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CITRUS GUMMOSIS: A GLOBAL THREAT TO CITRUS PRODUCTION AND QUALITY-DISTRIBUTION, DIAGNOSIS, AND MANAGEMENT STRATEGIES

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ABSTRACT

Citrus, the most economically important fruit crop worldwide, faces numerous challenges from various diseases caused by fungi, bacteria, viruses, nematodes, and spiroplasma. Among these, gummosis, primarily induced by *Phytophthora* spp., poses a significant threat to citrus orchards, leading to substantial yield losses. *Phytophthora* spp., *P. citrophthora* and *P. nicotianae*, are the most prevalent pathogens responsible for gummosis in citrus-growing regions worldwide. The symptoms of gummosis, characterized by gummy substance formation and rotting roots, are described in detail, along with observations of gum exudation, cracks, cankers, and necrotic lesions on various parts of the plant. The review explores the historical background of citrus gummosis, tracing its initial outbreaks and identifying *Phytophthora* spp., as the causal agent. Understanding the epidemiology and ecology of gummosis is crucial for effective disease management. Factors influencing disease development, such as humidity, temperature, soil moisture, and rainfall, are discussed based on studies conducted in different regions. This highlights the pathogen's life cycle, emphasizing the importance of soil contact and water in the dispersal of *Phytophthora* spp. The comprehensive review provides valuable insights into the current knowledge of citrus gummosis, facilitating further research and development of sustainable management strategies to combat this devastating disease.

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INTRODUCTION

The citrus is the world's most valuable fruit crop. The Punjab province accounts for a significant share of Pakistan's total citrus production. Citrus production is reduced due to biotic and abiotic factors. Citrus diseases (fungi, bacteria, viruses, nematode, and spiroplasma) are the main reason for lower yield (Ahmad et al., 2004; Iqbal et al., 2004a, b, 2005a,b, 2007; Mukhtar et al., 2007; Irshad et al., 2012; Saeed et al., 2019; Sajid et al.,

2022). A number of fungal diseases are responsible for low citrus yield (Fateh et al., 2017). A few are essential citrus diseases caused by *Phytophthora* spp., belong to Oomycetes (Rehman et al., 2022).

Phytophthora spp., is responsible for billions of dollars in losses every year. This pathogen is among the most influential disease-causing organisms responsible for many diseases in economically important plants worldwide, like citrus, rubber, cocoa, coconut, papaya,

forest trees, tomato, potato, and pepper. The citrus Gummosis disease is mainly caused by *Phytophthora* spp., such as *P. nicotianae*, *P. citrophthora* and *P. palmivora* (Drenth and Guest, 2004). Gummosis has been reported to cause approximately 30% per year of yield losses (Mounde et al., 2009). Naqvi and Singh (2002) stated that the fast spread of this disease and the fact that it is a soil-borne fungus make it challenging to track and manage. Once *Phytophthora* enters a field, it establishes and becomes an endemic problem. It assessed regarding propagule count up to 250-350/cc in infected orchard soil. In an orchard, the propagule count can generally be around 1-20/cc, but it can reach 100-200/cc in less time. The feeder and fibrous root can majorly affect the population exceeding 10 propagules/cc, causing their decay and resulting in sudden plant decline. A single sporangium releases 5-40 zoospores. The production and germination of sporangium depend on the soil, water and potential temperature. The zoospores produced from sporangia are motile and show flagella movement in the water at a short distance and are carried to a longer distance through soil water. Root exudates attract these zoospores, and they swim toward them and encyst upon contact. The cyst, upon germination, penetrates the cortex directly or through wounds, and natural openings. Zoospores can infect any part of the plant if the surface remains wet for at least 18 hours. It can form a germ tube penetrating the leaves, roots, twigs, or fruits, even without wounds (Graham and Timmer, 2006). *Phytophthora* spp., primarily consists of persistent, ovoid, and papillate sporangia and has a heterothallism and epigynous antheridia (Alves et al., 2016). The scale of sporangia recorded from 56×35 - $33 \times 2.5 \mu\text{m}$ with a length and breadth ratio of 1.4:1 (Santos et al., 2005). The intercalary or terminal chlamydospores have a diameter of 25.4 - $40.3 \mu\text{m}$ and the size of the oospores is $29 \mu\text{m}$. The sporangiophore has swollen hyphae and is sympodially branched. Antheridia and spherical oogonia have sexual structures measuring a diameter of $28.6 \mu\text{m}$ (Boughalleb-M'hamdi et al., 2018). High humidity and temperatures encourage the vigorous production of the disease in citrus orchards. Soil-borne pathogen, attacks the plant when the scion comes in contact with soil or irrigation water. The symptoms emerge after a few days, such as root discoloration, yellowing and wilting of young plants, reduced feeder roots, lesions forming, and emerging gummy material from them (Rajput et al., 2020). The most economically relevant fungal diseases in the citrus

industry are gummosis caused by *Phytophthora* spp., damping off, and root rot (Leoni and Ghini, 2006). The most recognized species of *Phytophthora* in citrus growing regions worldwide are *P. citrophthora* and *P. nicotianae*. Compared to *P. citrophthora*, *P. nicotianae* grows at a higher temperature while targeting the rootlets that cause gummosis and brown fruit rot (Cacciola and di San Lio, 2008).

Historical perspective

The first *Phytophthora* outbreak was found in citrus orchards in Azore, Ireland, between 1832 and 1836. It was mentioned in France in 1841 and spread to Portugal and the Mediterranean countries in 1845. In California, citrus regions were later confirmed to be infected by citrus gummosis in 1876 (Fawcett, 1936). The threatening pathogen was found to kill citrus orchards worldwide until 1914 (Fawcett, 1936). The cause of this outbreak was believed to be many bacteria and fungi, but then Charles Moore went to Spain and identified *Phytophthora* spp., as the cause of the huge damage. The true cause behind this disease was *P. citrophthora*, but later *P. parasitica* and *P. palmivora* were also isolated from the samples tested in California (Kaur et al., 2023). *P. nicotianae* was identified as the cause of citrus gummosis (Ho and Jong, 1989). Graham and Timmer, (2006) discussed the etiology and ecology of gummosis and foot rot. They studied the behaviour of the fungus and briefly explained symptoms, favorable environmental conditions and the growth cycle. *P. nicotianae* isolated from lavender and rose merry in Spain, which is the cause of rot and collar rot diseases (Alvarez et al., 2008). Ahmed et al., (2012) investigated about dominant *Phytophthora* spp., in the citrus grove in Syria. They selected samples, isolated fungal species, and performed their morphological tests. Results indicated that *P. citrophthora* was the predominant species in citrus orchards of Syria. Savita and Nagpal, (2012) studied different diseases of citrus caused by *Phytophthora* spp., in India. In this study history, Epidemiology, distribution, disease cycle, mode of action and spreading of *Phytophthora* spp., were also discussed. Choudhari et al., (2018) studied the epidemiology of citrus gummosis caused by *Phytophthora* spp., in Nagpur, India. Six different locations were selected for the collection of data. The result indicated that all soil factors, humidity, and rainfall have a positive relationship with disease development and a negative relationship with temperature.

Symptoms

Citrus gummosis is a well-known disease usually characterized by gummy substance formation or rotting roots. Gum forming is seen on the trunk and branches of the plants. The gum exudes from the blisters, and a gum pocket from which it secretes out on plant's surface is found in each blister (Rehman et al., 2022). The wood reveals a pinkish-orange colour under the blisters. Adult trees exhibit declined symptoms associated with leaf chlorosis, dieback of twigs, discolored or low-coloured fruits, wilting tips and withering leaves. On the above-ground areas near the soil, signs are visible. Cracks and cankers are evident on barks, roots, trunks, and exude gum. Branches give a bleeding experience. Pathogen is carried by the soil; the symptoms are found from the baseline to upwards on the primary to secondary branches of the tree. Necrosis (cell death) is found on taproots and crowns, with the increased severity of the infection spreading necrotic lesions and resulting in tree death (Naqvi, 2000). The gum exudes from the longitudinal cracks of bark covering the necrotic regions are detected. There appears a water-soaked discolouration that is persistent when the conditions are dry. The gummy content is naturally water-soluble, soaked away during the rain and ends in a discolored cortex. Boughalleb-M'hamdi et al., (2018) reported that citrus varieties were infected viz *C. hernandina* (73%), *T. navel* (16%), *Maltese* (10%), and *M. tangerine* (1%). The common symptom is longitudinal cracks with profuse gumming on the bark. Slimy, water-saturated, and reddish brown tends to be the bark near soil that becomes black. At an advanced stage, yellow, scattered vegetation is observed. Due to the pathogen, s girdling operation, the tree later collapses. The necrotic bark turns porous and sloughs away from the middle, forming callous. If signs of canker extend to more than 50% of the trunk, so there is a decline in the canopy (Cacciola and di San Lio, 2008). Leaf midrib and vein chlorosis, phyllotopsis, canopy thinning, and branch dieback occur. On the bark, Graham and colleagues recorded tiny cracks with gummy material exuding out. There was a confirmed distribution of lesions all over the trunk. They observed dead trees in infected orchards. Defoliated trees, twig dieback, and stunted flush growth have also been reported (Graham et al., 2003).

Prevalence of *Phytophthora* species

Phytophthora contains about 120 species and causes destructive diseases in crops, trees, fiber, and

ornamental plants worldwide, categorized into oomycetes (Rajput et al., 2015). Twelve species *P. boehmeriae*, *P. cactorum*, *P. capsici*, *P. cinnamomi*, *P. citricola*, *P. citrophthora*, *P. drechsleri*, *P. hibernalis*, *P. megasperma*, *P. palmivora*, *P. nicotianae*, and *P. syringe*, have been confirmed pathogenic to citrus from various citrus-growing regions of the world, resulting in substantial economic losses (Naqvi, 2004). Two more species viz. *P. insolita* and *P. humicola* recently isolated from the soil in a citrus orchard. *P. nicotianae*, *P. citrophthora*, and *P. palmivora* are most abundant and substantial species causing citrus diseases (Ippolito et al., 2004; Naqvi, 2006). *P. citrophthora* formed oomycete triggers inflammation, producing gum exudation from the trunks and branches (Alvarez et al., 2008).

Ecology, disease cycle and epidemiology

A single sporangium releases 5-40 zoospores. The production and germination of sporangium depend on the soil, water and potential temperature. The zoospores produced from sporangia are motile and show flagella movement in the water at a short distance and are carried to a longer distance through soil water. Root exudates attract these zoospores, and they swim toward them and encyst upon contact. The cyst, upon germination, penetrates the cortex directly or through wounds, natural openings. Zoospores can infect any part of the plant if the surface remains wet for at least 18 hours. It can form a germ tube. Savita and Nagpal (2012) explained the disease cycle of *P. parasitica* (*P. nicotianae*). They stated that the cycle begins with sporangia forming, which further releases many zoospores (that are motile), oospores, and chlamydospores in favorable conditions. Over time, the zoospores encyst, develop and germinate to form the mycelium. Mycelia requires an optimum temperature of 30-32°C for appropriate growth. Small deficit in meteoric water potential of -5 to -70 KPa, favors the sporangial growth. Depletion of nutrients and light stimulates sporangial formation. It can germinate from the germ tube under moist conditions, but indirect germination is crucial in gummosis and root rot disease. Further, sporangia require low temperatures and accessible water to produce zoospores. During unfavorable conditions, such as low oxygen levels, nutrient depletion, and low temperature (15-18°C), chlamydospores are produced. Below 15°C, chlamydospores enter the dormant state and germinate when the temperature rises to 27-30°C. During

infection, the pathogen establishes an intimate relationship with its hosts by forming haustoria, redirecting host metabolism and suppression (Alvarez et al., 2008). Choudhari et al. (2018) studied *Phytophthora* spp., outbreaks in four commercial citrus orchards to see if environmental and soil variables were linked to *Phytophthora* spp., root rot disease. Rainfall, temperature, relative humidity, soil moisture, soil pH, and soil EC all affected disease development. With the rise in rainfall and soil moisture, disease development was substantially advanced. The illness progressed well into October after a considerable rise in the second fortnight of August. There was a positive association between disease development and rainfall, soil moisture, relative humidity, soil EC, and an inverse correlation with air temperature (Benson et al., 2006). When the soil temperature falls below 12°C, citrus root development ceases; at this point, *P. nicotinae* reproduces chlamydospores and goes dormant to withstand the adverse conditions. Later, as the temperature rises again in the spring, the roots begin to develop, and the population of *P. nicotinae* increases. Because the roots develop all year in tropical locations, the seasonal variation in plant sensitivity to fungal infection is less noticeable (Cacciola and di San Lio, 2008). *P. parasitica* has been found in different ecological niches on five continents (Meng et al., 2014). *Phytophthora* spp., has a vast range and habitat present in solanaceous crops as well as several vegetables and forest trees, watersheds, medicinal herbs, mountain ecosystems, and natural ecosystem and in recycled irrigation water system (Prigigallo et al., 2015; Hulvey et al., 2010; Beever et al., 2009; Vannini et al., 2009). *P. nicotiana* is associated with loamy soil. The amount of the pathogen significantly higher in ten years old plants than in the five year old ones (Jung et al., 2000).

Diagnosis of *Phytophthora* spp. causing gummosis

For managing the disease, first step is to detect the reason behind the disease. It is necessary to detect the *Phytophthora* specie and its level present. Different methods have been developed for the detection of *Phytophthora* species. The baits method determines the quantity of *Phytophthora* inoculum in the citrus orchard soil. For this purpose, how frequently a ripped fruit present on the ground becomes infected is checked. Ripen lemon and sweet orange are used mainly to capture the *Phytophthora* from the soil (Ippolito et al., 2002; 2004). Leaf fragments of different citrus cultivars

are used as universal baits as they can capture all the species of *Phytophthora* present in citrus orchards. It is a highly sensitive method for molecular detection that can detect a very low population density of *Phytophthora*. Against *Phytophthora*, genus-specific monoclonal and polyclonal antibodies are produced. Enzyme-linked immunosorbent assay (ELISA) is developed to detect pathogens in soil debris and roots. The ELISA tests are not specific, so to confirm it, PCR is performed (Ippolito et al., 2002; 2004). Meng and Wang (2010), demonstrated specific primers for validating *P. nicotiana* against different fungal species. These primers detected *P. nicotiana* in citrus tissues, tobacco tissues, soil samples, and water suspensions. Therefore, Loop-mediated isothermal amplification (LAMP) was recently described as a specific, rapid, cost-effective, and easy-to-use method for nucleic acid amplification technology (Tomita et al., 2008). LAMP has been successfully developed to detect several *Phytophthora* species, including *P. nicotiana* (Li et al., 2015).

Management strategies

Citrus Gummosis can be managed by different strategies mentioned in Table 1 and Figure 1.

Agronomical management

To avoid the introduction of a fungus, the seeds to be planted are treated at 50°C for about 10 minutes. Using a medium also removes most *Phytophthora* problems at the nursery stage. The place should be chosen carefully, keeping it away from the infected region. The land needs to be fumigated before it can be used again. Replanting with methyl bromide or metam-sodium can decrease the inoculum at that location (Savita and Nagpal, 2012). Following sanitary practices, nursery and greenhouse production is kept free from *Phytophthora* spp. Soil preparation is the first and most crucial stage in managing soil-borne fungus through cultural management. Collar burying should be avoided when planting young trees (Schillaci and Caruso, 2006). Soil solarization is an efficient method of combating soil-borne pathogens. Next comes the management of the irrigation system. Use clean irrigation water (contamination free), keep the trunk dry, avoid flood irrigation, and provide proper water drainage. It is necessary to avoid premature irrigation during the spring season when the roots are inactive. Graham and Timmer, (2006) reported that removing soil from the collar region creates unfavorable habitat for the pathogen because

doing this prevents the bark at the foot region from remaining wet. It also helps the canker to heal up. They also mentioned that removing weeds around this region dries the area since the weeds prevent the bark from drying. To remove weeds, herbicides are used as they kill weeds and reduce the risk of wounds on the trunk, which might provide the penetration point to

the pathogen. Cacciola and di San Lio (2008), reported that mechanical tilling should be avoided, as it buries the trunk in the soil and makes it prone to infection. Deep tilling damages the roots and makes them susceptible. Grass growing between the rows during winter prevents the pathogen from coming in contact with fruits and leaves.

Table 1. Different strategies for the management of citrus gummosis.

Types	Action	References
Agronomical practices	Selection of areas for planting; practices of soil conservation; organic and mineral fertilization in adequate quantity; use of healthy seedlings with good genetic, agricultural, and phytosanitary characteristics; windbreaks; management of weeds. Fumigation or steam for thermal treatment of the lesions.	Graham and Feichtenberger, (2015); Savita and Nagpal, (2012)
Biological Management	Use of natural enemies of the pathogens.	Jagtap et al. (2012a, 2012b)
Chemical Management	Fungicides and resistance inducers made with calcium and potassium phosphite.	Gade and Koche, (2012)

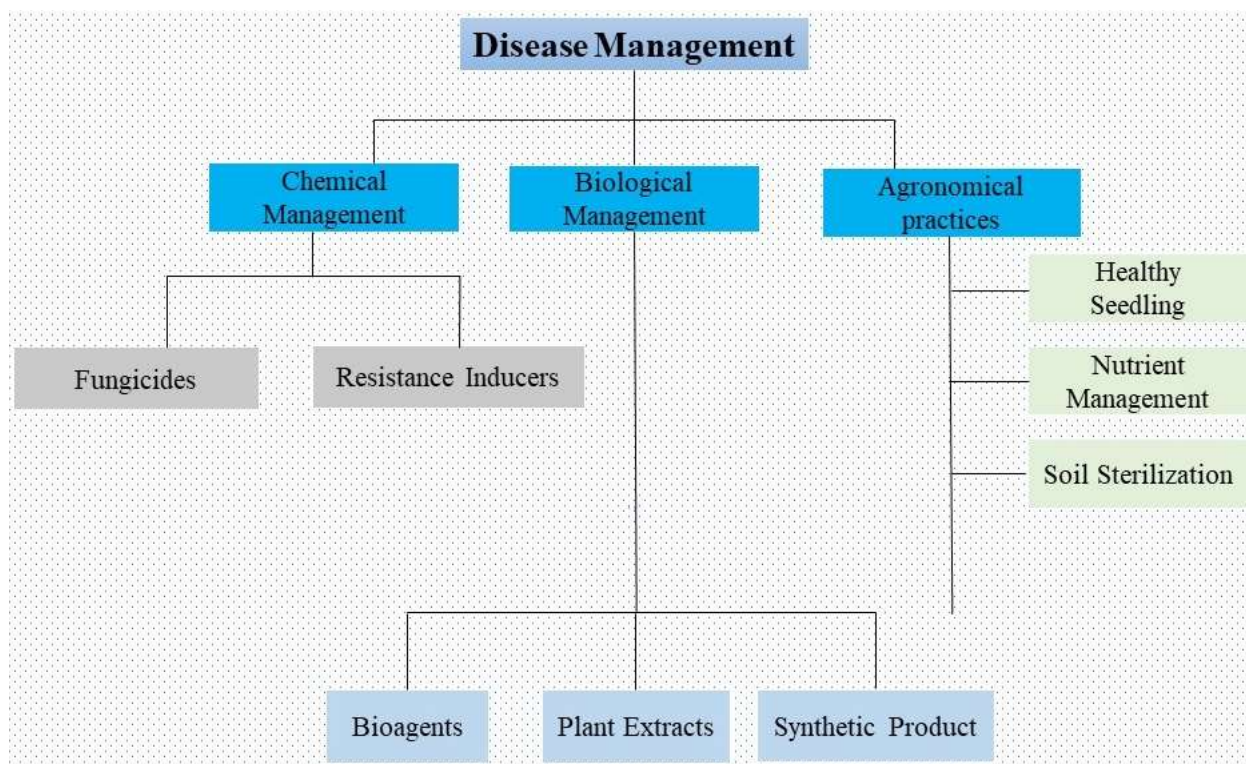


Figure 1. Disease management practices to manage the citrus gummosis.

Biological management

The bioagents produce systematic resistance in plants, which aids in disease resistance. Some plant species are also used to treat gummosis. Using leaf extract of neem,

eucalyptus, Acacia, Glyricidia, Dhatura, *Lawsonia inermis*, and bioagents such as *Trichoderma viride*, *T. hamatum*, *T. harzianum*, *T. lignorum*, *Gliocladium virens*, and *Pseudomonas fluorescens* against *Phytophthora* spp. has

shown promising results in the management of plant diseases caused by these pathogens. For biocontrol, a dual culture technique was used. *In vitro* study results showed that biocontrols significantly inhibited the mycelial growth of *P. citrophthora*. *T. harzianum* gave maximum inhibition, followed by *T. atroviridae*. *T. viridae* gave maximum inhibition (Jagtap et al., 2012a). Lantana was the most effective in inhibiting fungal growth *in vitro* conditions. *T. harzianum* is highly influential in bioagents, blocking mycelial growth (Jagtap et al., 2012b). Melo et al., (2017) evaluated the antagonistic properties of *Trichoderma* spp., and alfalfa seedling bioassay against *P. nicotianae*. Bairwa et al., (2015) conducted a field experiment to manage gummosis disease in kinnow mandarin. They used bioagents (*T. viride* and *P. fluorescens*) with Bordeaux paste. They noted a decrease in lesion size and minimum fruit dropping. The stem was painted with Bordeaux pastes and *T. harzianum*, and *P. fluorescens*. The reduction in symptoms was mild to moderate. Gade and Koche (2012) recorded a reduction in disease intensity of gummosis in Nagpur mandarin using *P. fluorescens* and Bordeaux mixture. Sadeghy et al. (2014) conducted an experiment in Kerman where they used 20 isolates of *Streptomyces* spp., from soil against gummosis. Quyet et al. (2016) used a saprophytic mesophilic fungus named *Chaetomium globosum* that resides on plants, straw, soil and dung against citrus gummosis. They mentioned in their studies that this fungus produces Chaetoglobosin-C, which has high inhibition abilities.

Chemical management

Management of these diseases is necessary to reduce yield losses and maintain fruit quality. Chemical control is the most efficient and successful method of eradicating illness and saving the crop. Various fungicides are used to treat various fungal infections which is the most widely utilized technique of disease control. Metalaxyl combined with mancozeb is the best systematic and proactive fungicide that inhibits mycelial growth and kills oomycetes (Rather et al., 2012). Gade and Koche (2012) suggested metalaxyl paint to be most effective in treating gummosis in Nagpur mandarin. Metalaxyl paint, in combination with Bordeaux paint, inhibits the production of oospores and chlamydospores quickly and quickly recovers the tree from disease. Rather et al. (2012) evaluated ten fungicides; among them, thiophanate methyl gave efficient results against *Phytophthora* spp. Iqbal and coauthors evaluated three

concentrations of different fungicides such as Topsin-M, Success, copper oxychloride, and cumulus, against *Phytophthora* species causing a citrus decline (Iqbal et al., 2020), among these, Topsin-M showed the best results because it is a broad-spectrum systematic fungicide followed by Success, kumulus and copper oxychloride. However, future studies are needed to adopt an integrated disease management (IDM) strategy, including using *Phytophthora*-free nursery, resistant rootstocks, and economical phytochemical fungicides to manage citrus gummosis disease best.

CONCLUSION AND FUTURE PERSPECTIVES

In conclusion, citrus gummosis, caused primarily by *Phytophthora* spp., is a significant disease affecting citrus orchards worldwide. It is a soil-borne pathogen that attacks the roots and trunk of citrus trees, leading to gum formation, root rotting, and decline of the trees. The disease has a long history, with outbreaks documented as early as the 19th century. *Phytophthora citrophthora* and *P. nicotianae* are the most commonly identified species causing gummosis in citrus-growing regions. Citrus gummosis poses a major challenge to citrus production, resulting in significant economic losses. It is responsible for yield reductions and even the complete decline of orchards. The disease spreads rapidly and establishes itself in infected fields, making it difficult to manage. Environmental conditions such as high humidity and temperatures favor the development of gummosis, further exacerbating its impact. Efforts have been made to understand the etiology, ecology, and pathogenicity of *Phytophthora* spp., associated with gummosis. Researchers have studied the morphology, behaviour, and pathogenicity of these fungi and their phylogenetic relationships. Various control measures, including fungicides and cultural practices, have been explored to manage gummosis and reduce its impact on citrus orchards. In the future, further research is needed to develop more effective and sustainable strategies for controlling and preventing citrus gummosis. This could involve the development of resistant citrus varieties, improve cultural practices, and identifying biological control agents. Continued surveillance and monitoring of *Phytophthora* spp., populations and their pathogenicity are essential to staying ahead of new strains or emerging species. Furthermore, raising awareness among citrus growers about the importance of early detection, proper sanitation practices, and integrated disease management

approaches can be crucial in minimizing the spread and impact of citrus gummosis. Addressing the challenges of citrus gummosis requires a multidisciplinary approach involving collaboration between researchers, citrus growers, and agricultural extension services. Combining knowledge, expertise, and resources makes it possible to develop sustainable strategies to mitigate the impact of this destructive disease on citrus production.

AUTHOR'S CONTRIBUTION

YI outlined and finalized the review, MM wrote initial draft; AS helped in figure preparations; AA used software and performed editing, MAZ validated the manuscript, US made necessary corrections; KA collected the literature.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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