Huanglongbing ('Candidatus Liberibacter africanus', 'Candidatus Liberibacter americanus', 'Candidatus Liberibacter asiaticus') and its vectors (African citrus psyllid (Trioza erytreae) and Asiatic citrus psyllid (Diaphorina citri))

> Prepared by Plant Health Australia December 2015

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1 Purpose and background of this contingency plan

This contingency plan framework is designed to provide background information on the pest biology and available control measures to assist with preparedness for an incursion into Australia of huanglongbing and its vector(s). It provides guidelines and options for steps to be considered and undertaken when developing a Response Plan for these pests. Any Response Plan developed using information in whole or in part from this contingency plan must follow procedures as set out in PLANTPLAN and be endorsed by the National Management Group (NMG) prior to implementation.

This contingency plan was developed for Citrus Australia, and is therefore focussed on scenarios relevant to an incursion of huanglongbing in citrus production areas. However, scenarios relevant to an incursion in the Nursery and Garden Industry, or an urban area are also considered.

The information for this plan has been primarily obtained from the documents as cited in the reference section and the contingency plan developed by GAC Beattie and Patricia Barkley in 2009, entitled "Huanglongbing and its Vectors – A Pest Specific Contingency Plan for the Citrus and Nursery and Garden Industries". This will be referred to as Beattie and Barkley (2009).

A contingency plan has also been developed by the Queensland Department of Agriculture, Fisheries and Forestry for the Nursery and Garden industry, entitled "Threat specific contingency plan for huanglongbing and its vectors", which will be referred to as QDAFF (2013).

2 Australian citrus industry

Currently around 12 million trees, covering about 29,780 hectares, form the base of the citrus industry in Australia. A large portion of citrus is bearing fruit (27,931 ha) and a further 1,845 ha is non-bearing. Individual farms are often mixed fruit growing operations and are relatively small (the average area harvested is 18 ha). The majority of growers (41%) have total citrus areas of between 0.25 and 5 ha.

Citrus is produced commercially in all states except for Tasmania, with most of Australia's commercial citrus producers concentrated in the irrigated horticulture regions of southern Australia and Queensland (see Figure 1), including the:

- Riverina, situated in the Murrumbidgee River and Lachlan River area of southern New South Wales
- Murray Valley growing area (including Sunraysia), located along both sides of the Murray River in north western Victoria and south western New South Wales
- Riverland, located along the Murray River in South Australia
- Central Burnett and Emerald regions in Queensland.

Smaller citrus growing areas are situated in various coastal and other locations in New South Wales (Central and North Coast, Narromine), the Northern Territory (Darwin, Katherine), Queensland (Bundaberg, Sunshine Coast, Mareeba area) and Western Australia (Carnarvon, Donnybrook, Gin Gin, Perth and Kununurra).

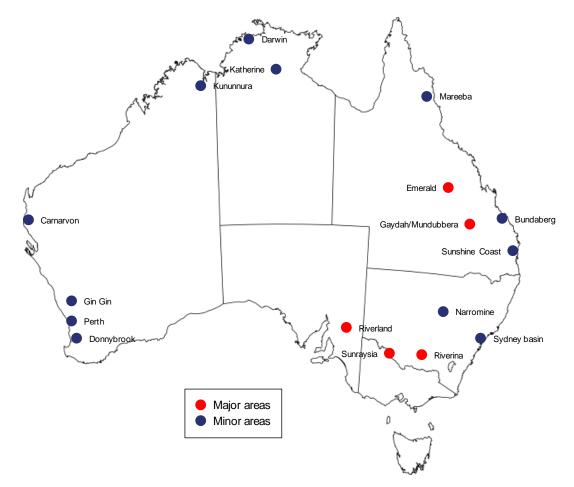


Figure 1. Major citrus production areas in Australia

Over 580,000 tonnes of citrus fruit were produced in Australia in 2013, with over 600,000 tonnes predicted for 2014 (Table 1). In the southern hemisphere, Australia is ranked as the fifth largest producer of citrus, after Brazil, Argentina, South Africa and Peru (FAO 2012). On the global scene, however, Australia is a relatively small player, producing less than 1 % of the world's citrus (FAO 2012).

Oranges are the most commonly grown citrus fruit in Australia, followed by mandarins, lemons, grapefruit, tangelos and limes. Varieties produced vary according to region. While oranges are the main citrus crop grown in the southern irrigation areas, mandarin production is dominant in Queensland. A summary of the main types of citrus fruit produced according to region is supplied in Table 1.

2013 Production (tonnes)								
Variety	Sunraysia	Mid Murray	Riverland	Riverina	NSW other	Qld	WA	Total
Navel	64,000	3,400	98,000	45,455		1,600	5,500	217,955
Valencia	25,500	5,500	50,000	100,000		500	2,700	184,200
Common orange				16,000	13,000			29,000
Imperial mandarin	9,500		11,000	200		25,250	1,500	47,450
Murcott mandarin	200		2,940			18,000	400	21,540
Afourer mandarin	12,800		10,500	1,000		3,000	100	27,400
Other mandarin	4,200		6,000			5,000	1,200	16,400
Lemon	1,500	400	5,000	1,000	1,000	10,000	1,000	19,900
Grapefruit	1,500	2,000	3,000	1,000		500	950	8,950
Tangelo	1,500		5,000					6,500
Lime	800		800	400	800	3,200	400	6,400
Total	121,500	11,300	192,240	165,055	14,800	67,050	13,750	585,695
		Pr	roduction esti	mate 2014 ((tonnes)			
Variety	Sunraysia	Mid Murray	Riverland	Riverina	NSW other	Qld	WA	Total
Navel	65,000	3,500	105,000	58,000		1,500	5,500	238,500
Valencia	25,000	5,000	51,000	110,000		500	2,800	194,300
Common orange				20,000	13,000			33,000
Imperial mandarin	6,500		7,700	150		20,000	2,000	36,350
Murcott mandarin	200		3,000			22,000	400	25,600
Afourer mandarin	13,000		10,500	1,500		3,000	100	28,100
Other mandarin	4,500		6,000			5,000	1,300	16,800
Grapefruit	1,500	2,000	2,000	1,000		500	900	7,900
Tangelo	1,000		3,300					4,300
Lemon	1,500	400	6,000	1,500	1,000	12,000	1,000	23,400
Lime	800		800	400	800	3,200	400	6,400
Total	119,000	10,900	195,300	192,550	14,800	67,700	14,400	614,650

Table 1. Citrus production in Australia (Citrus Australia)

2.1 Notification process for the reporting of suspect pests

Early detection and reporting may prevent or minimise the long-term impact of an incursion into Australia of huanglongbing and/or its vectors. The notification process is described in Figure 2.

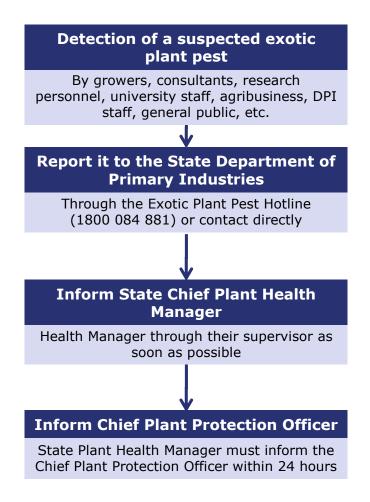


Figure 2. Notification process for the reporting of suspect pests

3 Eradication or containment decision matrix

If huanglongbing and/or its psyllid vectors are detected in Australia, they will be subject to eradication and/or containment processes. The decision to eradicate should be based on the potential economic impact of host damage resulting from the introduction of huanglongbing and/or its vectors, the cost of eradication and technical feasibility. Eradication costs must factor in long term surveys to prove the success of the eradication program. A minimum period with no detection of the pest(s) will be necessary before pest free status can be declared. Eradication of huanglongbing and/or its vectors may be technically feasible if detected while still contained within a small and/or isolated area.

The general decision process for the development of an eradication matrix for huanglongbing and its vectors is outlined in Figure 3 and Table 2 and should be followed in determining if an incursion will result in eradication or management/containment. The final decision between eradication and management will be made through the NMG.

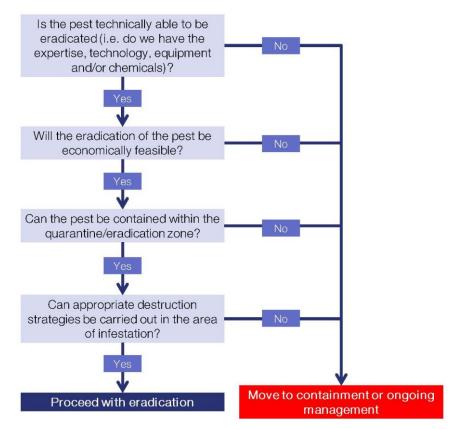


Figure 3. Decision outline for the response to an exotic pest incursion

Table 2. Factors considered in determining whether eradication or alternative action will be taken for an EPP Incident (taken from Table 2; Section 4.16 of PLANTPLAN)

a) the capability to accurately diagnose or identify the EPP.

b) the effectiveness of recommended control technique options, which are likely to be the most costeffective in eradicating the EPP.

c) the ability to remove or destroy all EPPs present by the recommended control techniques.

d) the ability to remove the EPP at a faster rate than it can propagate until proof of freedom can be achieved.

e) the recommended control techniques are publicly acceptable (taking into consideration cultural and social values, humaneness, public health impacts, non-target impacts and environmental impacts).

f) whether Emergency Containment measures have been put in place by the Lead Agency(s).

g) whether there are controls methods, commonly employed for endemic pests and diseases, that may limit or prevent the establishment or impact of the EPP.

h) any legislative impediments to undertaking an emergency response.

i) the resources e.g. chemicals, personnel etc. required to undertake an emergency response are accessible or available.

j) the ability to delimit the known area of infestation.

k) the ability to identify the pathway for entry into, and trace the spread of the EPP within Australia.

I) the ability to determine whether the likelihood of further introductions is sufficiently low.

m) the dispersal ability of the EPP (that is, whether the EPP is capable of rapid spread over large distances).

n) the capability to detect the EPP at very low densities for the purpose of declaring freedom, and that all sites affected by the EPP have or can be found.

o) the ability to put in place surveillance activities to confirm Proof of Freedom for sites possibly infested by the EPP.

p) whether community consultation activities have or will be undertaken

4 Pest information/status – Asiatic citrus psyllid

4.1 Pest details

Common names	Asiatic citrus psyllid, Asian citrus psyllid, oriental citrus psyllid, citrus psylla
Scientific name	Diaphorina citri Kuwayama
Synonym	Euphalerus citri Crawford
Taxonomic position	Kingdom: Animalia
	Phylum: Arthropoda
	Class: Insecta
	Order: Hemiptera
	Suborder: Sternorrhyncha
	Superfamily: Psylloidea
	Family: Liviidae (Previously: Psyllidae)
	Genus: Diaphorina

4.2 Background

The Asiatic citrus psyllid (*Diaphorina citri*) is a small sap-sucking insect that belongs to the Psyllidae family. It feeds on all varieties of citrus and closely related plants in the Rutaceae family (see section 4.5). The psyllid damages citrus directly by feeding on new growth, that can lead to twisting and curling of young leaves and death or 'burn back' of new shoots. As they feed, the psyllids consume large amounts of sap from the plant. The copious amount of excreted honeydew results in growth of sooty moulds that can blemish leaves and in extreme cases reduce photosynthetic capacity.

By itself, the *D. citri* is a relatively minor pest. However, it is considered one of the most serious pests of citrus in the world due to its ability to carry and spread huanglongbing, an extremely destructive disease that is threatening citrus production worldwide. *Diaphorina citri* has the ability to carry and spread all three strains¹ (Asiatic, American and African) of huanglongbing. See: Grafton-Cardwell *et al.*, (2013) and Hall et al., (2013) for further information on *D. citri*.



Figure 4. Adult Asiatic citrus psyllid (Diaphorina citri). David Hall, USDA Agricultural Research Service, Bugwood.org.

¹ Note: this document refers to three "strains" of huanglongbing. Many publications now refer to these as "putative species", for readability the term "strain" is used in this document to refer to the three putative species of bacteria causing huanglongbing (i.e. *'Candidatus* Liberibacter africanus', *'Candidatus* Liberibacter americanus', and *'Candidatus* Liberibacter asiaticus').



Figure 5. Adult Asiatic citrus psyllid (Diaphorina citri) feeding. David Hall, USDA Agricultural Research Service, Bugwood.org.

4.3 Life cycle

Diaphorina citri has a short life-cycle and high fecundity (Catling, 1970a). They prefer warm, dry conditions and are best adapted to regions with high saturation deficits (high temperatures and low relative humidity) (Halbert and Manjunath 2004). *Diaphorina citri* progress from egg through five nymphal instars to the adult stage. At 25°C the population generation time is reported to be 20-22 days (Liu and Tsai, 2000). However, this can vary considerably depending on environmental conditions and under orchard conditions total life lengths of 15-47 have been observed (Husain and Nath, 1927). There is no diapause, however, populations are generally lower during cooler conditions (e.g. winter), during dry times or when citrus is not flushing. *Diaphorina citri* has a strong preference for young, tender flush and when present, adult psyllids are commonly found aggregated on this new flush where they feed and mate. When young flush is not available, psyllid adults can be found on the underside of leaves feeding in the area of the mid-vein (Figure 5; Bonani *et al.*, 2008). Adults have a distinctive feeding stance, feeding with their head down, and the rest of the body raised from the surface at a 45° angle (Figure 4 and Figure 5).

Eggs are laid on the tips of growing shoots, on and between unfurling leaves, on half-folded leaves of the buds, in leaf axils or other suitable places on the young tender parts of the tree (Catling 1970a; Pande 1971; Figure 8 and Figure 9). Females can lay eggs throughout their lives if young leaves are present. They typically lay about 500-800 eggs over a 2 month period, up to a maximum of 1900, depending on environmental conditions and host plant (Hall *et al.*, 2013). Developmental times of eggs and nymphs vary with temperature, with the average development time form egg to adult varying between about 14 days at 28°C and 49 days at 15°C (Liu and Tsai, 2000). New adults reach reproductive maturity within 2-3 days and oviposition begins 1-2 days after mating (Wenninger and Hall 2007).

Humidity, temperature and host plant determine the survival and reproductive rate, with a preference for uniformly warm conditions with suitable flush continually present. Reports of the number of generations per year vary depending on the region. In regions of India and Pakistan, 9 generations

were reported per year, and 11-12 were predicted in the presence of suitable flush growth (Husain and Nath, 1927). Atwal et al (1970) reported 16 generations in India, and in China, 6-11 generations per year have been reported depending on the region where observations were made (Xie *et al.*, 1989; Yang *et al.*, 2006). Reports of average life span also vary. Nava *et al.* (2007) reported adult males live an average of 21-25 days and females 31-32 days at 24°C. Liu and Tsai (2000) report the adult female lifespan to be 51 days at 30°C, with the average lifespan increasing as temperatures become cooler. The maximum lifespan is reported by Liu and Tsai (2000) to be 117 days at 15°C; however, Richardson and Hall (2012) report the maximum life span of virgin females to be 188 days (with an average of 90 days) at 27°C, suggesting reproductive status may also influence survival.

Population fluctuations are closely correlated with the flushing rhythm of citrus trees, as eggs are laid on young flush points and nymphs are dependent on new growth for feeding. *Diaphorina citri* is attracted by the volatiles emitted by new shoot growth (Patt and Sétamou 2010), and therefore increases in psyllid numbers are most evident during periods of abundant new flush. In southern Florida, *D. citri* populations occur throughout the seasons on the ornamental shrub, orange jasmine (*Murraya exotica/ M. paniculata*), but there are population peaks, which are positively related to the weekly minimum temperature and rainfall (Tsai *et al.*, 2002). These periods also coincide with new flush growth of the orange jasmine. This plant is thought to serve as an alternative host for the psyllid when citrus is not in flush because of its more continuous flushing pattern.

While considered a pest of warmer environments, adults and nymphs can survive after being exposed for several hours to temperatures as low as -6°C and eggs can hatch after being exposed for several hours to -8°C. Mild to moderate freeze events are usually non-lethal to adult psyllids but a freeze that kills a flush would be expected to result in mass mortality (Hall *et al.* 2011). Adult *D. citri* (but not eggs) have been shown to become acclimated to colder winter temperatures (Hall *et al.* 2013).

4.4 Dispersal

Adult psyllids are not strong fliers, generally flying distances of about 30-100 m at one time (Hall *et al.*, 2013). These flight distances occur almost every month in Florida, and flight activity regularly peaks in spring (Hall and Hentz 2011). Dispersal over longer distances is wind assisted (Hall *et al.*, 2013), with psyllids moving at least 0.5-1 km during normal air current assisted dispersal flights (Hall *et al.*, 2013) and moving greater distances during high wind events, in particular in open orchards without windbreak protection (Aubert 1987a). Some flight experiments have shown that *D. citri* can fly for up to 3 hours, covering up to 2.4 km, the blue/green (coloured abdomen) morphotype has a greater flight duration than grey/brown morphotype (Martini *et al.*, 2014). The longest flight duration in a flight mill was approximately 50 minutes (Arakawa and Mivamolo, 2007). Recently, a study by Lewis-Rosenblum *et al.* (2015) showed that adults are able to disperse at least 2 km in 12 days and that wind direction did not correlate with dispersal.

Long distance dispersal of psyllids also occurs via the movement of infested host plant material containing viable egg masses or nymphs. *Diaphorina citri* is a well-established hitchhiker (Halbert *et al.*, 2010) and can survive up to 10 days on detached stems and up to 20-30 days on detached stems with fruit (Hall and McCollum 2011). Ornamentals such as orange jasmine (*Murraya exotica/ M. paniculata*) and food plants such as curry leaf (*Bergera koenigii*) have also been known to spread psyllids.

Long distance movement may also occur through wind dispersal and it has been speculated *D. citri* may have been carried up to 147 km across the Everglades in Florida in storms (Halbert *et al.* 2008).

In contrast to adults, psyllid nymphs are not expected to contribute significantly to dispersal as they are not active walkers, are relatively sessile and move only short distances on a branch when disturbed.

4.5 Host range

All known hosts of *D. citri* (see Appendix 1, Table 9) belong to the Rutaceae family, most within the Aurantioideae subfamily. Most commercial citrus are recorded as hosts including grapefruit, lemon, lime, mandarin, orange, pomelo, tangelo and trifoliate orange. Some Australian native citrus have been recorded as hosts overseas (e.g. Australian finger lime (*Citrus australasica*), Australian round lime (*C. australis*) and Australian desert lime (*C. glauca*)) while others (e.g. *Citrus gracilis* and *C. garrawayi*) may well be affected but are not currently recorded as hosts. Several citrus relatives are also hosts, some of which are distributed widely in naturalised vegetation, home gardens and parks, for example, orange jasmine (*Murraya exotica/M. paniculata*), a common ornamental plant and *Murraya paniculata* var. *ovatifoliolata* which grows naturally in north and north-eastern Australia (Brophy et al., 1994).

The suitability of hosts is influenced by a number of factors, including cultivar, ambient temperature, nutrition, soil moisture, light conditions, plant density, the nutritional value and frequency of flush growth and local biotypes of the *D. citri* (Beattie and Barkley, 2009). For example, young citrus trees that flush prolifically under ideal conditions are more suitable resources for the psyllid than older trees growing under identical conditions, or young and old trees grown under poor conditions (Beattie and Barkley, 2009).

4.6 Current geographic distribution

The Asiatic citrus psyllid is native to southern Asia and is currently present in tropical and subtropical Asia, from Arabia and Afghanistan through the Indian subcontinent, eastern Asia and Southeast Asia, including the Indonesian archipelago and Papua New Guinea. It is present in American Samoa, Guam, the Northern Mariana Islands, southern states of the US, Mexico, parts of South America, Central America, the Caribbean and the islands of Mauritius and Reunion. A list of the countries where the pest have been reported is given in Appendix 2, Table 10.

The Asiatic citrus psyllid was detected in Australia, in the Northern Territory, in 1915. Its presence followed an incursion of Citrus canker (*Xanthomonas citri* subsp. *citri*) that is assumed to be related to the introduction of Citrus from Asia in the early 1900s (Bellis *et al.*, 2005). The psyllid was eradicated by chance, as part of the 1916-1922 Citrus canker eradication program.

4.7 Potential geographic distribution in Australia

Diaphorina citri will survive a wide range of temperature extremes from –7°C to 45°C (Aubert 1990). In addition, psyllid populations prefer low altitudes (typically <800 m above sea level) and high saturation deficits (warm to very hot climates with low relative humidity) (Beattie and Barkley, 2009) but can be found outside of these limits. The ideal temperature range for psyllid development is between 25 - 28°C (Liu and Tsai, 2000). At these temperatures, the reproductive potential of Asian citrus psyllid is highest.

Diaphorina citri is distributed in a wide range of geographic regions (Halbert and Manjunath 2004), many of which are climatically similar to parts of Australia. Climate modelling, and observations in Asia, suggests that the Asiatic citrus psyllid is likely to survive in much of Australia, including all citrus growing areas (Aurambout *et al.*, 2009). Indications are that populations are likely to be highest in the citrus

production areas of the Central Burnett, Darwin, Kathrine, Kununurra, Riverland, Riverina and Murray Valley regions. In addition, most production nurseries across Australia would provide the right microclimate to support proliferation of *D. citri*.

4.8 Symptoms

New leaf growth is most severely damaged, and may show twisting, curling, notching and deformation, as well as premature defoliation, stunting and sometimes death of shoots. Heavily infested plants are sometimes characterised by the rosette appearance of shoot tips (OEPP/EPPO 2005a; Halbert and Manjunath 2004). Abscission (detachment) of leaves, death of the apical meristem and complete abscission of the terminal have also been reported (Michaud, 2004). Heavy infestations may lead to flower and fruitlet drop (OEPP/EPPO 2005a).

One of the most easily recognisable symptoms is the appearance of white waxy secretions and honeydew on new growth (Figure 6). This is produced as the psyllids feed and extract large quantities of sap and may lead to black sooty mould development. In hot humid weather $(36 - 38^{\circ}C)$ the honeydew melts to form clear droplets (Beattie and Barkley 2009).



Figure 6. White waxy secretions of the Asiatic citrus psyllid. Source: Douglas L. Caldwell, University of Florida IFAS Extension

4.9 Diagnostic information

A draft diagnostic protocol has been prepared for the diagnosis of Asiatic citrus psyllid and the African citrus psyllid (Malipatil and Semeraro 2007), but is yet to be endorsed by the Subcommittee on Plant Health Diagnostics (SPHD). Once endorsed it will be available from:

http://plantbiosecuritydiagnostics.net.au/.

The diagnostic information presented here has been adapted from Hall *et al.*, (2013) and OEPP/EPPO (2005a).

Adult *D. citri* are 2.7-3.3 mm long with a mottled brown forewing and long slender hind wings. The abdominal colour varies between three distinctive colour patterns (morphotypes) of grey/brown, blue/green or orange/yellow. Antennae have black tips with two small light brown spots on the middle segments. Adults are very active and will jump on the slightest disturbance. They have a distinctive feeding stance, holding their bodies at a 45° angle from the plant surface.

Eggs of *D. citri* are about 0.3 mm long by 0.15 mm wide, almond shaped and anchored to new flush with a tapered stalk (pedicel) at the posterior/lower end. They are light yellow in colour when freshly deposited, but gradually turn bright orange with two distinct red eye spots as they reach maturity (Figure 8). They are laid on buds and young flush, on the tips of growing shoots, and on and between unfurling leaves, with the long axis vertical to the surface. Many eggs may be found on a single flush shoot.

Nymphs pass through five instars, are light-yellow to dark-brown, with well-developed wing pods (Figure 10). They are usually found clustered in groups and are sedentary unless disturbed in which case they will quickly move away.

It should be noted that surveys by CSIRO have failed to detect any psyllids on Aurantioideae in Australia (Beattie and Barkley, 2009) and therefore any psyllids detected on citrus or other Aurantioideae should be considered exotic and potential vectors of huanglongbing.



Figure 7. Asiatic citrus psyllid (Diaphorina citri) adult. Source: Pest and Diseases Image Library, Bugwood.org



Figure 8. Asiatic citrus psyllid (Diaphorina citri) eggs. Source: David Hall, USDA Agricultural Research Service, Bugwood.org.



Figure 9. Asiatic citrus psyllid (Diaphorina citri) eggs. Source: Douglas L. Caldwell, University of Florida.



Figure 10. Nymph stages of the Asiatic citrus psyllid (Diaphorina citri). Source: David Hall, USDA Agricultural Research Service, Bugwood.org.

4.9.1 Determine if psyllids are carrying huanglongbing

As the Asiatic citrus psyllid can spread huanglongbing it is important to determine if they are carrying the bacterium. To do this, psyllids should be sent in a phial 70% ethanol (ethyl alcohol) for identification.

If confirmed to be Asiatic citrus psyllid specimens (minimum of three) should be sent (in a 95% ethanol solution) for PCR analyses for the presence of '*Candidatus* Liberibacter spp.'

4.10 Pest risk analysis – Asiatic citrus psyllid

Potential or impact	Rating
Entry potential	HIGH
Establishment potential	HIGH
Spread potential	HIGH
Economic impact	HIGH - EXTREME
Overall risk	HIGH - EXTREME

See Appendix 3, Section 12.3 for further details regarding these risks.

5 Pest information/status – African citrus psyllid

5.1 Pest details

Common names	African citrus psyllid, African citrus psylla, Citrus psylla, Two-spotted citrus psyllid
Scientific name	Trioza erytreae del Guercio
Synonyms	<i>Aleurodes erytreae</i> del Guercio <i>Spanioza eritreae</i> del Guercio
	Spanioza erythreae del Guercio Spanioza merwei Pettey Trioza citri Laing Trioza erythreae del Guercio
Taxonomic position	Trioza merwei Pettey Kingdom: Animalia Phylum: Arthropoda Class: Insecta Order: Hemiptera Suborder: Sternorrhyncha
	Superfamily: Psylloidea Family: Triozidae Genus: <i>Trioza</i>

5.2 Background

The African citrus psyllid (*Trioza erytreae*) is a small, sap-sucking insect (Figure 11) that belongs to the Triozidae family. It feeds on several varieties of citrus and closely related plants in the Rutaceae family (see section 5.5). This species damages citrus directly by feeding of new growth. Nymphs feeding on the underside of the leaves result in characteristic cup shaped distortions on the leaves (Figure 14 and Figure 15). Infestations can also cause mild chlorosis to develop on infested plants (Catling 1973).

By itself, *T. erytreae* is a moderate pest, causing leaf distortion and mild chlorosis. However, it is considered a serious pest of citrus due to its ability to vector huanglongbing, an extremely destructive disease that is threatening citrus production worldwide. *Trioza erytreae* has the ability to vector the Asiatic and African strains of huanglongbing (Massonie *et al.*, 1976).



Figure 11. Adult African citrus psyllid (Trioza erytreae). Source: S.P. van Vuuren, Citrus Research International, Bugwood.org)

5.3 Life cycle

Trioza erytreae has a short life-cycle and high fecundity (Van Den Berg *et al.*, 1991). Unlike the Asiatic citrus psyllid, the African citrus psyllid is sensitive to high temperatures and dry weather, preferring cooler, more humid areas, this is very similar to the conditions required by the African strain of huanglongbing. Saturation deficits above 45 millibars (i.e. warm dry conditions) results in 100 % mortality of eggs and nymphs (Aubert 1987b). High mortality has been reported when temperatures exceed 32°C for several hours per day (Catling and Green 1972), suggesting that establishment of this species in Australia could be limited by temperature.

Adult males are sexually mature as soon as they have completed their final moult (Van Den Berg *et al.*, 1991). Females are sexually mature a few days after their final moult and have been show to lay eggs 3-5 days (at 24-26°C) or 6-7 days (at 14-16°C) after their final moult. Mated females lay eggs for 16±1.5 days producing a mean of 591±60.8 eggs. A second mating usually occurs as females lay a mean of 982 eggs over their life (Van Den Berg *et al.*, 1991).

The small (0.5 mm long) orange coloured eggs (Figure 12) are usually laid on the margins of the young leaves of suitable host plants. Eggs are reported to have a 95% hatching rate in South Africa (Van Der Berg *et al.*, 1991). The eggs then take 6-15 days to hatch before passing through 5 nymphal stages

over 17-43 days (Catling 1973). Development of nymphs and eggs is temperature dependent (Catling 1973). Populations fluctuate in response to climatic conditions and the flushes of new growth on host plants, which provide suitable habitat for the development of nymphs (Catling 1972). Reproduction occurs year round and there is no diapause (Catling 1973).

Trioza erytreae is only able to reproduce on new growth of suitable host plants (Aubert 1987b). Although the adults are able to feed on a range of plants in the Rutaceae family there are fewer plants that eggs are laid on or nymphs can feed on. Nymphal feeding is only known to occur on the plants listed in Table 3.

Common name	Scientific name	Notes
False horsewood	Clausena anisata	Nymphs very commonly observed
White ironwood	Vepris lanceolata	Nymphs very commonly observed
Lemon	Citrus x limon	Nymphs very commonly observed
Sweet orange	Citrus x aurantium (syn. Citrus sinensis)	Nymphs often observed
Mandarin	Citrus reticulata	Nymphs often observed
pomelo	Citrus maxima (syn. Citrus grandis)	Nymphs occasionally observed
Orange jasmine	Murraya paniculata (syn. M. exotica)	Nymphs occasionally observed
Small knobwood	Zanthoxylum capense (Fagara capense)	Nymphs occasionally observed

Table 3 Plants able to support the development of African citrus psyllid nymphs (Source: Aubert 1987b)



Figure 12. African citrus psyllid eggs. Source: Peter Stephen, Citrus Research International, Bugwood.org



Figure 13. African citrus psyllid nymphs and cup shaped leaf galls. Source: Peter Stephen, Citrus Research International, Bugwood.org

5.4 Dispersal

Adult psyllids are able to jump or fly strongly over short distances when disturbed, but generally do not move large distances from suitable habitat (Catling 1973).

Artificial release and yellow sticky trap recapture studies have shown that adult *T. erytreae* are able to disperse at least 1.5 km from its point of release (Van Den Berg and Deacon 1988). Adults have been shown to survive for up to 85 hours (approximately 3.5 days) in the absence of host plants (Van Den Berg and Deacon 1988).

The movement of nursery material (budwood, trees, seedlings, and rootstock) can allow the dispersal of the African citrus psyllid (especially nymphs, which live in cup shaped galls on leaves) over long distances. Once in a new area adults fly to new hosts and establish new populations.

5.5 Host range

All known hosts of *T. erytreae* (see Appendix 4, Table 11) belong to the Rutaceae family. Most commercial citrus are recorded as hosts including grapefruit, lemon, lime, mandarin, orange, pomelo, tangelo and trifoliate orange. Some Australian native citrus species have been recorded as hosts overseas (e.g. Australian finger lime (*Citrus australasica*)). Several citrus relatives are also hosts, some of which are distributed widely in home gardens and parks (e.g. *Murraya* spp.).

5.6 Current geographic distribution

The African citrus psyllid is native to sub-Saharan Africa, but has now spread across much of southern and eastern Africa, to off shore islands (such as Madagascar, the Canary Islands Reunion and Saint Helena), parts of the Middle East and recently to Spain and Portugal.

A list of the countries where the bacteria has been reported is given in Appendix 5, Table 12.

5.7 Potential geographic distribution in Australia

The African citrus psyllid is less tolerant of high temperatures and dry conditions than the Asiatic citrus psyllid. Saturation deficits above 45 millibars (i.e. warm dry conditions) results in 100% mortality of eggs and nymphs (Aubert 1987b). High mortality has also been reported when temperatures exceed 32°C for several hours per day (Catling and Green 1972). This suggests that the climate in southern citrus growing regions of Australia may be more suitable for the establishment and spread of the African citrus psyllid than the subtropical and tropical regions of Australia.

5.8 Symptoms

T. erytreae lays eggs on new growth. Nymphs that hatch create a feeding site on the underside of the leaf. The leaf tissue around the feeding site becomes distorted creating a cup shaped gall on the leaf (Figure 13, Figure 14, Figure 15). This is one of the most obvious symptoms of this pest. Feeding can also cause leaf chlorosis to develop.

While feeding adults and nymphs produce a white granular excretion that can cover the tree and surrounding earth (Annecke and Cilliers 1963).



Figure 14. Feeding damage caused by the African citrus psyllid. Source: Peter Stephen, Citrus Research International, Bugwood.org



Figure 15. Feeding damage caused by the African citrus psyllid. Source: INRA-Bordeaux Archive, Institut National de la Recherche Agronomique, Bugwood.org

5.9 Diagnostic information

A draft diagnostic protocol has been prepared for the diagnosis of Asiatic citrus psyllid and the African citrus psyllid (Malipatil and Semeraro 2007), but is yet to be endorsed by the Subcommittee on Plant Health Diagnostics (SPHD). Once endorsed it will be available from:

http://plantbiosecuritydiagnostics.net.au/.

Trioza erytreae eggs are 0.5 mm long, yellow to orange in colour and have a thin "stalk" that attaches the egg to the plant. Eggs are laid on margins of leaves and occasionally on young thorns (Annecke and Cilliers 1963; Figure 12).

Newly hatched nymphs are orange and become orange, yellow or greenish coloured as they age. The nymphs feed on the underside of the leaf. Feeding creates a cup shaped depression in the leaf and the nymph lives in this (Figure 13). The nymphs are dorso-ventrally compressed and typically surrounded by a light coloured waxy fringe (Annecke and Cilliers 1963). Nymphs are sedentary and live in colonies, causing severe leaf distortion and chlorosis (OEPP/EPPO 2005b).

Adult psyllids are approximately 4 mm long, thin bodied and possess wings (OEPP/EPPO 2005b). Newly emerged adults are light green becoming yellow then become brown as they age. All have, black coloured eyes and hyaline wings. The hind legs of the adult psyllid are stouter than front pairs (Annecke and Cilliers 1963). Female psyllids have a pointed abdomen; males have a blunter shaped abdomen and are typically smaller and darker coloured than females (OEPP/EPPO 2005b). Dissection is required to identify *T. erytreae* to a species level. Further information is available in OEPP/EPPO (2005b).

When feeding adults sit with their abdomen raised at a 35° angle to the plant surface (OEPP/EPPO 2005b). Adults and nymphs produce a white granular excretion whilst feeding, which can cover the tree and surrounding earth (Annecke and Cilliers 1963). When disturbed adult psyllids jump or fly (OEPP/EPPO 2005b).

It should be noted that surveys by CSIRO have failed to detect any psyllids on Aurantioideae in Australia (Beattie and Barkley, 2009) and therefore all psyllids detected on citrus or other Aurantioideae should be considered exotic and potential vectors of huanglongbing.

Further diagnostic information is available from: OEPP/EPPO (2005b).

5.9.1 Determine if psyllids are carrying huanglongbing

As the African citrus psyllid can spread huanglongbing it is important to determine if they are carrying the bacterium. To do this, psyllids should be sent in a phial 70% ethanol (ethyl alcohol) for identification.

If confirmed to be African citrus psyllid specimens (minimum of three) should be sent (in a 95% ethanol solution) for PCR analyses for the presence of '*Candidatus* Liberibacter spp.'

5.10 Pest risk analysis – African citrus psyllid

Potential or impact	Rating
Entry potential	MEDIUM
Establishment potential	HIGH
Spread potential	HIGH
Economic impact	HIGH - EXTREME
Overall risk	HIGH - EXTREME

See Appendix 6, Section 12.6 for further details regarding these risks.

6 Other potential vectors of huanglongbing

There are also reports of two other psyllids (described in 6.1 and 6.1) that are capable of vectoring the bacteria. But they have not been as extensively studied as the Asiatic citrus psyllids and African citrus psyllids.

6.1 Pomelo psyllid (Cacopsylla citrisuga)

Pomelo psyllid (*Cacopsylla citrisuga*) is reported to vector the Asiatic strain of Huanglongbing ('*Candidatus* Liberibacter asiaticus') in China (Cen *et al.*, 2012).

Common names	Pomelo psyllid
Scientific name	Cacopsylla citrisuga Yang & Li
Synonyms	Psylla citrisuga Yang & Li
Taxonomic position	Kingdom: Animalia
	Phylum: Arthropoda
	Class: Insecta
	Order: Hemiptera
	Suborder: Sternorrhyncha
	Superfamily: Psylloidea
	Family: Psyllidae
	Genus: Cacophylla

6.1.1 Pest details

6.2 Black psyllid (Diaphorina communis)

Black psyllid (*Diaphorina communis*) in Bhutan was reported to contain the Asiatic strain of Huanglongbing ('*Candidatus* Liberibacter asiaticus') (Donovan *et al.*, 2012). Transmission studies have yet to confirm the vector status of this species.

6.2.1 Pest details

Common names	Black psyllid
Scientific name	Diaphorina communis Mathur
Synonyms	Diaphorina mathura Loginova
Taxonomic position	Kingdom: Animalia
	Phylum: Arthropoda
	Class: Insecta
	Order: Hemiptera
	Suborder: Sternorrhyncha
	Superfamily: Psylloidea
	Family: Liviidae
	Genus: Diaphorina

7 Pest information/status – huanglongbing

7.1 Pest details

Common names	Huanglongbing, HLB, citrus greening, blotchy mottle disease of citrus, greening, yellow branch disease, yellow shoot, yellow dragon disease, citrus dieback (India), leaf mottling (Philippines), blotchy mottle (Philippines), mottle leaf disease (Philippines), citrus vein phloem degeneration (Indonesia), likubin (Taiwan)	
Scientific names	<i>'Candidatus</i> Liberibacter africanus' (African strain) <i>'Candidatus</i> Liberibacter americanus' (American strain) <i>'Candidatus</i> Liberibacter asiaticus' (Asiatic strain)	
Synonyms	Candidatus Liberobacter africanum Monique Garnier Liberobacter africanum Monique Garnier Liberibacter americanus Candidatus Liberobacter asiaticum Monique Garnier Liberobacter asiaticum Monique Garnier	
Taxonomic position	Kingdom: Bacteria Phylum: Proteobacteria Class: Alphaproteobacteria Order: Rhizobiales Family: Rhizobiaceae Genus: <i>Candidatus</i> Liberibacter	

7.2 Background

Huanglongbing is the most serious citrus disease worldwide (Sechler *et al.*, 2009; Gottwald 2010). The bacterial disease causes fruit to become bitter, misshapen (lop-sided) and trees to go into decline and eventually die. In countries where the disease is endemic trees may only live for 5-8 years and never bear saleable fruit (Halbert and Manjunath 2004). In the absence of vectors, the disease is self-limiting as infected trees die as a result of the infection (although the disease can still be spread via grafts from infected trees) (Halbert *et al.*, 2008).

The disease is caused by three strains of gram-negative bacteria, which colonise the phloem sieve tubes of the infected plant (Lopes and Frare 2008; Teixeira *et al.*, 2008), limiting sugar transport (Tatineni *et al.*, 2008). All three strains are transmitted by grafts, and marcotting (air layering) although not all grafts taken from an infected tree may be infected as the bacteria are not evenly distributed in the tree. This also means that false negatives can occur (Gottwald 2010). Huanglongbing is also vectored by four species of citrus psyllid (the vector-bacteria relationship is described below). Experiments have also shown that dodder (*Cuscuta* spp.) can also transmit huanglongbing between plants (e.g. Zhang *et al.*, 2010).

The three² strains of bacteria that are reported to cause the citrus disease huanglongbing are:

- 'Candidatus Liberibacter africanus' (African strain)
- 'Candidatus Liberibacter americanus' (American strain)
- 'Candidatus Liberibacter asiaticus' (Asiatic strain)

The strains differ in their ability to tolerate different temperatures and their current geographic distribution. All cause similar symptoms (tree decline, leaf chlorosis, poorly developed (lop-sided) and poor tasting (bitter) fruit, etc.).

The Asiatic strain is the most damaging of the three strains and has recently been reported from the United States of America, where is has spread rapidly and is having a significant impact on the United States citrus industry (Gottwald 2010). The Asiatic strain of the bacteria is described as heat tolerant and can cause symptoms and multiply at temperatures above 35°C (Lopes *et al.*, 2009).

The African strain of huanglongbing was first reported in the 1920s in South Africa (Pietersen *et al.*, 2010). The strain is currently confined to Africa and the Middle East. This strain is reported to be heat sensitive, with plants able to recover from symptoms at temperatures above 32°C (Pietersen *et al.*, 2010). Symptoms are similar to the Asian strain but usually less severe die back is observed (Gottwald *et al.*, 2007), and it is considered to be less damaging than the Asian strain of the disease (Van Den Berg 1990).

The American strain of huanglongbing is the most recently described (Lopes and Frare 2008). It has been shown to be heat sensitive, with experiments showing that temperatures of over 32°C were detrimental to the bacteria (Lopes *et al.*, 2009). Symptoms and severity is considered to be similar to the Asian strain (Gottwald *et al.*, 2007). To date this strain has only been recorded in Brazil (Teixeira *et al.*, 2005) and north western Argentina (Ramallo *et al.*, 2008).

7.3 Disease cycle

Huanglongbing is a gram-negative, phloem-limited bacteria (Lopes and Frare 2008; Teixeira *et al.*, 2008) that is transmittable via grafting or a psyllid vector (Table 4). The incubation period is variable and ranges from months to years; there is a suggestion that older orchards (7-10 years old) have a longer latent period (1-2.5 years) than younger orchards (6-12 months) (Gottwald 2010).

The three strains differ in their ability to multiply or cause symptoms at high temperatures. Both the American and African strains do not produce obvious symptoms or multiply at temperatures of 32°C or above (Lopes *et al.*, 2009; Pietersen *et al.*, 2010), while the Asiatic strain is able to multiply at temperatures above 32°C (Pietersen *et al.*, 2010). Because of this the Asiatic strain is often described in the scientific literature as being heat tolerant (e.g. Tatineni *et al.*, 2008; Hoffman *et al.*, 2013).

Once infected the bacteria concentrates in the bark tissue, leaf midribs, roots, flowers and fruit, but does not colonise the endosperm or embryo (Tatineni *et al.*, 2008). Roots are reported to be colonised before the bacteria moves to the leaves and foliar symptoms are expressed (Johnson *et al.*, 2014). Seeds are not known to transmit the bacteria (*et al.*Albrecht and Bowman 2009; Hoffmann *et al.*, 2013).

² 'Candidatus Liberibacter caribbeanus' has recently been reported affecting *Citrus sinensis* in Columbia. This bacterium is reported to be vectored by the Asian citrus psyllid (*D. citri*) (Keremane *et al.*, 2015). This is likely to be a fourth strain of huanglongbing but limited research has been carried out to date.

7.4 Dispersal

Huanglongbing is transmitted via grafting, marcotting or by its psyllid vectors (Table 4). Seeds are not known to transmit the bacteria (*et al*.Albrecht and Bowman 2009; Hoffmann *et al.*, 2013). Dodder (*Cuscuta* spp.) can also act as a vector of huanglongbing and has been used experimentally to transmit the bacteria from citrus to other plants such as periwinkle (*Catharanthus roseus*) (e.g. Zhang *et al.*, 2010).

Table 4 Psyllid vectors of the three strains of huanglongbing

Strain	Confirmed vector(s)
'Candidatus Liberibacter asiaticus' (Asiatic strain)	Asiatic citrus psyllid (Diaphorina citri)
	African citrus psyllid (Trioza erytreae)
	Pomelo psyllid (<i>Cacopsylla citrisuga</i>) ³
	Black psyllid (<i>Diaphorina communis</i>) ⁴
'Candidatus Liberibacter africanus' (African strain)	Asiatic citrus psyllid (Diaphorina citri)
	African citrus psyllid (Trioza erytreae)
'Candidatus Liberibacter americanus' (American strain)	Asiatic citrus psyllid (Diaphorina citri)

The movement of infected nursery material (budwood, rootstock or nursery plants) would allow the rapid spread of the bacteria between regions. The bacteria could also be spread by the movement of infected psyllids, as adult, fourth and fifth instar psyllids can carry the bacteria (Xu *et al.*, 1988; Gottwald *et al.*, 2007) and infect new plants when feeding.

The disease can spread if infected trees are used as a source of budwood, top worked, by root grafting or if the vectors are present (currently the known psyllid vectors of huanglongbing are exotic and surveys by CSIRO have failed to detect any psyllids on Aurantioideae in Australia (Beattie and Barkley, 2009) and therefore all psyllids detected on citrus or other Aurantioideae should be considered exotic and potential vectors of huanglongbing).

Seeds are not known to transmit the bacteria (Albrecht and Bowman 2009; Hoffmann et al., 2013).

Drastic pruning is unlikely to be effective in eradication the disease as Futch *et al.* (2009) found 66% of stumps of infected trees contained one or more positive sprouts 180 after removal of the canopy.

³ Pomelo psyllid (*Cacopsylla citrisuga*) was recently reported by Cen *et al.*, (2012) to act as a vector of the Asian strain of Huanglongbing in China.

⁴ In Bhutan the Black psyllid (*Diaphorina communis*) was reported to contain the Asiatic strain of Huanglongbing ('*Candidatus* Liberibacter asiaticus') (Donovan *et al.*, 2012). Studies have yet to confirm the vector status of the Black psyllid.

7.5 Host range

Most of the known hosts of huanglongbing (see Appendix 7, Table 13) belong to the Rutaceae family, some exceptions include; periwinkle (*Catharanthus roseus*) (Apocynaceae), which was infected experimentally using dodder (*Cuscuta pentagona*) (Convolvulaceae) plants to transmit the bacteria from infected lemon trees to periwinkle plants (Zhang *et al.*, 2010). Dodder has also been used to experimentally transmit the bacteria to tobacco (*Nicotiana tabacum*) (Solanaceae) (Francischini *et al.*, 2007).

Most commercial citrus are recorded as hosts including grapefruit, lemon, lime, mandarin, orange, pomelo, tangelo and trifoliate orange. Some Australian native citrus species have been recorded as hosts overseas (e.g. Australian finger lime (*Citrus australasica*)). Several citrus relatives are also hosts, some of which are distributed widely in home gardens and parks (e.g. orange jasmine). See Appendix 7, Table 13 for a complete list of hosts.

7.6 Current geographic distribution

The three stains of the bacteria have different distributions. The Asian strain is the most widespread occurring in Africa, Asia, North America, South America, and in Central America. The African strain is the next most widely distributed, being found predominantly in Africa and in Yemen in western Asia. The American Strain of huanglongbing is confined to Brazil (Teixeira *et al.*, 2005) and north western Argentina (Ramallo *et al.*, 2008).

A list of the countries where the bacteria has been reported is given in Appendix 8, Table 14.

7.7 Potential geographic distribution in Australia

In the absence of a suitable vector (see Table 4) huanglongbing can be spread via graft transmission. The physiological factor limiting its potential distribution in Australia appears to be the bacteria's ability to survive different temperatures.

The Asiatic strain of huanglongbing is described as heat tolerant and can cause symptoms and multiply at temperatures above 35°C (Lopes *et al.*, 2009). The African strain of huanglongbing is reported to be heat sensitive, with plants able to recover from symptoms at temperatures above 32°C (Pietersen *et al.*, 2010). The American strain of huanglongbing has also been shown to be heat sensitive, with experiments showing that temperatures of over 32°C were detrimental to the bacteria (Lopes *et al.*, 2009).

Therefore, much of Australia would have conditions suitable for the establishment of one or more of the three strains of huanglongbing, based on the temperature thresholds reported overseas.

7.8 Symptoms

Huanglongbing infection causes a range of symptoms that vary in severity from mild to severe. Typically, lemons and limes show less severe symptoms than oranges, mandarins and tangelos, while *Poncirus* spp. are tolerant (but not immune) to huanglongbing. Disease incidence and severity is faster in younger compared with older blocks (Bassanezi and Bassanezi 2008).

Typical symptoms include: complete yellowing of leaves and growing shoots (Figure 16 and Figure 17) and mottling of leaves that crosses leaf veins and is asymmetrical on the leaf blade (Figure 18). Branch dieback and thickening of midribs and veins may also be observed. Other symptoms include unseasonal and heavy flowering on diseased branches and out of phase flushing. Chronically infected trees are sparsely foliated with extensive twig and limb dieback and small, upright leaves with compressed internodes. Eventually the tree goes into complete decline and dies.

Fruit may be small, lopsided (when cut longitudinally the axis of the fruit will appear curved, with some of the fruit more developed than the rest) (Figure 19), hard and bitter-tasting with dark, aborted seeds. Fruit may also remain partially green (Figure 20) or ripen backwards and excessive fruit drop may also be observed.

Huanglongbing can be confused with mineral deficiencies, particularly zinc, however, mottling of leaves crosses veins in huanglongbing and is asymmetrical, whereas in zinc deficiency, mottling occurs symmetrically between or along leaf veins. Huanglongbing also causes similar symptoms to the bacterial disease, Australian citrus dieback.

The foliar symptoms on *Murraya* spp. are similar to citrus symptoms (QDAFF 2013) and include: shoot yellowing and die back (Lopes *et al.*, 2010).

In Australia, the established disease most closely resembling huanglongbing symptoms is citrus dieback, a phytoplasma-associated disease.

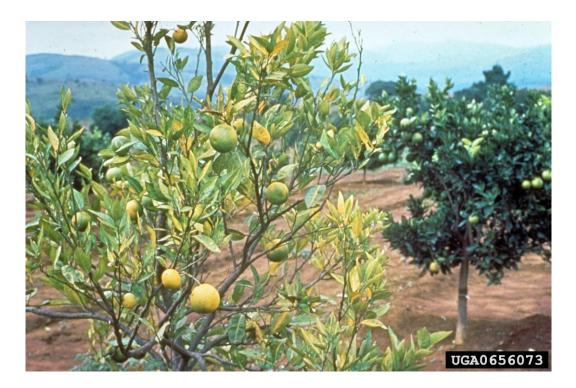


Figure 16 Yellowing of shoots on sweet orange caused by 'Candidatus Liberibacter asiaticus'. Source: H.D. Catling, Bugwood.org



Figure 17 Chlorotic leaves with green islands caused by 'Candidatus Liberibacter asiaticus'. Source: Hilda Gomez, USDA



Figure 18 Asymmetrical leaf mottling symptoms caused by 'Candidatus Liberibacter asiaticus'. Source: Hilda Gomez, USDA



Figure 19 Small lopsided fruit caused by 'Candidatus Liberibacter asiaticus'. Source: Jeffrey W. Lotz, Florida Department of Agriculture and Consumer Services, Bugwood.org



Figure 20 Mottled leaves and partially green fruit caused by 'Candidatus Liberibacter asiaticus'. Source: Jeffrey W. Lotz, Florida Department of Agriculture and Consumer Services, Bugwood.org

7.9 Diagnostic information

Diagnosis of huanglongbing symptoms must be confirmed using appropriate molecular methods. Conventional or real-time PCR (Polymerase Chain Reaction) is recommended for the diagnosis of the bacteria (Li *et al.*, 2008; Bertolini *et al.*, 2014). These tests can determine the presence or absence of huanglongbing (tests are more accurate when symptoms are present) and are capable of distinguishing between the three strains of the disease.

A nationally endorsed diagnostic protocol (NDP 25) has been developed for the diagnosis of 'Candidatus Liberibacter asiaticus' (SPHDS 2014). This protocol is available from: http://plantbiosecuritydiagnostics.net.au/resource-hub/protocols/national-diagnosticprotocols/.

The North American Plant Protection Organization (NAPPO) also has developed a diagnostic protocol for the three strains of bacteria causing huanglongbing (NAPPO 2012) that is available from: www.phytosanitary.info/sites/phytosanitary.info/files/NAPPO%20HLB%20DP%202%202012-04-10-e_0.pdf.

Similarly, the European and Mediterranean Plant Protection Organization has prepared a diagnostic protocol for the three strains of bacteria causing huanglongbing (EPPO 2014)

There is also a commercially available Agdia rapid method for the detection of '*Candidatus* Liberibacter spp.' in trees or vectors and can provide results in as little as 30 minutes (Russell *et al.* 2015).

7.10 Pest risk analysis – huanglongbing

Potential or impact	Rating
Entry potential	HIGH
Establishment potential	HIGH
Spread potential	HIGH
Economic impact	EXTREME
Overall risk	EXTREME

See Appendix 9, Section 12.9 for further details regarding these risks.

8 Pest management

8.1 General procedures for control

Eradication/management of huanglongbing and/or its vectors will be reliant on the use of pesticides to control the vector and the destruction of huanglongbing infected and surrounding plants⁵ (to remove them as sources of infection). The spread of the bacteria and vectors should be controlled by managing the movement of plant material, people and machinery to and from the area. Specific control measures will be determined by a CCEPP, however, general procedures include:

- Keep traffic out of affected areas and minimise movement in adjacent areas.
- Adopt best-practice property hygiene procedures to restrict the spread of plant material, including plant material that may be adhering to machinery, etc., between orchards and adjacent properties.
- After surveys are completed, and permission has been obtained from the Chief Plant Health Manager or the CCEPP, destruction of the infected/infested plant material, may be an effective control.
- On-going surveillance of infected/infested areas to ensure the pest is eradicated.

Table 5 provides a summary of the key activities required in the event of the detection of huanglongbing and/or its vectors. Note, these activities can be carried out concurrently.

Table 5 General procedures to follow in the event of the detection of huanglongbing and/or its w	/ectors
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Scenario	Activities required
If vectors present but disease has not been discovered	 Immediately treat all hosts in the immediate are with suitable chemicals (see Section 8.4) to destroy any psyllids in infested area. Hosts in the area may also be destroyed (or all leaves removed) to remove habitat for the psyllids
	• Carry out surveys in the surrounding area (including gardens, native bushland, commercial and abandoned orchards) for other psyllid populations
	Trace the movement of plant material to and from the infested area and control any populations discovered
	 Test any huanglongbing hosts in the area where psyllids were discovered to determine if huanglongbing is present
	Psyllids should also be tested for the presence of huanglongbing

⁵ Infection could be determined using PCR tests. The distance around infected plants that will need to be removed will depend on the presence or absence of vectors and management practices.

Scenario	Activities required
If disease is present but vectors have not been discovered	 Immediately destroy infected plants and surrounding plants Trace the movement of propagation material to and from the infected area and control any infections Carry out surveys in the surrounding area (including gardens, native bushland, commercial and abandoned orchards) for psyllid populations
If vector and disease are present	 Immediately destroy huanglongbing infected plants and surrounding plants Immediately treat all hosts in the immediate are with suitable chemicals (see Section 8.4) to destroy any psyllids in infested area. Hosts in the area may also be destroyed (or all leaves removed) to remove habitat for the psyllids Trace the movement of plant material to and from the infested area and control any huanglongbing infections or psyllid populations discovered Carry out surveys in the surrounding area (including gardens, native bushland, commercial and abandoned orchards) for other psyllid populations or huanglongbing infections

8.2 Control of infested/infected areas

If a tree, orchard or area is found to be infected with huanglongbing or infested with the citrus psyllid vectors the area will need to be managed to eradicate or contain the pest.

The establishment of psyllids will increase the chance of the successful establishment of huanglongbing (Halbert and Manjunath 2004) and for this reason the psyllids will need to be controlled, even in the absence of huanglongbing.

8.2.1 Control overview

Control would rely on the use of suitable pesticides (Section 8.4) to control the vectors (if present) together with the removal of infected trees to destroy the bacteria. All trees (including roots and above ground parts) in the infected/infested orchard will need to be sampled to determine the spread of the bacteria and/or vectors, and identify the source of infection.

Particular care must be taken to minimise the transfer of plant material that may allow the spread of either the bacteria or the psyllid vectors.

All equipment used on the site should be thoroughly cleaned down, with products such as a degreaser or potentially a 1% bleach solution and washed down with a pressure cleaner on the affected property. Evidence for efficacy of bleach treatments against huanglongbing is mixed and should be confirmed before recommending within a response program.

The clean down procedure should be carried out on a hard surface or preferably a designated washdown area to avoid mud being recollected from the affected site onto the machine.

8.2.2 Area wide management

In California and Florida, Area Wide Management of the psyllid has been implemented. The aim of area wide management is to encourage all growers within a defined area to implement control strategies for the pest at the same time. In Florida this has proven to be an effective tool in significantly reducing Asian citrus psyllid numbers (see: www.crec.ifas.ufl.edu/extension/chmas/index.shtml for further information). Although mostly used for the managing of the pest, this could still have a role in eradication as a way of engaging the community in controlling the pest.

8.3 Control of hosts

There are few cost effective methods for curing a tree infected by huanglongbing. Huanglongbing typically has a long latency period, especially in older trees (Gottwald 2010). The bacteria are reported to have an uneven distribution within the tree, making detection at low levels of infection difficult, and false negatives possible (Gottwald 2010). Heat therapy has been shown to be effective under controlled conditions (Hoffman *et al.* 2012),

As there is no cost-effective cure for huanglongbing infected host plants will need to be destroyed, along with any hosts identified as being at risk of being infected (e.g. trees that share common budwood sources with the infected plant(s), surrounding plants that may be root grafted to the infected plant(s), etc.).

Vectors will need to be destroyed and all host plants in the surrounding area (area to be determined by the flight ability of the psyllid and spread of plant material (see Sections 4.4 and 5.4 for further details on the dispersal ability of the psyllid vectors)) should be destroyed so that they cannot be utilised as a food source for the psyllid.

Trace forward and trace back of budwood and rootstocks will be required to identify the source of infection (especially if the psyllid vectors are absent). If the psyllid vectors are present it will be more difficult to identify the source of infection. Trace forward and trace back will guide destruction, eradication and surveillance efforts.

Skeletonisation and removal of all leaves from hosts could be considered as part of an eradication strategy as the psyllid cannot survive without host canopies. Chiyaki *et al.* (2012) outlines a model to characterize the dynamics of the vector and disease establishment.

8.4 Chemical control

Although huanglongbing is not able to be cured pesticides allow the control of the psyllid vectors that spread the disease between host plants. Therefore, in the event of an incursion of either Asiatic citrus psyllid or African citrus psyllid pesticides are likely to offer a method of control. However, before any chemical can be used in Australia it must be approved for that use by the APVMA. See the APVMA website (http://apvma.gov.au/node/611) for further information on permits.

Products included below were selected based on a review of the scientific literature, with products not registered for use on citrus in Australia removed from the list. Chemical control options change overtime and the most appropriate options will need to be selected at the time of an incursion based on the most up to date information.

8.4.1 Asiatic citrus psyllid

Management of the Asiatic citrus psyllid with pesticides will be required to obtain containment and eradication, both of the psyllid and of infection with huanglongbing. There are a number of different insecticides used in the US that provide effective control and are currently used in Australia on citrus (Table 6). Other chemicals used in the United States for the management of Asiatic citrus psyllids can be found at: www.ipm.ucdavis.edu/PMG/r107304411.html.

It will be important to strategically time these applications to kill eggs, nymphs and adults, and reduce feeding and oviposition by adults. In the US, growers are encouraged to target adult psyllids by making effective foliar applications during winter and prior to anticipated new growth during the growing season based on monitoring (Stansly *et al.*, 2009).

Chemical	Comments	Reference
Chlorpyrifos	Application to foliage of Valencia orange trees at a rate of 5 US pints/acre (5.9 L/ha) significantly reduced psyllid adults for 24 days after treatment, and significantly reduced the percentage of infested shoots and numbers of nymphs per shoot.	Qureshi and Stansly (2009)
Imidacloprid	Leaf spraying lead to 50-70 % mortality with decreased effectiveness one month after application. Imidacloprid was more effective than Thiamethoxam or Clothianidin. Trunk injections of Imidacloprid lead to 50 % psyllid mortality with effectiveness continuing for one month. Imidacloprid is applied in the US at rate of 0.56 kg/ha per year to provide good psyllid control in small citrus trees.	Ichinose <i>et al.</i> , (2010) Qureshi and Stansly (2007)
Methidathion	Application to foliage of Valencia orange trees at a rate of 1 US quart/acre (2.34 L/ha) lead to significantly fewer adults observed for more than a month after application, and significantly lower percentage of shoots infested with eggs, percentage of flush infested with nymphs and nymphal density.	Qureshi and Stansly (2009)
Spinetoram ⁶	Application to foliage of Valencia orange trees at rate of 4 fl. oz./acre (292 mL/ha) significantly reduced psyllid adult numbers, percentage of infested shoots and numbers of nymphs per shoot.	Qureshi and Stansly (2009)
Thiamethoxam	Leaf spraying resulted in 50–70 % psyllid mortality with decreased effectiveness reported one month after application. Trunk injections with Thiamethoxam lead to 50 % psyllid mortality with efficiency continuing for 1 month.	Ichinose <i>et al.</i> , (2010)

Table 6. Chemicals	used to contr	ol Asiatic citrus	psyllid	(D. citri)

⁶ Both 435 Oil (a.i mineral oil) and Induce (a.i Alkyl Aryl Polyoxylkane Ethers and Free Fatty Acids), non-ionic surfactants, appeared to increase the efficacy of Delegate WG (a.i. Spinetoram).

8.4.2 African citrus psyllid

Management of the African citrus psyllid with pesticides will be required to obtain containment and eradication, both of the psyllid and of infection with huanglongbing. There are a number of different insecticides used in South Africa that provide effective control of the pest and are also currently used on citrus in Australia (Table 7).

Chemical	Comments	Reference
Chlorpyrifos	Pyrinex 250 CS (250 g a.i/L) is used in South Africa for the control of the African citrus psyllid, when applied as a solution containing 115 ml/100 L water.	Pyrinex 250 CS
Dimethoate	Catling (1970b) showed Dimethoate to be effective at killing eggs and nymphs of the African citrus psyllid.	Catling (1970b);
	Dimethoate in the product Dimethoate EC (400 g a.i/L) is used in South Africa for the control of the African citrus psyllid.	Dimethoate EC
Imidacloprid	Kohinor 350 SC (350 g a.i/L) is used in South Africa for the control of the African citrus psyllid when applied as a soil drench at a rate of 9 ml (3.15 g a.i)/tree. All lemon fruit should be removed prior to application.	Kohinor 350 SC

8.5 Host-plant resistance

Host plant resistance is an area of interest overseas as it potentially offers a low cost way of managing pests and diseases such as huanglongbing or its vectors. However, it should be noted that resistance will only aid in the management of the pest not its eradication.

To date no commercial citrus varieties have been developed that are resistant to the huanglongbing. However, studies have shown that some varieties are less susceptible to, and supported lower populations of, the huanglongbing causing bacteria than others (Stover and McCollum 2011; Albrecht and Bowman 2011). Because of this, management of huanglongbing in countries where the disease is endemic instead focus on the use of disease free planting material, removal of vectors and the removal of infected trees, in order to keep the inoculum level as low as possible (Manjunath *et al.*, 2008).

8.6 Biological control

Biological control potentially offers a low cost method of managing the psyllid vectors of huanglongbing. However, biological control is not suitable for the eradication of the vectors, rather it is suitable for ongoing management after eradication has been deemed unfeasible.

The use of antagonistic organisms for the control of the Asiatic citrus psyllid and the African citrus psyllid has been investigated in numerous papers. A summary is provided below.

8.6.1 Asiatic citrus psyllid

Tamarixia radiata (an ectoparasitoid) and *Diaphorencyrtus aligarhensis* (an endoparasitoid) have been introduced into Florida to control the Asiatic citrus psyllid (McFarland and Hoy 2001). *Olla v-nigrum* and *Harmonia axyridis* are two North American beetles that have a greater impact on the Asiatic citrus psyllid than the introduced *T. radiata* (Michaud 2002). Michaud (2002) also reports that *Curinus coeruleus*, *Cycloneda sanguinea* and *Exochomus childreni childreni* feed on *D. citri*.

8.6.2 African citrus psyllid

Biological control of the African citrus psyllid has been demonstrated to assist in keeping the huanglongbing disease incidence low (Aubert *et al.*, 1984). *Tamarixia radiata* (syn. *Tetrastichus radiatus*) (an ectoparasitoid) and *Psyllaephagus pulvinatus* (an endoparasitoid) appear to be the main parasitoids of the African citrus psyllid (McDaniel and Moran 1972). *Tamarixia dryi* is another key species for the control of the African citrus psyllid in southern Africa (Van Den Berg and Greenland 2000).

9 Epidemiological study, surveillance and collection of samples

Information provided in Sections 9.1 and 9.2 provides a framework for the development of early detection and delimiting surveys for chewing insect pests. Sections 9.3 and 9.4 provide information on sampling and collection procedures.

9.1 Epidemiological study

The climatic conditions and the presence or absence of suitable psyllid vectors will affect the development and spread of huanglongbing. It should be remembered that the disease can take several years to develop and display symptoms, especially when older trees are infected. Gottwald (2010) provides a good overview of the epidemiology of huanglongbing.

9.1.1 Huanglongbing

Sampling of plants to determine the presence of huanglongbing within a district and beyond will be based upon the origins of the initial suspect sample(s). Factors to consider will be:

- The presence of either African citrus psyllids or Asiatic citrus psyllids. If present, pathways that spread the psyllid will need to be considered as these will also potentially spread the bacteria (if present) with infected vectors (see below).
- The source of any plant material used on the property.
- Huanglongbing is graft transmitted, therefore, other trees/orchards/nurseries that share the same mother trees, are potentially root grafted to the infected tree, or have been grown from material from the infected tree will need to be sampled.
- It takes time for the disease to reach detectable levels, generally older trees take longer to display symptoms than younger trees (Gottwald 2010).

9.1.2 Asiatic and African Citrus psyllids

Sampling of plants to determine the presence of the psyllid vectors within a district and beyond will be based upon the origins of the initial suspect sample(s). Factors to consider will be:

- The proximity of other susceptible plants to the initial infestation source.
- Machinery or vehicles that have been into the infested area or in close proximity to the source of infection.
- The movement of unprocessed fruit, which can spread the psyllids between areas (Halbert *et al.*, 2010).
- The extent of human movements into and around the infested area. A possible link to the recent importation of plant material (including budwood, nursery plants, foliage from host plants) from other regions should also be considered.
- The source of any plant material used on the property.

- The temperature and other environmental conditions, which affect the pest's reproduction rates and spread.
- The direction and strength of the prevailing wind. Psyllids can be dispersed by the wind (Hall *et al.*, 2013).
- It may take some time for psyllid populations to build to detectable levels.

9.2 Surveillance

9.2.1 Priorities

Detection and delimiting surveys are required to delimit the extent of the outbreak, ensuring areas free of the pest retain market access and appropriate quarantine zones are established.

Initial surveillance priorities include the following:

- Surveying all host growing properties and businesses (including commercial orchards, nurseries, gardens, bushland, parks etc.) in the pest quarantine area.
- Surveying all properties and businesses identified in trace-forward or trace-back analysis as being at risk.
- Surveying all host growing properties and businesses that are reliant on trade with interstate or international markets which may be sensitive to the presence of the pest.
- Surveying other host growing properties.

9.2.2 Technical information for planning surveys

When developing surveys for presence and/or distribution of the Asiatic citrus psyllid or African citrus psyllid, the following points provide the basic biological knowledge that informs the survey strategy:

- Host species in Australia are widely and irregularly dispersed and may be present within
 residential properties, commercial and abandoned orchards, nurseries (both retail and
 production), parks, retail outlets selling Citrus (commercial and native species and varieties, and
 species of *Murraya* (e.g., orange jasmine), *Bergera* (e.g., curry leaf) and *Clausena* (e.g., huangpi
 or wampee), weekend markets and roadside stalls. If native citrus occurs in the area, these must
 also be inspected.
- The likely distribution of all known hosts within a 5 km radius of the point of detections should be
 determined, confirmed and mapped. If the incursion is detected within an orchard or nursery, all
 trees within the orchard or nursery should be inspected for the presence of the disease or vector
 (using appropriate methods, e.g. visual inspections, PCR tests for the bacteria, etc.). In all other
 circumstances, host plants should be surveyed for psyllid infestations and symptoms of
 huanglongbing within a radius of 200 m of the initial point of detection (see Beattie and Barkley
 2009).
- After initial inspections the survey should resume where hosts are distributed, at 200 m intervals
 on all (e.g., in the Riverina) or part (e.g., narrow coastal regions or orchards adjacent to rivers) of
 the circumference of a circle with a 5 km radius from the point of detection. If hosts are distributed
 in a narrow coastal region, or along rivers, it is recommended that surveys also extend in 1 km
 increments outwards from the 5 km radius to 10 km (see Beattie and Barkley 2009).

- Distribution is influenced by people and equipment movements, wind intensity and direction and the proximity of adjacent plants. The psyllids are not strong flyers and dispersal over considerable distance is wind assisted or occurs via the movement of infested plants. Jumping/landing behaviour, such as when disturbed, is likely to be less than 8 m and dispersal is prompted by high populations (Beattie and Barkley 2009). Psyllids may move at least several kilometres during normal air current assisted dispersal flights and may move far greater distances during high wind events.
- *Diaphorina citri* can move on shipments of fresh, unprocessed citrus fruit (Halbert *et al.*, 2010). *Trioza erytreae* is likely to be spread in the same way as *D. citri*.
- The adult *D. citri* has a distinctive feeding stance, feeding with its head down, almost touching the leaf, and the rest of its body is raised from the surface at an almost 45-degree angle with its tail end in the air. No other endemic insect pest of citrus positions its body this way while feeding.
- The adult *T. erytreae* have a similar feeding stance to *D. citri*, feeding with the abdomen raised at a 35° angle to the plant surface (OEPP/EPPO 2005b).
- Huanglongbing infected trees have been reported to be more common on the edges (including along roads, etc.) than the interior of infected orchards. It is suggested that this is because psyllids congregate in these areas (but are also present elsewhere in the planting but at lower levels) (Gottwald 2010).
- Production areas and significant proportions of Australia may have favourable climatic conditions for the pests spread and establishment.
- Surveys must be completed as soon as possible as any delay may be detrimental to restricting further spread.
- Any psyllids collected should be sampled to determine if huanglongbing causing bacteria are present.

It is recommended that surveys to determine the extent of an incursion immediately after detection should comprise visual inspection of young flush growth for the presence of eggs, nymphs, adults and honeydew and include beating of foliage on hosts within 5 km of the point of detection using the following methods:

- visual inspections for adults on mature leaves, particularly on the underside of leaves in between flush cycles, particularly in regions with distinct winters;
- visual inspections for eggs and nymphs on flush growth from 5 mm to 50 mm long, particularly in spring, within 14 days of buds opening;
- visual inspections of young flush growth (5 mm to 50 mm long) for honeydew, particularly in spring, within 14 days of buds opening;
- beating foliage, particularly young flush growth, to dislodge adults into a pan containing a shallow amount of mineral oil;
- use of D-vac machines to suck psyllid adults from within canopies; the use of these will be limited by the number of trees to be sampled and would not be practical in urban areas, nurseries or orchards unless used to sample a large number of trees collectively to determine presence in a defined area.

 use of yellow sticky traps to trap flying adults (see Aubert and Xia 1990; Samways 1987; Samways 1990); this is limited in urban situations and orchards by the distances *D. citri* and *T. erytreae* can fly and their need to disperse from a host plant. Their use in nurseries and small urban areas would be appropriate if inspected every 7 days (Beattie and Barkley 2009).

9.2.3 Delimiting surveys in the event of an incursion

In the event of an incursion, delimiting surveys are essential to inform the decision-making process. Delimiting surveys should comprise local surveys around the area of initial detection concentrating on areas of dieback or discolouration. The normal procedure is to collect symptomatic plants and to test them to confirm the presence of the bacteria and the vector. If confirmed, plant samples taken at random from the same area should be tested to enable an estimate to be made of the pest incidence. Surrounding areas would then be surveyed. The extent of the survey beyond the initial infected/infested area should be guided by the test results from surrounding areas and the likely spread pathway (i.e. graft or vector transmission).

When establishing delimiting surveys for huanglongbing and/or its psyllid vectors the following should be considered:

- The size of the survey area (Figure 21) will depend on the size of the infected/infested area, as well as potential movement of plant material (especially budwood) during the period prior to detection. It is recommended delimiting surveys should comprise local surveys around the area of initial detection concentrating on areas of leaf discolouration, or other symptoms of the bacteria/vector.
- A high intensity of field sampling is needed for a high degree of confidence, especially as false negatives for huanglongbing can occur due to the uneven distribution of the disease within the plant (see Section 7.2 and 8.3)
- If huanglongbing is present without vectors, trace-forward or trace-back of budwood will be essential.
- All potential host species of huanglongbing (see Sections 12.1) and its vectors (if present) (see Sections 12.4 and 12.7), should be surveyed.
- In addition to inspection of possible host plants, material should be collected for diagnostic purposes (refer to Section 9.4).
- If the incursion is in a populated area, publication and distribution of information sheets and appeals for public assistance may be helpful.

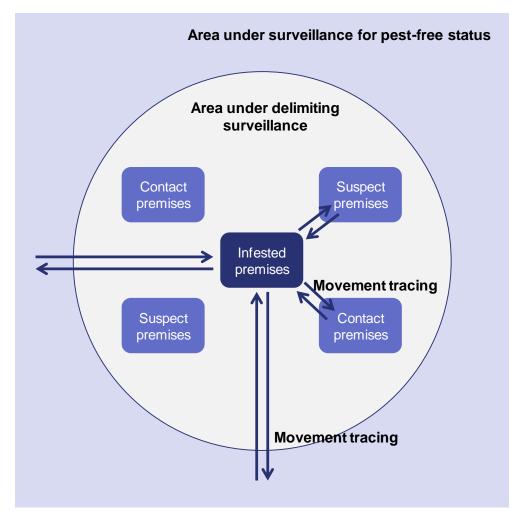


Figure 21. Diagram of a delimiting survey showing surveillance activities from the infected/infested premises

9.3 Sampling for huanglongbing and its vectors

9.3.1 Sampling for huanglongbing

Huanglongbing has a long latency period and cannot be consistently detected until symptoms are present due to the uneven distribution of the bacteria within the plant (Manjunath *et al.*, 2008). Roots are reported to be colonised before the bacteria moves to the leaves and foliar symptoms are expressed (Johnson *et al.*, 2014). It is therefore important to collect root samples, phloem samples from the trunk and symptomatic material if possible as this material is more likely to have the bacteria present at levels that can be detected using polymerase chain reaction (PCR) methods.

If present, psyllids should also be tested for the presence of huanglongbing, as Manjunath *et al.*, (2008) suggests that the bacteria may be detected in the vector before it is present at detectable levels in infected plants.

The National Diagnostic Protocol developed for the Asian strain of the disease (NDP 25) providessamplinginformation.Theprotocolisavailablehttp://plantbiosecuritydiagnostics.net.au/wordpress/wp-content/uploads/2014/09/NDP-25-Huanglongbing-Candidatus-Liberibacter-asiaticus-V1.0.pdf.

A detailed sampling protocol has also been prepared for use in the Florida, which provides details on the procedures for sampling plant material for huanglongbing (Irey, date unknown). Although this protocol was developed for North America, the information would still be useful for developing an Australia sampling protocol in the event of an incursion of huanglongbing.

9.3.2 Sampling for psyllids

Citrus psyllids should be sampled considering the points discussed in Sections 9.3.2.1 and 9.3.2.2.

9.3.2.1 Asiatic citrus psyllid

Specific colours do not consistently attract adult Asiatic citrus psyllid (some studies have found blue sticky cards to be more attractive than yellow and others show the opposite to be the case (Hall *et al.*, 2007)).

Hall *et al.*, (2007) looked at the effectiveness of different coloured sticky cards, stem tapping (stem or branch is tapped to encourage the psyllid to drop onto a board or pan held under the branch), Multi-Lure traps, and CC traps for sampling and monitoring Asiatic citrus psyllid. They found stem tapping, and sticky card traps (blue or yellow) to be the most effective. Multi-Lure traps, and CC traps have been found to be ineffective at capturing psyllids. Sweep netting has also been shown to be an effective way of detecting the pest (Arevalo *et al.*, 2011).

Nymphs and eggs are sampled for in the United States by examining flush shoots and leaves (Hall *et al.*, 2008).

A yellow sticky trap and stem tapping sampling protocol has been developed in the United States (Hall and Hentz 2010) and should be referred to when sampling for the Asiatic citrus psyllid. A recent study suggests that Lime green traps (baited with an attractant) are more attractive than yellow sticky traps (Czokajlo *et al.*, 2015).

9.3.2.2 African citrus psyllid

The African citrus psyllid is reported to be highly attracted to yellow-green colour (530 nm) (Samways 1987). Therefore, traps incorporating this colour can assist in capturing this species. Samways (1987) also describe a trap suitable for indicating the abundance of the African citrus psyllid.

Sweep netting and stem tapping techniques used for the Asiatic citrus psyllid are also likely to be suitable for the sampling of the African citrus psyllids, as it shares behavioural traits with the Asiatic citrus psyllid.

9.4 Collection and treatment of samples

Once initial samples have been received and preliminary diagnosis made, follow up samples to confirm identification of the bacterium/vector will be necessary. This will involve sampling directly from the infected/infested location and sampling over a larger area to determine the extent of the pests' distribution.

Protocols for the collection, transport and diagnosis of suspect Emergency Plant Pests (EPPs) must follow PLANTPLAN (Plant Health Australia 2014). Details are provided in the Guidelines for the

Collection of suspect Emergency Plant Pests and Transport of suspect Emergency Plant Pests PLANTPLAN available as supporting documents of (Plant Health Australia. 2014) (www.planthealthaustralia.com.au/wp-content/uploads/2014/12/Guidelines-Collection-ofsuspect-Emergency-Plant-Pests.pdf www.planthealthaustralia.com.au/wpand content/uploads/2014/12/Guidelines-Transport-of-Suspect-Emergency-Plant-Pests.pdf). Any personnel collecting samples for assessment should notify the diagnostic laboratory prior to submitting samples to ensure expertise is available to undertake the diagnosis.

The total number of samples collected at this point may run into the hundreds or even thousands. It is therefore vital that a system of sample identification is determined early in the procedure to allow for rapid sample processing and accurate recording of results. Data collected should include details such as geographical location (using GPS), host information (including age, variety, plant part affected), symptoms, level of pest prevalence, detection method, movements of plants, people and equipment on the property, climatic events (e.g. storms and prevailing wind directions) and orchard management (e.g. top-working, spray regimes, etc.).

Samples should be initially collected over a representative area of the infected/infested crop to determine the bacteria's distribution. The disease may appear as patches within the crop depending on the source of the disease.

It is important to note the distribution of bacteria/vectors in the initial crop, as this can help indicate where the bacterium/vector originated from.

It is also important that all personnel involved in crop sampling and inspections take all precautions to minimise the risk of spreading the bacteria/vector between areas by decontaminating between properties.

It should also be noted that except in exceptional circumstances (decided by State Coordination Centre (SCC) and/or Chief Plant Health Manager (CPHM)), no live insects should be sent to diagnostic laboratories.

The Chief Plant Health Manager will select the preferred laboratory. Samples will be forwarded to the nominated diagnostic laboratories for processing. All sample containers should be clearly labelled with the name, address and contact phone number of both the sending and receiving officers. In addition containers should be clearly labelled in accordance with the Guidelines for the *Collection of suspect Emergency Plant Pests* and *Transport of suspect Emergency Plant Pests* available as supporting documents of PLANTPLAN (Plant Health Australia, 2014) (www.planthealthaustralia.com.au/wp-content/uploads/2014/12/Guidelines-Collection-of-suspect-Emergency-Plant-Pests.pdf and www.planthealthaustralia.com.au/wp-content/uploads/2014/12/Guidelines-Transport-of-Suspect-Emergency-Plant-Pests.pdf). Containers should be carefully sealed to prevent loss, contamination or tampering of samples.

10 Course of action – eradication methods

Additional information is provided by the IPPC (1998b) in Guidelines for Pest Eradication Programmes. This standard describes the components of a pest eradication programme which can lead to the establishment or re-establishment of pest absence in an area. A pest eradication programme may be developed as an emergency measure to prevent establishment and/or spread of a pest following its recent entry (re-establish a pest free area) or a measure to eliminate an established pest (establish a pest free area). The eradication process involves three main activities: surveillance, containment, and treatment and/or control measures.

For a range of specifically designed procedures for the emergency response to a pest incursion and a general communication strategy refer to PLANTPLAN (Plant Health Australia 2014).

10.1 Surveillance and tracing

Detection and delimiting surveys are required to delimit the extent of the outbreak, ensuring areas free of the pest retain market access and appropriate quarantine zones are established.

Initial surveillance priorities include the following:

- Surveying all host growing properties and businesses in the pest quarantine area.
- Surveying all properties and businesses identified in trace-forward or trace-back analysis as being at risk.
- Surveying all host growing properties and businesses that are reliant on trade with interstate or international markets which may be sensitive to the presence of the pest/bacterium.
- Surveying other host growing properties and backyards.

10.1.1 Survey regions

Establish survey regions around the surveillance priorities identified above. These regions will be generated based on the zoning requirements (see Section 10.3), and prioritised based on their potential likelihood to currently have or receive an incursion of huanglongbing and/or its vectors. Surveillance activities within these regions will either allow for the area to be declared pest free and maintain market access requirements or establish the impact and spread of the incursion to allow for effective control and containment measures to be carried out. Detailed information regarding surveys for huanglongbing and its vectors have been outlined elsewhere in this plan (refer to Section 9.3).

Steps outlined in Table 8 form a basis for a survey plan. Although categorised in stages, some stages may be undertaken concurrently based on available skill sets, resources and priorities.

Phase 1	Identify properties that fall within the buffer zone around the infected/infested premise. Complete preliminary surveillance to determine ownership, property details, production dynamics and tracings information (this may be an ongoing action).		
Phase 2 .	Preliminary survey of host crops on properties in buffer zone establishing points of pest detection.		
Phase 3 •	Surveillance of an intensive nature, to support control and containment activities around points of pest detection.		
Phase 4 .	Surveillance of contact premises. A contact premise is a property containing susceptible host plants, which are known to have been in direct or indirect contact with an infected/infested premises or infected/infested plants. Contact premises may be determined through tracking movement of materials from the property that may provide a viable pathway for spread of the bacteria or its psyllid vectors. Pathways to be considered are:		
	 Items of equipment and machinery which have been shared between properties including bins, containers, vehicles and equipment. 		
	• The producer and retailer of infected/infested material, if this is suspected to be the source of the outbreak.		
	 Labour and other personnel that have moved from infected/infested, contact and suspect premises to unaffected properties (other growers, tradesmen, visitors, salesmen, crop scouts, harvesters and possibly beekeepers). 		
	 Movement of plant material (especially budwood) from controlled and restricted areas. 		
	 Storm events that could assist the dispersal of the psyllid vectors and any bacterium that they may be carrying. 		
Phase 5 .	Surveillance of orchards, gardens and public land where plants known to be hosts of the bacteria or its vectors occur.		
Phase 6	Agreed area freedom maintenance, post-control and containment.		

Table 8. Phases to be covered in a survey plan

10.2 Quarantine and movement controls

Consult PLANTPLAN (Plant Health Australia 2014) for further administrative details and procedures.

10.2.1 Quarantine priorities

- Plant material (budwood, rootstocks, etc.) at the site of infestation/infection to be subject to
 movement restrictions as such material could potentially spread the bacteria/vector to new
 areas.
- Machinery, equipment, vehicles and disposable equipment in contact with infected/infested plant material to be subject to movement restrictions (especially if the psyllid vectors are present, as machinery, etc. could spread the psyllids to new areas).

10.2.2 Movement controls

If Restricted or Quarantine Areas are practical, movement of equipment or machinery should be restricted and movement into the area only occurs by permit. The industry affected (the citrus and the nursery and garden industries) will need to be informed of the location and extent of the pest occurrence.

Movement of people, vehicles and machinery, from and to affected farms, must be controlled to ensure that infected/infested plant material (which may carry the bacteria or the vector (if present)) is not moved off-farm. This can be achieved through the following; however, specific measures must be endorsed in the Response Plan:

- Signage to indicate quarantine area and restricted movement into and within these zones.
- Fenced, barricaded or locked entry to quarantine areas.
- Movement of equipment, machinery, plant material by permit only. Therefore, all non-essential operations in the area or on the property should cease.
- Where no dwellings are located within these areas, strong movement controls should be enforced.
- Where dwellings and places of business are included within the Restricted and Control Areas movement restrictions are more difficult to enforce, however, limitation of contact with infested plants should be enforced.
- Residents should be advised on measures (e.g. minimise movement of plant material etc.) to minimise the inadvertent transport of the pest from the infected/infested area to unaffected areas.
- Plant material or plant products must not be removed from the site unless part of an approved disposal procedure.
- All machinery and equipment should be thoroughly cleaned down with a high pressure cleaner or scrubbed with products such as a farm degreaser or a 1% bleach (available chlorine) solution, prior to leaving the affected area.

10.3 Zoning

The size of each quarantine area will be determined by a number of factors, including the location of the incursion, biology of the pest, climatic conditions and the proximity of the infected/infested property to other infected/infested properties. This will be agreed by the National Management Group during the production of the Response Plan. Further information on quarantine zones in an Emergency Plant Pest (EPP) incursion can be found in Section 4.1.4 of PLANTPLAN (Plant Health Australia 2014). These zones are outlined below and in Figure 22.

10.3.1 Establishing Quarantine Zones

Delimiting surveillance will inform the establishment of quarantine zones and identify the Restricted Area(s) (RA), Control Area (CA) and Pest Free Area (PFA). The size of each quarantine zone will be determined by a number of factors including location of the incursion, climatic conditions, the biology of the bacteria/vector and proximity of an Infected Premises (IP) to other IPs.

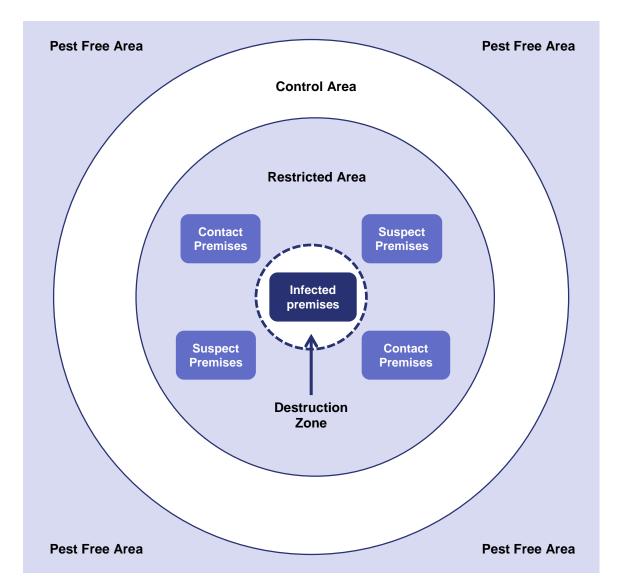


Figure 22. Schematic diagram of quarantine zones used during an EPP incursion (not drawn to scale)

10.3.2 Destruction Zone

The size of the Destruction Zone (i.e. zone in which the pest and all host material is destroyed) will depend on, distribution of the pest (as determined by delimiting surveys), and the ability of the pest to spread.

All host plants should be destroyed after the level of infection has been established. The delimiting survey will determine whether or not neighbouring hosts are infected/infested and need to be destroyed. If spread is likely to have occurred prior to detection, the Destruction Zone may include contiguous areas that have been in contact with, or are associated with the same management practices as, the infected/infested area. Particular care needs to be taken to ensure that plant material is not moved into surrounding areas.

10.3.3 Restricted Area

Data collected from surveys and tracing (trace back and trace forward) will be used to define the RA, which comprises all properties where the pest has been confirmed (Infected Premises or IP), properties which have come into direct or indirect contact with an IP or infected/infested plants (Contact Premises or CP) and properties which may have been exposed to the pest (Suspect Premises or SP). The RA will be subject to intense surveillance and movement control, with movement out of the RA to be prohibited and movement into the RA to occur by permit only.

10.3.4 Control Area

A CA is established around a RA to control the movement of susceptible hosts and other regulated materials until the extent of the incursion is determined. There may be multiple RAs within one CA. When the extent of the EPP Incident has been confidently defined, the RA and CA boundaries and movement controls may need to be modified, and where possible reduced in size commensurate with appropriate controls.

Additional zones can be utilised as required for operational purposes.

10.3.5 Pest free Area guidelines

Determination of PFAs should be completed in accordance with the International Standards for Phytosanitary Measures (ISPMs) 8 and 10 (IPPC 1998a, 1999).

Additional information is provided by the IPPC (1995) in Requirements for the Establishment of Pest Free Areas. This standard describes the requirements for the establishment and use of PFAs as a risk management option for phytosanitary certification of plants and plant products. Establishment and maintenance of a PFA can vary according to the biology of the pest, pest survival potential, means of dispersal, availability of host plants, restrictions on movement of produce, as well as PFA characteristics (size, degree of isolation and ecological conditions).

In the event of an incursion, specific guidelines for surveys and monitoring will be provided by the Consultative Committee on Emergency Plant Pests (CCEPP). General points to consider are:

- Design of a statistical delimiting survey (see Section 9 for points to consider).
- Plant sampling should be based on the rates required to give an appropriate level of confidence and taken at random within the orchard/area.
- Preliminary diagnosis can be based on plant symptoms and vector morphology.
- The results are confirmed by diagnosis in another recognised laboratory or by another diagnostician.
- Surveys should also consider alternative host plants (see Sections 4.5, 5.5 and 7.5) and not be limited to commercial citrus.
- Information (including absence of the bacteria/vector) should be recorded.

10.4 Destruction strategy

10.4.1 Priorities

- Confirm the presence of huanglongbing/psyllid vectors
- Limit movement or people and prevent movement of vehicles and equipment through affected areas.
- Stop the movement of any propagation material that could contain/spread the bacteria.
- Stop the movement of any plant material or machinery that could be carrying the psyllids or their eggs from the infected area.
- Determine the strategy for the destruction of psyllid hosts.
- Determine the strategy for the destruction of huanglongbing infected host material.
- Determine the extent of the infestation through survey and plant material trace back and trace forward. If huanglongbing is present without the vector, surveillance will be simplified as budwood material would likely be the source of the disease.

10.4.2 Destruction protocols

- No plant material should be removed from the infected/infested area unless part of the disposal procedure.
- Disposable equipment, infested/infected plant material, etc. should be disposed of by autoclaving, high temperature incineration or deep burial.
- Any equipment removed from the site for disposal should be double-bagged (where practical).
- All vehicles and farm machinery that enter the infested/infected field should be thoroughly washed, preferably using a detergent, farm degreaser or a 1% (available chlorine) bleach solution, to stop the accidental movement of psyllid vectors (if present).
- If huanglongbing is present, the infected and surrounding trees should be treated with an appropriate pesticide (see Section 8.4) to control any psyllids that may be present, cut down, the waste disposed of (see Section 10.4.4) then the stump treated with an appropriate herbicide. If psyllids are present without the disease than the area should be treated with an appropriate pesticide (see Section 8.4) to control the psyllids. Host plants should then be destroyed to remove possible food and breeding sites for the psyllid.

10.4.3 Decontamination protocols

If decontamination procedures are required, machinery, equipment and vehicles in contact with infected/infested plant material or present within the Quarantine Area, should be washed to remove soil and plant material using high pressure water or scrubbing with products such as a farm degreaser or a 1% bleach solution in a designated wash down area.

General guidelines for decontamination and clean-up are as follows:

• Refer to the Guideline for the *Disinfection and decontamination guidelines* (available as a supporting document of PLANTPLAN (Plant Health Australia, 2014)

(www.planthealthaustralia.com.au/wp-content/uploads/2015/12/Guidelines-Disinfection-and-decontamination.pdf)) for further information.

- Keep traffic out of affected area and minimise it in adjacent areas.
- Adopt best-practice property hygiene procedures to retard the spread of the pest between fields and adjacent properties.
- Machinery, equipment and vehicles in contact with infected/infested plant material, should be washed using high pressure water or scrubbing with products such as a degreaser or a bleach solution in a designated wash down area
- Only recommended materials are to be used when conducting decontamination procedures, and should be applied according to the product label.
- Infected/infested plant material should be disposed of by autoclaving, high temperature (enclosed) incineration or deep burial.

10.4.4 Plants, by-products and waste processing

- If the psyllid vectors are present and material moved from the site will need to be covered or contained to ensure that psyllids are not spread to new areas.
- Any infected/infested plant material removed from the site should be destroyed by (enclosed) high temperature incineration, autoclaving or deep burial.
- Infected/infested areas should remain free of susceptible host plants (including weeds, alternative hosts and volunteer plants) until the area has been shown to be free from the pest.

10.5 Post-eradication surveillance

The period of pest freedom sufficient to indicate that eradication of the pest has been achieved will be determined by a number of factors, including climatic conditions, the control measures applied and the pest(s) biology.

Specific methods to confirm the eradication of huanglongbing include:

- Establishment of sentinel plants at the site of infection (to detect the bacteria and/or vectors).
- Maintain good sanitation and hygiene practices throughout the year.
- Monitoring of plants for signs of huanglongbing.
- If detected, samples are to be collected and stored and plants destroyed.
- Non-host crops should be grown on the site and any self-sown/regrowth of host plants destroyed.
- Surveys should be undertaken for a minimum of 3 years after eradication has been achieved (or as endorsed by a CCEPP).

Specific methods to confirm the eradication of psyllid vectors include:

- Establishment of sentinel plants and sticky traps at the site of infection (to detect vectors).
- Maintain good sanitation and hygiene practices throughout the year.
- Monitoring of plants for signs of the psyllid.

- If detected, samples are to be collected and stored and plants destroyed (so they cannot act as a potential host of the bacteria, which may be present with the psyllid).
- Non-host crops should be grown on the site and any self-sown/regrowth of host plants destroyed.
- Surveys should be undertaken for a minimum of 3 years after eradication has been achieved (or as endorsed by a CCEPP).

11 Technical debrief and analysis for stand down

The aim of the Stand Down Phase is to provide guidance for moving from emergency response arrangements to normal business.

The Stand Down Phase is activated by one of the following:

- The Investigation and Alert Phase fails to confirm the presence of an EPP.
- The eradication of a confirmed EPP is determined by the NMG (upon advice by the CCEPP) not to be technically feasible and cost beneficial (refer section 4.1.7 of PLANTPLAN).
- Following implementation of a Response Plan, the NMG formally declares that the Incident has ended as the EPP has been successfully eradicated or eradication is no longer considered feasible or eradication was unsuccessful (refer section 4.2.4 of PLANTPLAN).

Refer to section 4.3 of PLANTPLAN (Plant Health Australia 2014) for further details.

12 Appendices

12.1 Appendix 1: Recorded hosts of Asiatic citrus psyllid

Table 9. Known hosts of Asiatic citrus psyllid (Diaphorina citri). Adapted from Beattie and Barkley (2009)⁷; Westbrook et al., (2011). Note: citrus nomenclature based on Mabberley (1997; 1998; 2004; 2008); Scott et al., (2000); Samuel et al., (2001); and Bayer et al., (2009) with synonyms of Citrus listed in Appendix 10.

Species or hybrid	Common name	Species or hybrid	Common name
Aegle marmelos	Bael	Citrus x aurantium	Sweet orange (navel and Valencia), sour orange, grapefruit, chinotto
Aeglopsis chevalieri		Citrus x floridana	Limequat
Afraegle gabonensis	Gabon powder flask fruit	Citrus x insitorum	Citrange
Afraegle paniculata	Nigerian powder flask fruit	Citrus x limon	Lemon
Atalantia buxifolia syn. Severinia buxifolia	Chinese box orange	Citrus x microcarpa	Calamondin, calamandarin, calamansi
Atalantia monophylla	Indian atalantia	Citrus x oliveri	Faustrimedin, Australian sunrise lime
Balsamocitrus dawei	Uganda powder flask fruit	Citrus x taitensis	Rough lemon, Mandarin lime, rough lemon, Rangpur
Bergera koenigii syn. Murraya euchrestifolia, M. koenigii	Curry leaf/tree	Citrus x virgata	Sydney hybrid
Casimiroa edulis	White sapote	Clausena anisumolens	Kayumanis
Citropsis articulata syn. Citropsis schweinfurthii	West African cherry orange	Clausena excavata	Pink wampee
Citropsis gilletiana	Gillet's cherry orange	Clausena harmandiana	
Citrus amblycarpa	Nasnaran	Clausena indica	
Citrus australasica	Australian finger lime	Clausena lansium	Wampee
Citrus australis	Australian round lime, dooja	Glycosmis pentaphylla	Orangeberry, ginberry
Citrus cavaleriei (Syn. Citrus ichangensis)	Ichang (Yichang) papeda	Limonia acidissima	Indian wood apple, elephant apple, wood apple

⁷ See Beattie and Barkley (2009) for specific references cited within.

Species or hybrid	Common name	Species or hybrid	Common name
Citrus glauca	Australian desert lime	Merrillia caloxylon	Malay lemon
Citrus glauca x Citrus reticulata (Eremocitrus hybrid)		Murraya exotica ⁸ , Murraya paniculata	Orange jasmine, orange jessamine, mock orange
Citrus halimii	Mountain citron, sultan Iemon	Murraya paniculata var. ovatifoliolata ⁹	
Citrus hystrix	Kaffir lime, leech lime, Mauritius papeda	Naringi crenulata	Hesperethusa
Citrus inodora	Russell River lime, large leaf Australian wild lime	Pamburus missionis syn. Atalantia missionis	
Citrus japonica	Kumquat	Swinglea glutinosa	Tabog
Citrus latipes	Khasi papeda	Ravenia spectabilis syn. Lemonia spectabilis	Limonia, pink ravenia
Citrus longispina	Talamisan, winged lime	Tetradium ruticarpum syn. Evodia rutaecarpa	Evodia
Citrus maxima	Pomelo, pummelo	Toddalia asiatica	Orange-climber, forest pepper
Citrus medica	Citron	Triphasia trifolia	Limeberry, triphasia
Citrus reticulata	Mandarin	Vepris lanceolata	White ironwood
Citrus trifoliata	Trifoliate orange	Zanthoxylum ailanthoides	Japanese prickly ash
Citrus wintersii	Brown River finger lime	Zanthoxylum fagara	Lime prickly-ash
Citrus x aurantiifolia	Limes e.g., sweet lime, key/Mexican lime		

⁸ The status of this species is complex. Some reports consider *M. exotica* and *M. paniculata* to be synonyms; while others have concluded they are separate species. Hence there is some controversy over whether the "orange jasmine" commonly found as a host for *D. citri* in Florida and Brazil is *M. paniculata* or *M. exotica*, because the two names at times have been used interchangeably. Regardless of this, it is known that the common cultivated species grown in parks and home gardens is a host as well as *Murraya paniculata* var. *ovatifoliolata* that grows naturally in north and north-eastern Australia are hosts of the psyllid. ⁹ Large and small leaflet forms grow naturally in north and north-eastern Australia.

12.2 Appendix 2: Geographical distribution of Asiatic citrus psyllid

Continent	Country	Continent	Country
Asia	Afghanistan	Africa	Mauritius
	Bangladesh		Reunion
	Bhutan	North America	Mexico
	Cambodia		United States ¹⁰
	China	Central America and Caribbean	Antigua and Barbuda
	East Timor		Bahamas
	India		Barbados
	Indonesia		Belize
	Iran		Cayman Islands
	Israel		Costa Rica
	Japan		Cuba
	Laos		Dominica
	Malaysia		Dominican Republic
	Maldives		Guadaloupe
	Myanmar		Haiti
	Nepal		Jamaica
	Oman		Puerto Rico
	Pakistan		United States Virgin islands
	Philippines	South America	Argentina
	Saudi Arabia		Brazil
	Singapore		Paraguay
	Sri Lanka		Uruguay
	Taiwan		Venezuela
	Thailand	Oceania	American Samoa
	United Arab Emirates		Guam
	Vietnam		Northern Mariana Islands
	Yemen		Papua New Guinea

Table 10. Geographic distribution of Asiatic citrus psyllid (D. citri). Adapted from CABI 2014a.

¹⁰ Present in Alabama, Arizona, California, Florida, Georgia, Hawaii, Louisiana, Mississippi, South Carolina and Texas. Refer to **www.hungrypests.com/the-spread/index.php?pest=acp**.

12.3 Appendix 3: Pest risk analysis – Asiatic citrus psyllid

Potential or impact	Rating
Entry potential	HIGH
Establishment potential	HIGH
Spread potential	HIGH
Economic impact	HIGH - EXTREME
Overall risk	HIGH - EXTREME

12.3.1 Entry potential

Rating: HIGH

Diaphorina citri could enter Australia via the movement of infested host plant material containing viable egg masses or nymphs. This may occur through legal importation of infested material that has been inadequately treated and inspected, or illegal importation of plants, leaves, fruit and propagating material. Legal importation of commercially packed fruit is considered to pose a low risk.

Passive transport of adult psyllids, which are strongly attracted to light, could also occur via commercial and military aircraft.

The psyllid is present close to Australia, in Papua New Guinea, Indonesia and East Timor and may enter via the Torres Strait through people carrying host material.

Winds created by tropical storms and cyclones may also lead to the entry of psyllids into northern Australia.

From this information the entry potential has been rated as High.

12.3.2 Establishment potential

Rating: HIGH

Given that *D. citri* has a history of establishment overseas, a high reproductive rate, long-lived adults, the presence of alternative hosts and the fact they live in environmental conditions similar to those found in Australia, the establishment potential has been rated as **High**.

12.3.3 Spread potential

Rating: HIGH

D. citri can survive up to 10 days on detached stems and up to 20-30 days on detached stems with fruit (Hall and McCollum 2011). It can be distributed by wind (Hall *et al.*, 2013), or by hitchhiking on citrus propagation material, ornamentals such as orange jasmine (*Murraya*) and food plants such as curry leaf (*Bergera koenigii*) (Halbert *et al.*, 2010). It therefore has a **High** risk of spread.

12.3.4 Economic impact

Rating: HIGH - EXTREME

Heavy infestations are reported to lead to flower and fruitlet drop, and young shoots may be twisted and growth stunted. Leaves may become severely curled and covered in honeydew and sooty mould (OEPP/EPPO 2005a). Abscission of leaves, death of the apical meristem and complete abscission of the terminal have also been reported (Michaud, 2004). Based on these observations, the economic impact of *D. citri* is considered to be **High**. If *D. citri* enters Australia, there is a significant chance it will be vectoring huanglongbing, and if this is the case, the economic impact rating is **Extreme** (refer to risk rating for huanglongbing).

12.3.5 Overall risk

Rating: HIGH - EXTREME

Based on the individual ratings above, the combined overall risk of Asiatic citrus psyllid is considered to be **High** (without huanglongbing) or **Extreme** (with huanglongbing).

12.4 Appendix 4: Recorded hosts of African citrus psyllid

Table 11. Known hosts of African citrus psyllid (Trioza erytreae). Adapted from Beattie and Barkley (2009). Note: citrus nomenclature based on Mabberley (1997; 1998; 2004; 2008); Scott et al., (2000); Samuel et al., (2001); and Bayer et al., (2009) with synonyms of Citrus listed in Appendix 10.

Species or hybrid	Common name	Species or hybrid	Common name
Calodendrum capense	Cape chestnut	Citrus x aurantium	Orange, grapefruit, tangelo
Citrus australasica	Australian finger lime	Citrus x limon	Lemon
Citrus japonica	Kumquat	Clausena anisata	False horsewood
Citrus maxima	Pomelo	Murraya paniculata (syn. M. exotica)	Orange jasmine
Citrus medica	Citron	Toddalia asiatica	Orange climber, forest pepper
Citrus reticulata	Mandarin	Triphasia trifolia	
Citrus trifoliata (syn. Poncirus trifoliata)	Trifoliate orange	Vepris lanceolata	White ironwood
Citrus x aurantiifolia	Limes e.g., sweet lime, key/Mexican lime	Zanthoxylum capense	Small knobwood

12.5 Appendix 5: Geographical distribution of African citrus psyllid

Continent	Country	Continent	Country
Asia	Saudi Arabia		Reunion
	Yemen		Rwanda
Africa	Angola		Saint Helena
	Cameroon		Sao Tome and Principe
	Canary Islands		South Africa
	Comoros		Sudan
	Congo Democratic Republic		Swaziland
	Eritrea		Tanzania
	Ethiopia		Uganda
	Kenya		Zambia
	Madagascar		Zimbabwe
	Malawi	Europe	Portugal
	Mauritius		Spain

Table 12. Geographic distribution of African citrus psyllid (Trioza erytreae). Adapted from CABI 2014b.

12.6 Appendix 6: Pest risk analysis – African citrus psyllid

Potential or impact	Rating
Entry potential	MEDIUM
Establishment potential	HIGH
Spread potential	HIGH
Economic impact	HIGH - EXTREME
Overall risk	HIGH - EXTREME

12.6.1 Entry potential

Rating: MEDIUM

Trioza erytreae could enter Australia via the movement of infested host plant material containing viable egg masses or nymphs. This may occur through legal importation of infested material that has been inadequately treated and inspected, or illegal importation of plants, leaves, fruit and propagating material. Legal importation of commercially packed fruit is considered to pose a low risk.

The psyllid is currently confined to Africa and the Middle East, meaning natural dispersal is unlikely.

From this information the entry potential has been rated as Medium.

12.6.2 Establishment potential

Rating: HIGH

The African citrus psyllid is less tolerant of high temperatures and dry conditions than the Asiatic citrus psyllid. The African citrus psyllid is currently only found in Africa, surrounding islands, ,parts of the Middle East, and the Iberian peninsula in Europe. As it has spread to off shore islands and the Middle East it appears that this species can adapt to new areas if suitable hosts and climatic conditions are available.

The African citrus psyllid prefers cool humid conditions and high mortality has been reported at a temperature of 32°C or higher (Catling and Green 1972). This suggests that the climate in some of the southern citrus growing regions of Australia would be more suitable for the establishment of the African citrus psyllid than the subtropical and tropical regions.

From this information the establishment potential has been rated as High.

12.6.3 Spread potential

Rating: HIGH

Adult *T. erytreae* have been shown to survive for up to 85 hours (~3.5 days) in the absence of host plants (Van Den Berg and Deacon 1988). This means that adults could survive a flight from infested countries to Australia. The movement of nursery material (budwood, trees, seedlings, and rootstock) could also allow the dispersal of adults, nymphs and eggs over long distances.

Over shorter distances the adult psyllids are capable of flight and experiments have shown that they can fly 1.5 km if required (Van Den Berg and Deacon 1988), but generally do not move large distances from a suitable habitat (Catling 1973).

It therefore has a **High** risk of spread.

12.6.4 Economic impact

Rating: HIGH - EXTREME

Infestations of African citrus psyllids cause leaf distortion and chlorosis (OEPP/EPPO 2005b). Based on these observations, the economic impact of *T. erytreae* is considered to be **High**. If *T. erytreae* enters Australia, there is a chance it will be vectoring huanglongbing, and if this is the case, the economic impact rating is **Extreme** (refer to risk rating for huanglongbing).

12.6.5 Overall risk

Rating: HIGH - EXTREME

Based on the individual ratings above, the combined overall risk of Asiatic citrus psyllid is considered to be **High** (without huanglongbing) or **Extreme** (with huanglongbing).

12.7 Appendix 7: Recorded hosts of huanglongbing

Table 13. Known hosts of huanglongbing. Adapted from Beattie and Barkley (2009). Note: citrus nomenclature based on Mabberley (1997; 1998; 2004; 2008); Scott et al., (2000); Samuel et al., (2001); and Bayer et al., (2009) with synonyms of Citrus listed in Appendix 10.

Species or hybrid	Common name
Aeglopsis chevalieri	Chevalier's aeglopsis
Atalantia buxifolia	Chinese box orange
Balsamocitrus dawei	Uganda powder-flask-fruit
Bergera koenigii	Curry leaf/tree
Calodendrum capense ¹¹	Cape chestnut
Catharanthus roseus	Periwinkle
Citrus amblycarpa	Nasnaran
Citrus australasica	Australian finger lime
Citrus cavaleriei	Ichang (Yichang) papeda
Citrus hystrix	Kaffir lime, leech lime, Mauritius papeda
Citrus japonica	Kumquat
Citrus maxima	Pomelo
Citrus medica	Citron
Citrus reticulata	Mandarin
Citrus trifoliata	Trifoliate orange
Citrus x aurantiifolia	Limes e.g., sweet lime, key/Mexican lime, West Indian lime
Citrus x aurantium	Orange, grapefruit, tangelo
Citrus x insitorum	Citrange
Citrus x junos	Yuzu
Citrus x latifolia	Tahitian lime
Citrus x limon	Lemon
Citrus x microcarpa	Calamondin, calamandarin, calamansi
Citrus x taitensis	Rough lemon (referred to as 'bush lemons in Australia', 'lemandarins' of Swingle, mandarin lime, Rangpur lime)
Clausena anisata	False horsewood
Clausena indica	
Clausena lansium	Wampee

¹¹ Host of Ca. L. africanus subsp. capense. This Ca. L sub-species is not associated with huanglongbing in commercial citrus.

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Species or hybrid	Common name
Cuscuta australis	Dodder
Cuscuta campestris	Dodder
Limonia acidissima	Indian wood apple, elephant apple
Murraya exotica	Orange jasmine
Nicotiana tabacum	Торассо
Pamburus missionis	
Solanum lycopersicum	Tomato
Swinglea glutinosa	Tabog
Triphasia trifolia	Limeberry
Vepris lanceolata	White ironwood

12.8 Appendix 8: Geographical distribution of huanglongbing

Country	Continent/region	Reference
<i>'Candidatus</i> Liberibacter africanus' (African strain)		
Burundi	Africa	Batool et al., (2007); Aubert et al., (1988)
Cameroon	Africa	Batool et al., (2007); Aubert et al., (1988)
Central African Republic	Africa	Batool <i>et al.</i> , (2007)
Comoros	Africa	Batool <i>et al.</i> , (2007)
Ethiopia	Africa	Batool et al., (2007); Aubert et al., (1988)
Kenya	Africa	Batool et al., (2007); Bove (2006)
Madagascar	Africa	Batool et al., (2007); Bove (2006)
Malawi	Africa	Batool et al., (2007); Aubert et al., (1988)
Mauritius	Africa (Indian ocean)	Batool et al., (2007); Garnier and Bové (1996)
Reunion	Africa (Indian ocean)	Batool et al., (2007); Garnier and Bové (1996)
Rwanda	Africa	Batool et al., (2007); Aubert et al., (1988)
Saint Helena	Africa (Atlantic Ocean)	Van Den Berg and Greenland (2000)
Somalia	Africa	Batool <i>et al.</i> , (2007)
South Africa	Africa	Batool et al., (2007); Garnier and Bové (1996)
Swaziland	Africa	Batool et al., (2007); Catling and Atkinson (1974)
Tanzania	Africa	Batool <i>et al.</i> , (2007); Swai (1988)
Yemen	Asia	Bové and Garnier (1984); Batool et al., 2007
Zimbabwe	Africa	Batool et al., (2007); Garnier and Bové (1996)
<i>'Candidatus</i> Liberibacter americanus' (American strain)		
Argentina	South America	Ramallo <i>et al.</i> , (2008)
Brazil	South America	Teixeira <i>et al.</i> , (2005)
'Candidatus Liberibacter asiatic	us' (Asiatic strain)	
Argentina	South America	Ramallo <i>et al.</i> , 2008
Bangladesh	Asia	Catling <i>et al.</i> , (1978)

Table 14. Geographic distribution of huanglongbing

PLANT HEALTH AUSTRALIA | Contingency Plan for Huanglongbing and its vectors

Country	Continent/region	Reference
Belize	Central America	Manjuath <i>et al.</i> , (2010); Matos <i>et al.</i> , (2013)
Brazil	South America	Coletta-Filho <i>et al.</i> , (2004)
Bhutan	Asia	Doe <i>et al.</i> , (2003)
Cambodia	Asia	Garnier and Bové (1996); Garnier and Bove (2000)
China	Asia	Garnier and Bové (1996)
Costa Rica	Central America	Matos <i>et al.</i> , (2013)
Cuba	Central America	Martinez et al., (2009); Matos et al., (2013)
Dominican Republic	Central America	Matos et al., (2009); Matos et al., (2013)
East Timor	Australasia	Weinert <i>et al.</i> , (2004)
Ethiopia	Africa	Saponari <i>et al.</i> , 2010
Guatemala	Central America	Matos <i>et al.</i> , (2013)
Honduras	Central America	Matos <i>et al.</i> , (2013)
India	Asia	Garnier and Bové (1996)
Indonesia	Asia	Garnier and Bové (1996)
Iran	Asia	Faghihi <i>et al.</i> , (2009)
French West Indies	Central America	Cellier <i>et al.</i> , (2014)
Guadeloupe	Central America	Cellier <i>et al.</i> , (2014)
Jamaica	Central America	Matos <i>et al.</i> , (2013)
Japan	Asia	Miyakawa and Tsuno 1986)
Laos	Asia	Garnier and Bove (2000)
Malaysia	Asia	Garnier and Bové (1996)
Martinique	Central America	Cellier <i>et al.</i> , (2014)
Mauritius	Africa (Indian ocean)	Garnier and Bové (1996)
Mexico	North America	Robles-González (2013); Matos <i>et al.</i> , (2013)
Myanmar	Asia	Garnier and Bove (2000)
Nicaragua	Central America	Matos <i>et al.</i> , (2013)
Nepal	Asia	Garnier and Bové (1996)

PLANT HEALTH AUSTRALIA | Contingency Plan for Huanglongbing and its vectors

Country	Continent/region	Reference
Pakistan	Asia	Chohan <i>et al.</i> , (2007)
Papua New Guinea	Australasia	Davis <i>et al.</i> , (2005); Weinert <i>et al.</i> , (2004)
Philippines	Asia	Garnier and Bové (1996)
Puerto Rico	Central America	Matos <i>et al.</i> , (2013)
Reunion	Africa (Indian ocean)	Garnier and Bové (1996)
Saudi Arabia	Asia	Bové and Garnier (1984)
Sri Lanka	Asia	Garnier and Bové (1996)
Thailand	Asia	Garnier and Bové (1996)
Taiwan	Asia	Garnier and Bové (1996)
Vietnam	Asia	Garnier and Bové (1996)
Virgin Islands (United States)	Central America	NAPPO (2010)
United States of America	North America	Gottwald (2010)

12.9 Appendix 9: Pest risk analysis – huanglongbing

Potential or impact	Rating
Entry potential	HIGH
Establishment potential	HIGH
Spread potential	HIGH
Economic impact	EXTREME
Overall risk	EXTREME

12.9.1 Entry potential

Rating: HIGH

Huanglongbing could enter either on infected budwood or infected vectors (adult, fourth and fifth instar psyllids can carry the bacteria (Xu *et al.*, 1988; Gottwald *et al.*, 2007)). Seeds are not thought to transmit the bacteria (Hoffmann *et al.*, 2013; Albrecht and Bowman 2009).

Budwood can potentially enter via illegal pathways, while infected psyllids could enter via various means as discussed in Sections 12.3.1 and 12.6.1.

From this information the entry potential has been rated as High.

12.9.2 Establishment potential

Rating: HIGH

Given that huanglongbing has a history of establishment overseas, the presence of suitable host plants (in the natural, urban and rural environments) and the fact the bacteria has established in areas with similar environmental conditions to those found in Australia, the establishment potential has been rated as **High**.

12.9.3 Spread potential

Rating: HIGH

Huanglongbing is spread via the movement of infected plants, budwood and vectors. It is likely that the movement of infected plants will allow the long distance spread of the bacteria to new areas. If the vector is also present, the bacterium could be rapidly spread between plants.

For these reasons huanglongbing is thought to have a **High** risk of spread.

12.9.4 Economic impact

Rating: EXTREME

Huanglongbing causes a range of symptoms including bitter tasting and deformed fruit (i.e. not suitable for the fresh market). Infections ultimately result in plant death and there is no cure for the disease. In countries where the disease is endemic trees may only live for 5-8 years and never bear saleable fruit (Halbert and Manjunath 2004). For these reasons huanglongbing is considered to pose an **Extreme** economic impact to the citrus industry.

12.9.5 Overall risk

Rating: EXTREME

Based on the individual ratings above, the combined overall risk of huanglongbing is considered to be **Extreme**.

12.10 Appendix 10: Citrus nomenclature

Table 15. Citrus species and common hybrids (adapted from Beattie and Barkley, 2009 in which classification was based on Mabberley 1997; 1998; 2004; 2008; Scott et al., 2000; Samuel et al., 2001; and Bayer et al., 2009)

Scientific name	Common name
Citrus species	
<i>Citrus amblycarpa</i> (Hassk.) Ochse (a possible hybrid)	Nasnaran, nasnaran mandarin, 'Celebes' papeda
Citrus australasica F. Muell. (syn. Microcitrus australasica)	Australian finger lime
Citrus australis (Mudie) Planch. (syn. Microcitrus australis)	Australian round lime
Citrus cavaleriei H. Léveillé ex Cavalerie (C. ichangensis Swingle, C. latipes)	Ichang (Yichang) papeda/lime/lemon
Citrus fragrans (Montr.)	Fragrant oxanthera
Citrus garrawayi F. M. Bailey	Mount White lime, Garraway's Australian wild lime
<i>Citrus glauca</i> (Lindley) Burkill (syn. <i>Eremocitrus glauca</i>)	Australian desert lime
Citrus gracilis Mabb.	Humpty Doo lime
Citrus halimii BC Stone	Sultan lemon
<i>Citrus hystrix</i> DC. (<i>C. macroptera</i> Montrouz.)	Leech lime, kaffir lime, Mauritius papeda
Citrus inodora F. M. Bailey (syn. <i>Microcitrus inodora</i>)	Russell River lime, large leaf Australian wild lime
Citrus japonica Thunb. (syn. Fortunella japonica, F. crassifolia, F. hindsii, F. margarita, F. polyandra)	Kumquat
Citrus maxima (Burm.) Merr. (syn. Citrus decumana, C. grandis, C. obovoidea)	Pomelo (pummelo); cultivars include Chandler
Citrus medica L.	Citron; cultivars include Etrog, Buddha's hand/Fingered citron
Citrus neocaledonica Guill.	False orange, large-leaf oxanthera
Citrus oxanthera Beauvisage	Orange flower oxanthera
Citrus polyandra Tanaka (syn. Clymenia)	
Citrus reticulata Blanco (syn. C. nobilis, C. deliciosa, C. depressa, C. sunki)	Mandarin, tangerine, clementine , satsuma: includes <i>C</i> . x <i>suhuiensis</i> Hort. ex Tanaka, known as Canton mandarin
Citrus trifoliata L. (syn. Poncirus trifoliata)	Trifoliate orange
Citrus undulata Guill.	
Citrus warburgiana F. M. Bailey	Milne Bay lime, New Guinea wild lime

Scientific name	Common name
Citrus wintersii Mabb. (syn. <i>Microcitrus papuana</i> H. F. Winters)	Brown River finger lime
Common hybrids within the <i>Citrus</i> genus	
Citrus x aurantiifolia (Christm.) Swingle (syn. Citrus limettoides, C. pennivesiculata)	Lime; cultivars include Mexican/Key lime
Citrus x aurantium L. (syn. C. aurantium L. and C. sinensis (L.) Osbeck, C. maxima var. racemosa, C. sinensis, Citrus x paradisi)	Sour, sweet, Valencia and navel oranges; grapefruit; tangor; tangelo
Citrus x floridana (syn. Citrofortunella floridana)	Limequat
Citrus x indica Tanaka	Indian wild orange
<i>Citrus x insitorum</i> Mabb. (<i>x Citroncirus webberi</i> J. Ingram and H. E. Moore)	Citrange
<i>Citrus x latifolia</i> (Yu. Tanaka) Tanaka	Tahitian lime
Citrus x limon (L.) Osbeck (syn. Citrus assamensis, C. limonia, C. medica var. acida, C. medica var. limetta, C. medica var. limonum, C. meyeri, C. volkameriana)	Lemon
Citrus x macrophylla Wester	Alemow
Citrus x microcarpa Bunge (syn. Citrus madurensis)	Calamondin, calamandarin, calamansi
Citrus x oliveri (hybrid of Citrus australasica and C. x microcarpa)	Faustrimedin, Australian sunrise lime
<i>Citrus x taitensis</i> Risso (syn <i>C. x jambhiri</i> Lush., <i>C. x limonia</i> Osbeck)	Rough lemon (referred to as 'bush lemons in Australia', 'lemandarins' of Swingle, mandarin lime, Rangpur lime)
Citrus x virgata Mabb. (syn. Microcitrus sp., Microcitrus hybrid (M. australis x M. australasica))	Sydney hybrid

Scientific name	Common name
Citrus australasica F. Muell.	Australian finger lime
Citrus australis (Mudie) Planch.	Australian round lime
Citrus garrawayi F. M. Bailey	Mount White lime, Garraway's Australian wild lime
Citrus glauca (Lindley) Burkill	Australian desert lime
Citrus gracilis Mabb.	Humpty Doo lime
Citrus inodora F. M. Bailey	Russell River lime, large leaf Australian wild lime

12.11 Appendix 11: Australian native citrus species

12.12 Appendix 12: Standard diagnostic protocols

For a range of specifically designed procedures for the emergency response to a pest incursion refer to Plant Health Australia's PLANTPLAN (**www.planthealthaustralia.com.au/plantplan**).

12.13 Appendix 13: Resources and facilities

Formal diagnostic services for plant pests in Australia are delivered through a network of facilities located in every state and territory. These services are provided by a range of agencies, including state and territory governments, the Australian Government, commercial and private diagnostic laboratories, museums, CSIRO and universities. A current listing of these facilities can be found at http://plantbiosecuritydiagnostics.net.au/.

The national network is supported by the Subcommittee on Plant Health Diagnostics (SPHD), which was established to improve the quality and reliability of plant pest diagnostics in Australia. SPHD also manages the production of National Diagnostic Protocols.

For more information on the diagnostic services, or to identify an appropriate facility to undertake specific pest diagnostic services, refer to http://plantbiosecuritydiagnostics.net.au/ or contact the SPHD Executive Officer on **SPHD@agriculture.gov.au**.

12.14 Appendix 14: Communications strategy

A general Communications Strategy is provided in Section 4.1.5 of PLANTPLAN (Plant Health Australia, 2014).

Exercise Yellow Dragon simulated the response to an incursion of huanglongbing in Asiatic citrus psyllid in an urban environment (PHA 2015). One of the key learnings from this exercise was that a community engagement plan targeted specifically for urban environments was required. This was particularly the case for an Asiatic citrus psyllid/huanglongbing response but would have general application for other pests where hosts are common in urban environments. Key audiences in the response included: households with hosts, citrus industry generally, citrus industry within incidence zone, first detectors (for industry surveillance), production and retail nurseries, plant hire companies, exporters and traders, general public, export market, retailers, local government/councils, Local Land Services, school groups, private horticultural contractors, and potentially ethnic groups (where translated communication may be required).

12.15 Appendix 15: Market access impacts

Within the Department of Agriculture and Water Resources Manual of Importing Country Requirements (MICoR) database (**www.agriculture.gov.au/micor/plants/**) export of some material may require an additional declaration regarding freedom from the pest and/or bacteria. Should huanglongbing or the Asiatic or African citrus psyllids become established in Australia, some countries may require specific declarations. Latest information can be found within MICoR, using the search function.

The Department of Agriculture and Water Resources MICoR database was searched in October 2014 for current trade restrictions relating to huanglongbing (*Candidatus* spp.) and the Asiatic or African citrus psyllids. Note that there are several synonyms and it was necessary to search for '*Candidatus*' as well as 'huanglongbing' and 'citrus greening' to find these results on MICoR. No countries were identified on the Department of Agriculture and Water Resources MICoR database as having trade restrictions regarding the African citrus psyllids and only one country (Italy) has restrictions in lace relating to the Asiatic citrus psyllid. See Table 16 for details.

Country	Commodity	Requirements/restrictions
Italy	Microcitrus australasica	Declaration required that states Citrus greening (' <i>Candidatus</i> Liberibacter spp.') and <i>Diaphorina citri</i> are known not to occur in Australia
New Zealand	Citrus spp. seed	Declaration required that states Citrus greening (' <i>Candidatus</i> Liberibacter spp.') is not known to occur in Australia
New Zealand	Fortunella spp. seed	Declaration required that states Citrus greening (' <i>Candidatus</i> Liberibacter spp.') is not known to occur in Australia
New Zealand	Poncirus spp. seed	Declaration required that states Citrus greening (' <i>Candidatus</i> Liberibacter spp.') is not known to occur in Australia
South Africa	<i>Citrus</i> spp. nursery stock - cuttings	Declaration required that states Citrus greening (' <i>Candidatus</i> Liberibacter africanus', ' <i>Candidatus</i> Liberibacter asiaticus') and American greening (' <i>Candidatus</i> Liberibacter americanus') are not known to occur in Australia
South Africa	<i>Citrus</i> spp. nursery stock – tissue culture	Declaration required that states Citrus greening (' <i>Candidatus</i> Liberibacter africanus', <i>'Candidatus</i> Liberibacter asiaticus') and American greening (' <i>Candidatus</i> Liberibacter americanus') are not known to occur in Australia
South Africa	Citrus spp. seed	Declaration required that states Citrus greening (' <i>Candidatus</i> Liberibacter africanus', <i>'Candidatus</i> Liberibacter asiaticus') and American greening (' <i>Candidatus</i> Liberibacter americanus') are not known to occur in Australia
United States of America	Citrus spp. seed	Declaration required that states huanglongbing is not known to occur in Australia

Table 16. Countries identified on the Department of Agriculture and Water Resources MICoR

 database that have trade restrictions in relation to huanglongbing (as at December 2015)

Country	Commodity	Requirements/restrictions
United States of America	Clymenia polyandra seed	Declaration required that states huanglongbing is not known to occur in Australia
United States of America	<i>Microcitrus australasica</i> seed	Declaration required that states huanglongbing is not known to occur in Australia

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