cladosporium} sp. nr. \textit{oxyспорum} Berk and M.A. Curtis and \textit{Capnodium citri} Mont. Are known to cause high nymphal mortality in \textit{D. citri} under high relative humidity conditions (Aubert 1987). One ectoparasite, \textit{Tamarixia radiata} (Weterston) and one endoparasite, \textit{Diaphorescyrurus algarghensis} (Shaffee et al.) are effective primary parasites of \textit{D. citri}, but their effectiveness can be greatly affected by hyperparasitism (Aubert 1987, Garnier and Bové 1993). Both parasites have been introduced in Florida with limited success, but only \textit{T. radiata} is established in Florida (McFarland and Hoy 2001, Michaud 2002). Routine removal of symptomatic branches and trees and planting barrier windbreaks have been practiced in China (Ke and Xu 1990).

Like any other perennial woody trees affected by a vectored pathogen, citrus greening disease is very complex and difficult to manage. Currently, the most feasible and workable management practices in China and Taiwan include the use of certified greening free nursery stock, psyllid control and removing sources of inoculum (Hung et al. 2000, Ke and Xu 1990, Su et al. 1986).

Citrus greening is becoming a global problem threatening the very survival of the citrus industry in every citrus producing country. Considerable progress has been made in the study of the dynamics, epidemiology, and molecular characteristics of the causal pathogens of citrus greening disease, but vector-pathogen-plant relationships are still not well understood. It must be hoped that this problem will attract the attention of decision makers from resource rich countries so that more resources are committed to research on molecular aspects of cross protection and genetic breeding and thereby provide a long term solution to this serious problem.

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