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### SYNERGY OF MEASURES AND CONTROL STRATEGIES IN HUANGLONGBING DISEASE (HLB) OF CITRUS: INTEGRATED APPROACHES FOR PREVENTION AND EFFECTIVE COMBAT

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#### SUMMARY

Citrus Greening, also known as Huanglongbing (HLB), is one of the most devastating diseases affecting citrus crops worldwide. This disease is caused by a bacteria transmitted by*psyllids*, insects that feed on plant sap. Greening results in serious symptoms, such as wilting and yellowing of leaves, compromising fruit production and quality, and, in many cases, leading to the death of trees. Losses have been very significant in several parts of the world, especially in Florida, where crops have been decimated, resulting in major economic losses for the citrus sector. Faced with this threat, the adoption of appropriate control measures and strategies is essential for preventing and effectively combating the disease. The general objective of this work is to investigate and present a synergistic and integrated approach to control measures and strategies for Citrus Greening, aiming to prevent the spread of the disease, combat its spread and promote adequate and sustainable management of citrus crops. The methodology used in this work consists of a comprehensive and systematic bibliographical review on the topic of controlling Citrus Greening.

Key words:Citrus Greening. Huanglongbing. Integrated pest management. Citrus diseases.

## 1. INTRODUCTION

Citrus farming is an agricultural activity of extreme importance for the global market, contributing significantly to the economy and food supply in different regions of the world. Citrus production, which includes oranges, lemons, tangerines and other citrus fruits, represents a significant share of the agricultural industry and generates billions of dollars annually (Citrus Defense Fund, Fundecitrus, 2023).

However, this vital sector of agriculture has faced an increasing challenge due to one of the most devastating diseases affecting citrus: Citrus Greening, or Huanglongbing (HLB). This disease is caused by a bacteria transmitted by psyllids, sap-sucking insects that spread the disease by feeding on diseased and healthy plants. Greening results in serious symptoms in trees, such as yellowing and wilting of leaves, as well as deformed and low-quality fruits (Girardi*et al.*, 2017).

According to Khan and Razi (2018), Citrus Greening is considered the worst citrus disease worldwide, and has already caused substantial losses in several producing regions. One of the most alarming examples is Florida, United States, where citrus farming is an economic activity of great importance. According to data from the United States Department of Agriculture (USDA), citrus crops in Florida have been severely affected by greening, resulting in a drastic reduction in production in recent years (USDA, 2021).

This disease is caused by the bacteria *Candidatus Liberibacter*spp., transmitted mainly by psyllids *Diaphorina citri*It is *Trioza erytreae*(Girardi*et al.*, 2017). Greening symptoms include irregular yellowing of leaves, reduction in fruit size and quality, and eventually tree death (Liberato, 2017). These devastating effects have put the sustainability of citrus farming and the livelihoods of producers in different parts of the world at risk (Khan; Razi, 2018).

It is essential to highlight that Greening has challenged control efforts through isolated <u>approaches. S</u>tudies by several researchers indicate that, although some management tactics have 1 Graduated in Agribusiness from the Faculty of Technology of Rio Preto, FATEC; Specialist in Production Engineering. two Graduated in Agribusiness from the Faculty of Technology of Rio Preto, FATEC, specialist in Environmental and Sanitary Engineering (studying).

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demonstrated some success in reducing the population of *psyllids*transmitters, have not been sufficient to effectively contain the spread of the disease (Bové, 2006; Hall et al., 2013). These failures emphasize the need to adopt an integrated approach to control, combining different techniques and strategies (FUNDECITRUS, 2021).

The synergy of control measures has shown promise in reducing the incidence of greening and mitigating its negative effects on citrus farming. The use of resistant plants, adequate fertilizer management and the judicious application of selective pesticides are some examples of integrated tactics that have been proposed (Baldwin*et al.*,2019; Tomaseto*et al.*, 2021). Furthermore, constant monitoring and early detection of the presence of the disease have proven to be fundamental to allow a quick and effective response (Grafton-Cardwell*et al.*, 2013).

In this context, the general objective of this article is to investigate and present a synergistic and integrated approach to control measures and strategies for Citrus Greening, aiming to prevent the spread of the disease, combat its spread and promote adequate and sustainable management of citrus crops. citrus. To achieve this objective, a comprehensive bibliographical review will be carried out, using data from scientific studies and renowned research, which will serve as a basis for discussing the best practices for controlling and managing Greening in citrus farming.

#### 2 CITRUS GREENING: BIOLOGICAL AND EPIDEMIOLOGICAL ASPECTS

Citrus Greening, also known as Huanglongbing (HLB), is a disease of great relevance to the citrus industry worldwide. Its occurrence has become a growing concern for producers and researchers due to the devastating impacts it causes in citrus groves. According to Gottwald (2010), Greening is one of the most serious diseases with the greatest economic impact on the citrus sector worldwide.

The disease is caused by a bacteria of the genus *Candidatus Liberibacter*spp., which is transmitted by psyllids, small sap-sucking insects. According to Bové (2006), the interaction between the bacteria, the vector psyllids and the host plants is complex and directly influences the development and spread of Greening.

One of the main symptoms of Greening is the irregular yellowing of the leaves, also known as " *mottle chlorosis*", which affects photosynthesis and nutrient translocation in plants. Furthermore, the disease results in low quality fruits, due to reduced size and poor formation, damaging the productivity and commercialization of citrus (Grafton-Cardwell et al., 2013).

The spread of Greening occurs mainly through the action of psyllid vectors, which feed on infected plants and subsequently transport the bacteria to healthy trees. These insects are highly mobile and can travel long distances, facilitating the rapid spread of the disease in citrus growing areas (Hall et al., 2013).

Understanding the epidemiological aspects of greening is essential for developing effective control strategies. Early detection of the disease and implementation of appropriate management measures can help limit the spread of the pathogen and minimize damage to orchards. According to Tomaseto et al. (2021), the rapid identification of diseased trees and the eradication of these plants are important measures to control the disease.

Furthermore, the resistance of host plants to Greening has been the subject of study and research. The search for disease-resistant citrus varieties is a promising strategy to reduce the impacts of greening in citrus farming (Tomaseto et al., 2021).

The international spread of Greening has aroused the concern of phytosanitary authorities, who have implemented strict control measures to prevent the entry and spread of the disease in different countries. The adoption of quarantines, restrictions on trade in plant material and monitoring and control actions are some of the actions adopted to minimize the risks associated with Greening (Bové, 2006).

The complexity of the interaction between the bacteria that causes Greening, the psyllid vectors and citrus plants makes understanding the epidemiology of the disease fundamental. Studies such as those by Grafton-Cardwell et al. (2013) provide relevant information about the behavior and population dynamics of vectors, as well as their relationships with pathogens and host plants. This knowledge is essential for

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adoption of control measures that are effective in reducing the spread of the disease.

The evolution of Citrus Greening and its rapid dissemination have raised concerns about the sustainability of citrus farming on a global scale. The economic and social implications resulting from the damage caused by the disease are significant, as evidenced by studies by Bové (2006) that describe the impacts of Greening in different regions of the world. Coordinated actions and appropriate public policies are essential to guarantee the viability of citrus farming and food security in the face of this threat.

Faced with this challenging scenario, it is essential to promote integrated greening control and management actions, based on scientific knowledge and the exchange of information between researchers, producers and phytosanitary authorities. The search for sustainable and environmentally responsible solutions, as suggested by Tomaseto et al. (2021), is essential to guarantee the protection of citrus groves and the health of agricultural ecosystems.

## 2.1 Chemical Control Tactics and Integrated Pest Management for Citrus Greening

Controlling Citrus Greening is a challenging task that requires the adoption of several strategies, including chemical control and integrated pest management. The use of agricultural pesticides has been one of the main approaches to control the psyllid vectors of the disease. Studies such as those by Hall et al. (2013) have evaluated the effectiveness of different insecticides in combating psyllids, aiming to reduce the population of these transmitting insects.

However, the exclusive use of pesticides has proven to be insufficient to effectively control Greening. An integrated approach that also includes other management tactics is needed. The use of sticky traps to monitor the presence of psyllids, for example, is an important measure to assess the population density of these insects in orchards (Grafton-Cardwell et al., 2013).

Monitoring the psyllid population is a fundamental recommendation in the management of Citrus Greening. Using the yellow sticky trap, it is possible to detect insect entry points into orchards and determine the appropriate time to apply control measures. Studies indicate that the trap is more efficient than other monitoring methods, especially in areas with strict chemical control. However, identifying captured psyllids requires periodic training and retraining of inspectors. Regular monitoring and correct identification of insects are essential to ensure effective control and contribute to the protection of orchards and the sustainability of citrus farming (FUDECI-TRUS, 2017).

Furthermore, integrated pest management involves controlling invasive plants, eliminating diseased plants, and implementing appropriate cultural practices. Control of excessive sprouting, elimination of ladies of the night in the region, monitor psyllid *Diaphorina citri*, pruning affected branches and balanced plant nutrition are actions that can reduce the susceptibility of trees to greening (Dutra, 2018).

To ensure the efficiency and safety of chemical control tactics, compliance with good agricultural practices is essential. The correct application of pesticides, the choice of selective products and consideration of withdrawal periods are measures that must be strictly followed by producers (Baldwin et al., 2019).

The resistance of host plants to psyllids and the Greening pathogen is another important front of research for disease control. Studies such as those by Tomaseto et al. (2021) have investigated the identification and selection of citrus varieties that are more resistant to infection and damage caused by the bacteria.

Understanding psyllid ecology and pathogen transmission dynamics is also crucial to guide the efficient application of chemical control tactics and integrated pest management. The interaction

between control measures and vector biology can significantly influence the effectiveness of control measures. strategies adopted (Hall et al., 2013).

# 2.2 Host Plant Resistance and Other Promising Strategies for Greening Control of Citrus

Host plant resistance is a promising strategy for controlling citrus greening. Studies have been conducted to identify citrus varieties that present greater resistance to infection

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by bacteria *Candidatus Liberibacter asiaticus*, the main causative agent of the disease. Research such as that of Tomaseto et al. (2021) have investigated the resistance mechanisms present in different citrus genotypes, seeking to identify those that can be used in the development of cultivars more resistant to greening.

According to Arenas (2017), infection by the bacteria *Candidatus Liberibacter asiaticus* is the main causative agent of Greening disease in Citrus. This bacterium is transmitted by vector psyllids, such as *Diaphorina citri*, when they feed on contaminated plants. Once inside the tree, the bacteria multiplies and spreads, causing obstruction of the sap-conducting vessels, which leads to the development of characteristic symptoms of the disease, such as yellowing leaves, deformations in the fruits and reduced production. O*Candidatus Liberibacter asiaticus*It is highly destructive to citrus plantations, representing a significant threat to global citrus farming and making the search for effective control and management strategies to combat Greening essential.

In addition to host plant resistance, other promising strategies have been investigated to control Citrus Greening. Among them, the use of biological control agents stands out, such as natural enemies of the psyllid vectors and the use of pheromone traps for monitoring and controlling pests. Studies by Grafton-Cardwell et al. (2013) have demonstrated the effectiveness of parasitoids and predators in reducing the psyllid population, contributing to controlling the spread of greening.

The use of appropriate cultivation and management techniques also plays an important role in controlling the disease. Correct pruning, balanced plant nutrition and control of invasive plants can help reduce trees' susceptibility to greening (Gottwald, 2010). Studies by Baldiwin et al. (2019) demonstrated that appropriate cultural practices can contribute to improving the health of citrus plants and the quality of the fruits.

Another promising approach is the use of gene editing techniques to develop plants that are more resistant to greening. Researchers have explored CRISPR-Cas9 technology to modify genes in citrus and improve their disease resistance (Tomaseto et al., 2021). This approach has the potential to create citrus varieties with specific resistance characteristics, contributing to the control of greening in a sustainable way.

Furthermore, awareness and active participation of everyone involved in the citrus production chain are fundamental to the success of greening control strategies. Collaboration between producers, researchers, phytosanitary authorities and society as a whole is essential to implement and improve disease control measures.

Host plant resistance, the use of biological agents, appropriate cultivation techniques and gene editing are promising strategies that have been studied to control Citrus Greening. The search for integrated and sustainable solutions is essential to protect citrus farming and ensure the continuity of citrus production in the face of this global threat.

#### **3 DISCUSSION**

The analysis of the studies presented highlights the importance of adequate management and synergy of strategies in combating Citrus Greening. Studies show that simply adopting a single combat strategy is not enough to eradicate the disease. On the contrary, it is crucial to apply several tactics, such as chemical control, the elimination of diseased plants, monitoring of psyllids, the use of traps and the resistance of host plants, in a synergistic way. The integration of these measures allows you to maximize control efficiency and prevent the spread of Greening.

The discussion about the importance of adequate management is highlighted, as studies show that adequate cultural practices, such as correct pruning, balanced nutrition and control of invasive plants, can reduce the susceptibility of trees to greening. This reinforces the need to train producers with knowledge about good agricultural practices, in order to guarantee the health and productivity of orchards.

Li's study*et al.* (2021), with the aim of analyzing current strategies from physical, chemical and biological aspects and discussing their environmental impacts, proposed a green and ecological strategy to control HLB based on existing methods and the results of previous research. The study proposes a combination of physical, chemical and biological techniques and strategies, such as the use of insecticides and the release of enemy pests.

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natural. In addition to strategies, such as the use of organic fertilizers and the spraying of immune activators, aiming to improve the plant growth environment and increase plant immunity, also removing diseased plants, to reduce transmission because the effect of medicines is guided due to the severity of the disease in citrus infected by HLB.

The adoption of technology is also relevant in preventing and controlling greening. The use of pheromone traps, for example, allows efficient monitoring of psyllid vectors, enabling early detection of the pest and rapid control measures. Furthermore, the application of gene editing techniques can boost the development of citrus varieties that are more resistant to greening, strengthening citrus farming against this threat.

Taken together, these strategies and tools represent a holistic approach to controlling Citrus Greening. The synergy between management measures, chemical control, host plant resistance and the use of technology is essential to sustainably combat this devastating disease, ensuring food security, the economy and the health of citrus orchards around the world. Raising awareness among all actors involved in the production chain and collaboration between researchers, producers and phytosanitary authorities are fundamental to the success of this global confrontation. Only with an integrated and collaborative approach will it be possible to overcome the challenges posed by Greening and preserve citrus farming for future generations.

In addition to the strategies already mentioned, it is important to highlight that the resistance of host plants has also been the subject of intense studies aimed at controlling Greening. The search for citrus varieties that present greater resistance to the bacteria *Candidatus Liberibacter asiaticus*. It is a promising perspective for minimizing the effects of the disease on orchards. Several researchers have dedicated themselves to identifying the resistance mechanisms present in different citrus genotypes, seeking to develop cultivars that are more resistant to Greening. These resistant varieties would not only contribute to disease prevention, but would also reduce the need for excessive pesticide applications, making control more sustainable (ALVES*et al.* 2022).

Synergy between the different strategies is essential to obtain more effective results in the fight against Greening. The combined use of pheromone traps to monitor and control the psyllid population, along with the eradication of diseased plants and targeted chemical control, can help reduce the incidence of the disease in orchards. Integrated management, by covering a variety of tactics, provides a more complete and balanced approach to facing the Greening challenge.

Furthermore, the incorporation of technology in orchard management also stands out as a powerful tool for preventing and controlling greening. The use of gene editing techniques, such as CRISPR-Cas9 technology, can open new perspectives for the development of citrus varieties resistant to the disease. Gene editing makes it possible to modify specific genes to strengthen plant resistance, paving the way for an innovative and precise approach to greening control.

However, it is important to emphasize that none of these strategies should be considered in isolation. Adopting an integrated approach, which considers the combination of different measures and technologies, is crucial to tackle Greening in an efficient and sustainable way. Furthermore, collaboration between research institutions, producers and government authorities is essential to promote the implementation and continuous improvement of these strategies.

#### **4 CONCLUSION**

Effective control of Citrus Greening is a complex challenge that requires an integrated and sustainable approach. The strategies discussed in this article demonstrate that the synergy between different measures, such as Proper management, the use of technology and the search for resistance in host plants is essential for face this devastating disease. The combination of these tactics can result in more efficient and lasting control, reducing the negative impacts of greening on orchards and citrus farming as a whole. Host plant resistance presents itself as a promising perspective for control of Greening, allowing the development of citrus varieties that are more resistant to infection by the

bacteria that cause the disease. Furthermore, the use of pheromone traps and other advanced technologies enables precise monitoring of the psyllid vector population, allowing for timely and efficient intervention.



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The importance of adequate management is also highlighted, as appropriate cultural practices, such as Correct pruning and control of invasive plants can strengthen plants and reduce their susceptibility to greening. Combined, these integrated approaches contribute to the sustainability of citrus farming and the protection of plantations against this serious phytosanitary threat.

Therefore, it is essential that producers, researchers, government authorities and society in general are engaged in this joint effort to control Greening. Collaboration between different actors, the search for innovation and sharing of knowledge are fundamental to achieving success in combating this disease. Only through an integrated and cooperative approach, with a focus on sustainability and synergy of strategies, will we be able to protect and strengthen citrus farming, guaranteeing the production of quality citrus and food security on a global scale.

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