

***Candidatus Liberibacter asiaticus***

**System:** Terrestrial

Kingdom	Phylum	Class	Order	Family
Bacteria	Proteobacteria	Alphaproteobacteria	Rhizobiales	Rhizobiaceae

<b>Common name</b>	dieback (English, India), blotchy mottle (English, South Africa), vein phloem degeneration (English, Indonesia), huanglongbing (HLB) (Chinese), greening (English, South Africa), mottle leaf (English, Philippines), citrus greening disease (English), yellow shoot disease (English), likubin (English, Taiwan), Enverdecimiento (Spanish), yellow branch (English, South Africa)
<b>Synonym</b>	<i>Candidatus Liberobacter asiaticus</i> <i>Candidatus Liberobacter asiaticum</i>
<b>Similar species</b>	<i>Candidatus Liberibacter africanus</i> , <i>Candidatus Liberibacter americanus</i>
<b>Summary</b>	Huanglongbing (HLB) or citrus greening disease is a destructive disease of citrus caused by gram-negative phloem-restricted bacteria belonging to the genus <i>Candidatus Liberibacter</i> . The genus comprises three known species: <a href="#">Candidatus Liberibacter asiaticus</a> , occurring in Asian countries and, to a lesser extent, in Brazil and the USA (Florida), <a href="#">Candidatus Liberibacter africanus</a> , recorded from African countries, and <a href="#">Candidatus Liberibacter americanus</a> present in Brazil and Florida. It is thought that each <i>Liberibacter</i> species evolved in the continent after which it is named. HLB is transmitted between trees by the psyllids <i>Trioza erytrae</i> in Africa and <i>Diaphorina citri</i> in Asia and America. HLB affects all commercial citrus varieties, causing mottling of leaves, stunting of growth and formation of small, deformed fruit which fail to colour properly. HLB can destroy citrus groves within 5 to 8 years. Apart from prevention there are no control measures currently available, causing HLB to often be described as the most destructive and serious disease of citrus.



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## Species Description

The disease known commonly as huanglongbing (HLB) is caused by gram-negative bacteria with a double-membrane cell envelope in the genus *Candidatus Liberibacter*. There are three known species which cause HLB in different regions of the world: [Candidatus Liberibacter asiaticus](#), [Candidatus Liberibacter africanus](#) and [Candidatus Liberibacter americanus](#). None of the *Candidatus* species of *Liberibacter* have been cultured despite efforts by researchers (Li *et al.* 2008), hence the term *Candidatus* which indicates an organism that has not been cultured and is characterized on the basis of DNA properties (Murray and Schleifer 1994 in Bove and Ayres 2007). However a very recent study by Sechel *et al.* (2009) reports the successful cultivation of all three species of the genus.

The three known species of *Liberibacter* cause essentially the same symptoms wherever HLB occurs. Bove (2006) reports that "Infected trees show a blotchy mottle condition of the leaves that results in the development of yellow shoots, the early and very characteristic symptom of the disease. Trees are stunted, declining and bear a few, small-sized, and deformed (lop-sided) fruits, that are poorly coloured (greening) and with coloration starting at the peduncular end (colour inversion)." However there are no specific symptoms of HLB that can be used as a diagnostic (Bove 2006). Time from infection to appearance of symptoms ranges from less than a year to several years depending on season, environmental conditions, tree age, host species/cultivar and health of tree (Jepson 2008).

For these reasons additional techniques are required for detection. Electron microscopy (EM) has been used in the past for detection of HLB. Detection is based on the location of HLB in the sieve tubes, and the presence of a cell wall; features which no other citrus-infecting bacteria possess (Bove 2006). However there are no morphological differences to distinguish between the *Liberibacter* species which cause HLB.

The African and Asian form can be distinguished based on serological methods or by temperature sensitivity, as [Candidatus Liberibacter africanus](#) is heat sensitive, while [Candidatus Liberibacter asiaticus](#) is heat tolerant (Garnier *et al.* 1991; Gao *et al.* 1993 in Bove 2006). [Candidatus Liberibacter americanus](#) is also heat tolerant (Das *et al.* 2007). Other qualitative methods used for detection or identification of HLB pathogens include biological indexing (Roistacher 1991 in Li *et al.* 2008b), chemifluorescence (Schwarz 1968 in Li *et al.* 2008b) and enzyme-linked immunosorbent assay (Garnier and Bove 1993 in Li *et al.* 2008b). Gottwald *et al.* (2007) report that "PCR is now the main confirmatory test and is routinely used in many areas and particularly in Florida and Brazil both for diagnostics and as a prelude to disease management." Two PCR systems are commonly used; based on the 16S rRNA gene and the  $\beta$  operon (Bove and Ayres 2007).

For high quality images and descriptions please see Gottwald *et al.* (2007): [Citrus Huanglongbing: The Pathogen and Its Impact](#).

## Notes

Although HLB is currently attributed to *Candidatus Liberibacter* spp. it is possible that other plant pathogens contribute to this disease. Because *Ca. Li.* spp. are unable to be cultured, Koch's postulates which confirm the causal agent of a disease, cannot be fulfilled. Furthermore PCR results from citrus showing symptoms sometimes yield negative results. While the common explanation is that the bacterial titer is below detection limits, it may be that another pathogen is involved. A survey conducted in 2006-2007 in China sampled citrus showing HLB symptoms. A phytoplasma, termed HLB-associated phytoplasma, was associated with HLB symptoms more than was *Ca. L. asiaticus*. 29.1% of samples were detected with phytoplasma only; compared to just 14.2% detected with just *Ca. L. asiaticus*. This study indicates that phytoplasma may have a role in HLB but "more data, particularly those from controlled experiments, are needed to better establish the association of phytoplasma with HLB" (Chen *et al.* 2009).

## Lifecycle Stages

*Candidatus Liberibacter* are gram-negative bacteria with a double-membrane cell envelope found in the sieve tube elements of phloem. The bacteria are transmitted by psyllids as they feed. [Candidatus Liberibacter asiaticus](#) and [Candidatus Liberibacter americanus](#) are transmitted by the adults of the citrus psyllid *Diaphorina citri* Kuwayana. *Ca. L. africanus* is transmitted by the adult psyllid *Trioza erytreae* Del Guercio. The bacteria can be acquired by the insects in the nymphal stages and may be transmitted throughout the lifespan of the psyllid (Jepson 2008). The bacteria have been detected in the haemolymph and salivary glands of both psyllid vectors. *D. citri* has been demonstrated to retain the bacteria for 12 weeks, indicating that the bacteria may be replicating in the insect (Hung *et al.* 2004 in Manjunath *et al.* 2008). There is a remote possibility that [Candidatus Liberibacter asiaticus](#) bacteria are transmitted transovarially [transmission from parent to offspring] (Manjunath *et al.* 2008).

## Uses

Due to their devastating effects on citrus trees and their efficient transmission by the psyllid vectors, '*Ca. L. africanus*' and '*Ca. L. asiaticus*' have been classified by the US Government as 'select agents' with potential for bioterrorism (Teixeira *et al.* 2005)

## Habitat Description

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## Nutrition

*Candidatus Liberibacter* spp. are restricted to the sieve tube elements of the phloem. Kim *et al.* (2008) report that phloem is an ideal habitat for many pathogens due to the presence of rich nutrients in phloem sap.

## General Impacts

Of all citrus diseases HLB is often described as the most destructive and lethal (Bove 2006; Bove and Ayres 2007; Gottwald *et al.* 2007). Bove (2006) reports that “HLB symptoms are virtually the same wherever the disease occurs. Infected trees show a blotchy mottle condition of the leaves that results in the development of yellow shoots, the early and very characteristic symptom of the disease. Trees are stunted, declining and bear a few, small-sized, and deformed (lop-sided) fruits, that are poorly coloured (greening) and with coloration starting at the peduncular end (colour inversion).” The taste of the fruit is also affected. David Hall, an entomologist with USDA in Fort Pierce, Florida describes the taste as “jet fuel mixed with Vicks VapoRub” (Stokstad 2006). Kim *et al.* (2008) analyzed citrus response to HLB infection. Microarray analysis of citrus revealed that infection affected the expression of 624 genes including those involved with sugar metabolism, plant defense, phytohormone and cell wall metabolism. Anatomical analyses showed that HLB infection caused phloem disruption, sucrose accumulation, and plugged sieve pores. Phloem disruption and blockage is likely to be caused by host responses rather than by HLB bacterial aggregations. Plugged sieve pores and upregulation of genes involved in sucrose biosynthesis is thought to cause accumulation of sucrose in leaves leading to nutrient deficiencies in sink organs, stunting of plant growth, fruit maturation and seed development (Kim *et al.* 2008). HLB affects almost all citrus cultivars, and causes substantial economic losses to the citrus industry by shortening the lifespan of trees and making fruit inedible (Das *et al.* 2007). Gottwald *et al.* (2007) report that “almost 100 million trees have been affected and destroyed in many countries of South and Southeast Asia, Indonesia, Philippines, India, Arabian Peninsula, and South Africa, compromising the local citriculture (Aubert *et al.* 1985; Bove 1986; Halbert and Manjunath 2004; Toorawa 1998). Since 2004, more than 500 thousand trees were officially eliminated in Brazil due to HLB and it is estimated that an additional 300 to 400 thousand trees were unofficially eliminated by commercial citrus growers.”

## Management Info

At present there are no curative methods to control HLB. Thus control measures focus largely on prevention of infection. Please follow this link for more on the [management and control of Huanglongbing \(\*Candidatus Liberibacter\* spp.\)](#)

## Principal source:

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## Review:

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## ALIEN RANGE

[1] BELIZE  
[1] INDONESIA  
[1] SOLOMON ISLANDS

[1] CUBA  
[1] IRAN, ISLAMIC REPUBLIC OF  
[3] UNITED STATES

## BIBLIOGRAPHY

28 references found for *Candidatus Liberibacter asiaticus*

### Managment information

Beattie, G.A.C. & Barkley, P. 2009. Huanglongbing and its vectors: A pest specific contingency plan for the citrus and nursery garden industries, Volume 2.

- da Graca, J.V. & Korsten, L. 2004. Citrus huanglongbing: review, present status and future strategies. In S.A.M.H. Naqvi (Ed.), *Diseases of Fruits and Vegetables. Diagnosis and Management Volume 1* (pp. 229-245). Kluwer Academic Publishers.
- Ding, F., Shuangxia, J., Hong, N., Zhong, Y., Cao, Q., Yi, G. & Wang, G. 2008. Vitrification-cryopreservation, an efficient method for eliminating *Candidatus Liberibacter asiaticus*, the citrus Huanglongbing pathogen, from in vitro adult shoot tips. *Plant Cell Reports* 27: 241-250.
- Lopes, S.A., Frare, G.f., Yamamoto, P.T., Ayres, A.J. & Barbosa, J.C. 2007. Ineffectiveness of pruning to control citrus huanglongbing caused by *Candidatus Liberibacter americanus*. *European Journal of Plant Pathology* 119: 463-468.
- Manjunath, K.L., Halbert, S.E., Ramadugu, C., Webb, S. & Lee, R.F. 2008. Detection of *Candidatus Liberibacter asiaticus* in *Diaphorina citri* and its importance in the management of citrus huanglongbing in Florida. *Phytopathology* 98:387-396.
- McKenzie, C.L. & Puterka, G.J. 2004. Effect of sucrose octanoate on survival of nymphal and adult *Diaphorina citri* (Homoptera: Psyllidae). *Journal of Economic Entomology* 97(3): 970-975.
- Nakata, T. 2008. Effectiveness of micronized fluorescent powder for marking citrus psyllid, *Diaphorina citri*. *Applied Entomology and Zoology* 43(1): 33-36.
- Qureshi, J.A. & Stansly. 2008. Rate, placement and timing of aldicarb applications to control Asian citrus psyllid, *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae), in oranges. *Pest Management Science* 64: 1159-1169.
- Urasaki, N., Kawano, S., Mukai, H., Uemori, T., Takeda, O. & Sano, T. 2008. Rapid and sensitive detection of *Candidatus Liberibacter asiaticus* by cleavage isothermal and chimeric primer-initiated amplification of nucleic acids. *Journal of General Plant Pathology* 74: 151-155.

## General information

- APHIS. 2005. U.S. Department of Agriculture and Florida Department of Agriculture confirm detection of citrus greening. USDA Press Release.
- Bove, J.M. 2006. Huanglongbing: a destructive, newly-emerging century-old disease of citrus. *Journal of Plant Pathology* 88(1): 7-37.
- Bove, J.M. & Ayres, A.J. 2007. Etiology of three recent diseases of citrus in Sao Paulo state: sudden death, variegated chlorosis and huanglongbing. *IUBMB Life* 59(4-5): 346-354.
- Chen, J., Pu, X., Deng, X., Liu, S., Li, H. & Civerolo, E. 2009. A phytoplasma related to *Candidatus Phytoplasma asteri* detected in citrus showing huanglongbing (yellow shoot disease) symptoms in Guangdong, P. R. China. *Phytopathology* 99:236-242.
- Das, A.K., Rao, C.N. & Singh, S. 2007. Presence of citrus greening (huanglongbing) disease and its psyllid vector in the North-Eastern region of India confirmed by PCR technique. *Current Science* 92(12): 1759-1763.
- Davis, R.I. & Tsatsia, H. 2009. A survey for plant diseases caused by viruses and virus-like pathogens in the Solomon Islands. *Australasian Plant Pathology* 38: 193-201.
- [European and Mediterranean Plant Protection Organization \(EPPO\), 2009. EPPO Reporting Service, No. 5. Paris, 2009-05-01.](#)
- Garnier, M., Jagoueix-Eveillard, S., Cronje, P.R., Le Roux, H.F. & Bove, J.M. 2000. Genomic characterization of a liberibacter present in an ornamental rutaceous tree, *Calodendrum capense*, in the Western Cape province of South Africa. Proposal of *Candidatus Liberibacter africanus* subsp. *capensis*. *International Journal of Systematic and Evolutionary Microbiology* 50: 2119-2125.
- Gottwald, T. R., da Graça, J. V., and Bassanezi, R. B. 2007. Citrus Huanglongbing: The pathogen and its impact. Online. Plant Health Progress doi:10.1094/PHP-2007-0906-01-RV.
- Jepson, S.B. 2008. *Citrus Greening Disease (Huanglongbing)*. Oregon State University.
- Kim, J.S., Sagaram, U.S., Burns, J.K., Li, J.L. & Wang, N. 2009. Response of sweet orange (*Citrus sinensis*) to *Candidatus Liberibacter asiaticus* infection: Microscopy and microarray analyses. *Phytopathology* 99:50-57.
- Li, W., Levy, L. & Hartung, J. S. 2009. Quantitative distribution of *Candidatus Liberibacter asiaticus* in citrus plants with citrus huanglongbing. *Phytopathology* 99:139-144.
- Li, W., Li, D., Twieg, E., Hartung, J. S., & Levy, L. 2008. Optimized quantification of unculturable *Candidatus Liberibacter* spp. in host plants using real-time PCR. *Plant Disease* 92:854-861.
- NAPPO. 2008. Confirmation of Huanglongbing or Citrus Greening ( *Candidatus Liberibacter asiaticus* ) in Louisiana United States
- Qureshi, J.A., Rogers, M.E., Hall, D.G. & Stansly, P.A. 2009. Incidence of invasive *Diaphorina citri* (Hemiptera: Psyllidae) and its introduced parasitoid *Tamarixia radiata* (Hymenoptera: Eulophidae) in Florida citrus. *Journal of Economic Entomology* 102(1): 247-256.
- Sechler, A., Schuenzel, E.L., Cooke, P., Donnua, S., Thaveechai, N., Postnikova, E., Stone, A.L., Schneider, W.L., Damsteegt, V.D. & Schaad, N.W. 2009. Cultivation of *Candidatus Liberibacter asiaticus*, *Ca. L. africanus*, and *Ca. L. americanus* associated with huanglongbing. *Phytopathology* 99:480-486.
- Stokstad, E. 2006. New disease endangers Florida's already-suffering citrus trees. *Science* 312: 523-524.
- Teixeira, D.C., Saillard, C., Eveillard, S., Danet, J.L., Costa, P.I., Ayres, A.J. & Bove, J. 2005. *Candidatus Liberibacter americanus*, associated with citrus huanglongbing (greening disease) in Sao Paulo State, Brazil. *International Journal of Systematic and Evolutionary Microbiology* 55: 1857-1862
- Trivedi, P., Sagaram, U.S., Kim, J.S., Brlansky, R.H., Rogers, M.E., Stelinski, L.L., Oswalt, C. & Wang, N. 2009. Quantification of viable *Candidatus Liberibacter asiaticus* in hosts using quantitative PCR with the aid of ethidium monoazide (EMA). *European Journal of Plant Pathology*, in press.