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Journal of Citrus Pathology

REVIEW

Citrus Phantom Disorders of Presumed Virus and Virus-like Origin: What Have We Learned in the Past Twenty Years?

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Abstract

In the process of recording and studying citrus diseases in the 20th century, citrus pathologists reported several suspected graft-transmissible disorders that were thought to be of virus or virus-like origin. While later work clarified and characterized most of these disorders, others were left unaddressed beyond their initial reports, and their status has remained unresolved for decades. For this reason, and for lack of a better term, such disorders are considered "phantoms". In this work, our group performed an extensive literature review and communicated with renowned citrus pathologists and members of the International Organization of Citrus Virologists from around the world. Here we summarize and present in an organized manner the most up to date information for multiple phantom disorders, including disorders that have been subsequently characterized as a result of research efforts of the past 20 years. This review article could act as a reference point for citrus pathologists, regulatory agencies, and industry to clear up any confusion regarding citrus phantom disorders.

Keywords: citrus disorders, citrus pathology, citrus virus, graft-transmissible, causal agent, disease etiology

Introduction

Citrus represents one of the most widespread fruit crops in the world. Different varieties of citrus are grown in more than 140 countries with suitable tropical or subtropical environments, with the five major citrus-producing countries being China, Brazil, the United States of America (USA), Mexico, and India (FAO, 2021). In certain countries with substantial production, the citrus industry is usually a major contributor to the economy. For instance, in 2020-21, the citrus industry in Florida, USA generated an estimated total of $6.935 billion in industry output contributions to the state economy, with $5.334 billion from citrus juice manufacturing, $1.425 billion from citrus fruit production, and $177 million from fresh citrus marketing, and supported a total of 32,542 jobs in the state with $1.606 billion in labor income contributions (Cruz et al., 2023). In California, the total economic impact of the citrus industry was valued at $7.6 billion and supported a total of 24,247 jobs (Babcock, 2022).

Citrus production can be affected by various pathogens. Among those, graft-transmissible pathogens of viral or bacterial nature continue to cause significant damage to the citrus crop worldwide (Timmer et al., 2000; Talon et al., 2020). Over the decades, the scientific contributions of many citrus pathologists have greatly enhanced our knowledge of citrus disorders and their causative agents. This knowledge has enabled the citrus industry to mitigate the damage and spread of devastating citrus diseases. "The Compendium of Citrus Diseases (2nd ed.)" (Timmer et al., 2000), "The Citrus Industry (Vol. IV)" (Reuther et al., 1978), and other reviews (Knorr 1968; Childs 1968; Knorr 1973; Klotz, 1973), provided a comprehensive description of citrus diseases and disorders by presenting the information available at the time of their publication. However, several suspected virus, virus-like, or undetermined etiology graft-transmissible disorders affecting *Citrus* species made only a few appearances in the literature and were never mentioned afterward, while recurrences of these diseases or disorders on the field have not been reported, and greenhouse positive controls and type isolates are not known to exist today. For lack of a better term, such disorders are herewith termed as "phantoms".

Phantom disorders lack published work associating them with a pathogen or an abiotic cause. Some of these disorders cannot be studied beyond their original reports either due to the loss of type isolates or due to their elimination from citrus plantings following the use of pathogen-free propagative materials. These can, perhaps, be considered true phantoms, as we might never know what caused them. Other disorders, which were previously considered phantoms, have been subsequently attributed to known causal agents or conditions or, at least, shown to be abiotic or non-viral in nature as a result of research efforts in the past 20 years. However, some of these former phantoms still lack the published work declaring these new findings to the wider citrus community. As a result, they continue to be regarded as phantoms or of unknown etiology simply because the new knowledge has not been published and disseminated. Ultimately, there are a number of either true or former citrus phantom disorders, which can still cause confusion in the scientific, grower, and regulatory communities, and there is a need to update their information and clarify their status.

In an effort to disseminate in an organized fashion the most up to date current knowledge for the citrus phantom disorders and direct possible research, bibliography, policy, and regulation, our group performed an extensive literature review and contacted several renowned citrus pathologists and members of the International Organization of Citrus Virologists (IOCV) from different countries, for any related information on over 100 potential phantom cases reported from various citrus-growing regions of the world. The outcome of our efforts is presented in this review article, with a list of 55 citrus phantom disorders that should be disregarded and not used in the scientific literature as they cannot be considered as being associated with a pathogenic etiology or should have their names updated to reflect today's knowledge and their association with currently known pathogens or abiotic factors.

A. Phantom citrus virus-like disorders reported in multiple citrus regions of the world

1. Gum pocket/gummy pitting/wood pitting

These names all correspond to the same type of trifoliate orange (*Citrus trifoliata* L. (syn. *Poncirus trifoliata* (L.) Raf.)) rootstock disorders, which have been reported from many citrus growing regions of the world, in some cases, in correlation with citrus viroid infection (van Vuuren & da Graca, 1996; Duran-Vila et al., 2002). The trunk of the affected trees exhibited gum-filled pits, with gumming present in both the phloem and the xylem. Replicated field trials with pathogen-free rootstocks and scions have demonstrated that such symptoms could appear in trees inoculated with viroids and also in non-inoculated control trees. Therefore, Koch's postulates have not been fulfilled for citrus viroids as causal agents of gum pocket and gummy and wood pitting. These disorders are possibly of physiological or stress origin that could be enhanced by viroid infection (Duran-Vila et al., 2002; Vernière et al., 2002, 2004).

2. Measles

Citrus leaves exhibiting pale yellow spots were described in Florida, USA and Brazil (Knorr 1973; Lee et al., 1993). No double-stranded RNA (dsRNA) or virus particles were associated with the disorder, but some evidence of graft-transmissibility was presented (Lee et al., 1993; Lee, 2015). An abstract reported trees in California, USA putatively exhibiting symptoms associated with measles. However, laboratory tests, including dsRNA analysis and transmission electron microscopy, did not identify any etiological agent (Lee et al., 2008). In addition, two bioindexing experiments in California (CCPP #3023 and USDA-ARS #08-10.27) produced no symptoms on four different citrus indicator species (Vidalakis & Yokomi, personal communications), in agreement with earlier multi-year graft-transmission experiments reported from Florida, USA, indicating the non-infectious nature of this disorder (Knorr, 1968; Knorr 1973). Measles is still occasionally observed in South Africa (Glynnis Cook, personal communication), but since no clear association with a viral or graft-transmissible agent has been identified, and it is not known if a type isolate is available or whether any samples have been preserved, measles can be considered as a phantom disorder.

3. Popcorn psorosis

This disorder was reported in sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.) plants from São Paulo, Brazil, by Rossetti and Salibe (1965). According to the authors, a similar disease referenced with a similar name was also identified in Florida, USA by J. F. L. Childs and in Setubal, Portugal by V. Rossetti in the 1960s. The popcorn psorosis-like symptoms were characterized by the presence of small (less than 1 cm in diameter) scales or flakes on the outer bark that often were loose, tearing apart, and leaving small pustules. Gum exudation was also frequently observed (Rossetti & Salibe, 1965). In 2012, some old Navel sweet orange trees grown on rough lemon (*Citrus* × *granulata* Raf.) rootstock in South Africa exhibited popcorn psorosis-like symptoms. The original trees were removed. However, bark-inoculated 'Madame Vinous' sweet orange trees have been maintained in South Africa, and no clear association with a viral/graft-transmissible pathogen was identified (Glynnis Cook, personal communication). Furthermore, as suggested by Pedro Moreno (personal communication), the term "popcorn psorosis" should not be used to refer to this disorder, since there has been no association of the syndrome with citrus psorosis virus (CPsV), which causes psorosis, and its graft transmissibility was not demonstrated.

4. Shellbark

This disorder is typically characterized by cracking and peeling of the bark, and it was reported in approximately 10-year-old trifoliate orange and Eureka lemon (*Citrus* × *limon* (L.) Osbeck) trees in Australia (Olson, 1968; Broadbent & Dephoff, 1992). However, similar symptoms were detected in other citrus genotypes and in different countries, usually, in association with one or more viroids, mainly, citrus exocortis viroid (CEVd) (Fernandez-Valiela, 1961; Olson, 1968). However, the causal agent was not definitively determined. Currently, shell bark is not considered a distinct disease, but it could have been a result of mixed viroid infections, including CEVd. Although it was originally widespread in Eureka lemon cloned trees in some regions of Australia, the adoption of trees propagated from polyembryonic seeds reduced the incidence of symptomatic trees (Broadbent & Dephoff, 1992). There are no type samples available in Australia or other places where shell bark has been reported, i.e., Argentina (Fernandez-Valiela, 1961), where the disease has not been found for decades (Julia Figueroa, personal communication). Therefore, further examination of this disorder is not possible, and it should not be referenced as a specific graft-transmissible or viral disease of citrus.

5. Rumple of lemon

This disorder was reportedly observed in Sicily, Italy since the early 20th century (Salerno, 1963). However, the first report of the disorder was from Florida, USA (Knorr,1958). Since then, Rumple has been reported in many Mediterranean countries (Del Rivero 1967; Knorr & Koo 1969; Ozbek et al. 1976) and to date remains a problem in many lemon-producing regions in the Mediterranean (personal communications: Antonio Ippolito; Megan Dewdney; Moshe Bar-Joseph). The fruit symptoms have variable distribution patterns, which can either be spots that start as slightly sunken faint chlorotic specks or rings of an area of about 4-5 oil glands. These specks continue to sink and change color until the oil glands collapse, and the lesions become necrotic. These lesions can form vermiculate networks on the rind (Knorr & Koo, 1969). The etiology of this disorder remains unknown. There have been several attempts to ascertain the graft-transmissibility of the disorder, which led to inconclusive results (Cartia & Catara, 1974; Majorana & Continella, 1984). Attempts to attribute the disorder to nutrient deficiencies were also inconclusive (Salerno et al., 1968; Knorr & Koo, 1969; Ozbek et al., 1976). The yearly fluctuation of the incidence of rumple has led some to postulate that climatic conditions could be responsible (Knorr & Koo 1969; Alarcón et al. 1996; Del Rivero 1967). Recently, fungal populations were reported to be associated with the disorder (Valero et al., 2010). However, a fungal infection seems to be an association or a secondary infection of weakened tissue rather than the causative agent of the disease. Therefore, rumple of lemon should not be referenced as a virus-like, graft-transmissible citrus disease. The disorder requires further investigation to assess its possible association with a pathogen or, alternatively, to confirm its physiological, nutritional, or fungal nature. In the latter case, Koch's postulates should be completed to establish a causal agent-disease relationship.

6. Citrus dieback disease, Leaf mottle yellows disease, and Citrus vein-phloem degeneration

These disorders were described by J. M. Wallace in the chapter "Virus and virus-like diseases" of "The citrus industry-Vol. IV" based on reports from India, the Philippines, and Indonesia (Wallace, 1978). After a careful review of the symptoms described, electron microscopy finds, vector transmission, and antibiotics experiments, it can safely be concluded that the various names used in the referenced publications for these disorders all describe the citrus greening or the Huanglongbing (HLB) disease of citrus. Therefore, these names should not be used in conjunction with the description of such phenotypic disorders, which actually represent manifestations of the HLB disease of citrus.

7. Citrus crinkly leaf virus

Citrus crinkly leaf was reported as a disorder with vein-flecking, circular clear spots, and crinkled leaf symptoms (Yot-Dauthy and Bové, 1968). However, such symptoms resembled those caused by citrus variegation virus (CVV, syn. infectious variegation virus of citrus, Grant and Smith, 1960). A series of studies in the 1960s, which included graft-transmission and mechanical inoculation of various citrus and herbaceous indicators, as well as virus purification experiments, indicated that citrus crinkly leaf virus (CCLV) and CVV are closely related, with virtually identical properties (Dauthy and Bové, 1965; Majorana and Martelli, 1968; Yot-Dauthy and Bové, 1968). Therefore, CCLV and CVV could be considered strains of the same virus. In addition, one of the CCLV isolates used in the studies in the 1960s was identified as the "California strain 81-A-65 obtained from Dr. J. M. Wallace" (Dauthy and Bové, 1965; Yot-Dauthy and Bové, 1968) or as the "CCLV isolate from California derived from a naturally infected seedling" (Majorana and Martelli, 1968). This isolate is still available at the Citrus Clonal Protection Program (CCPP) at the University of California, Riverside under the name "IV-400-Crinkly leaf: Original crinkly leaf source from seed transmitted seedling". The CCPP record for IV-400-Crinkly leaf indicates that this source was introduced to the CCPP on 5/16/1962 by Dr. J. M. Wallace from a seed transmitted source (one lemon seedling out of 982 with pin-point spotting, planted on 11/7/1952; buds originally from a Eureka lemon in Davey grove collected by Dr. Fawcett) (Wallace, 1978). The CCPP record also indicated that the IV-400-Crinkly leaf source is a direct inoculation of the 81A-65 plant (Experiment No 81A- plant No 65) that transferred inoculum from the original lemon seedling to sweet orange on 10/5/1953. The CCPP IV-400-Crinkly leaf isolate was recently sequenced, and the three viral RNAs had very high sequence similarity with CVV (GenBank Assembly GCA_000870745.1, G. Vidalakis-CCPP, personal communication). Therefore, CCLV should be considered synonymous with CVV.

B. Phantom citrus virus-like disorders reported in the USA

8. Milam lemon stem pitting

This disorder was identified in the early to mid-1970's in Florida, USA and was characterized by severe stem pitting symptoms observed in some Milam lemon seed-source trees (Garnsey, 1973). Symptoms were similar to those associated with tristeza, although citrus tristeza virus (CTV) was not found in all symptomatic trees, and transmission tests were inconclusive. Some mandarin hybrids also exhibited milder pitting. Even in symptomatic trees, vigor was not affected, and the disorder never became a problem (Ferguson & Garnsey, 1993). Milam lemon stem pitting has not been seen in the field in the past decades, and to the best of our knowledge, no type isolate is available.

9. Fovea

This disorder was initially reported in Florida, USA on Murcott tangor (*Citrus* × *aurantium* L. var. *chrysocarpa* (Hassk.) ined.) and other mandarin hybrids showing tree decline and inverse stem pitting (Timmer et al., 2000). Symptoms reported were similar to those of the viroid induced cachexia on Murcott. There has been no mention of Fovea following the initial report, and it is not known if a type isolate is available or whether any samples have been preserved.

10. Grapefruit bark scaling

Bark scaling symptoms were observed on grapefruit (*Citrus* × *aurantium* L. var. *racemosa* (Risso) ined.) in Florida, USA. The disease was non-graft transmissible, and the association with a specific virus was not found (Timmer et al., 2000). There have been no further reports of the disease, and it is not known if a type isolate is available.

11. Failure of Rangpur lime on sweet orange

A series of experiments between 1958-1961 in California, USA reported a failure of Rangpur lime (*Citrus* × *limon* (L.) Osbeck) scions grafted on sweet orange rootstocks. The syndrome was not observed in Rangpur lime grafted on sour orange (*Citrus* × *aurantium* L.) or in rooted Rangpur lime cuttings (Frolich, 1958). Similar failure was observed in the citron (*Citrus medica* L.) scions grafted onto sweet orange (Frolich & Hodgson, 1961). The causal agent was found to be graft-transmissible. In addition, the growth of the sweet orange rootstock was markedly repressed, compared to that of the scion. There have been no further reports on the disease, and, to the best of our knowledge, no type isolate is available.

12. Mechanically-transmitted citrus ringspot virus (CRV-MT)

CRV-MT was first described when indexing 'Zatima' Navel sweet orange trees from an old citrus variety collection in Florida, USA on herbaceous indicator plants (Garnsey, 1975). CRV-MT produced varied symptoms on herbaceous indicator hosts and could be mechanically transmitted to citrus from cucumber, red kidney bean, and citrus. In citrus, the symptoms observed were similar to those previously described to be produced by citrus ringspot virus (CRV) (Wallace & Drake, 1968). At the time, because of the similarity to the previously described CRV, the virus was termed CRV-MT, since CRV was considered not mechanically transmissible. In a later publication, the same author cited the original 1975 paper when describing the isolate as CRSV-2 (Garnsey & Timmer, 1980). It appears the author of the original paper has re-characterized CRV-MT as an isolate of CRSV (presently referred to as CPsV) rather than a distinct virus. As such, CRV-MT should be considered an isolate of CPsV.

13. Algerian navel orange virus (ANOV)

ANOV was originally reported based upon indexing of asymptomatic Algerian navel sweet orange trees from an old citrus variety collection in Florida, USA on herbaceous indicator plants (Garnsey, 1975). The inoculated leaves of *Chenopodium quinoa* and *Crotalaria spectabilis* developed chlorotic mottle symptoms and local lesions, respectively. The suspected virus could be mechanically re-inoculated back into citrus and then reintroduced into the indicator plants. In the herbaceous hosts, the pathogen consistently induced the symptoms observed previously. However, no symptoms were seen in citrus. Additionally, electron microscopy revealed 780 ± 25 nm long, flexuous, rod-shaped particles in the samples taken from both citrus and herbaceous hosts (Garnsey, 1975; Timmer et al., 2000). Apart from the original report, there is no other mention of ANOV, and, to the best of our knowledge, no type isolate is available today.

14. Dweet mottle virus (DMV)

DMV was first reported from Riverside, California, USA in 1968 during reindexing of a Cleopatra mandarin (*C. reticulata* Blanco) variety at the Citrus Variety Improvement Program, which is now referred to as CCPP. DMV produced leaf-mottling symptoms similar to but distinct from the symptoms of psorosis and concave gum in 'Dweet' tangor (*C. reticulata* Blanco x *C. sinensis* (L.) Osbeck) (Roistacher et al., 1968). Full genome sequence analysis of the CCPP-DMV isolate revealed high nucleotide identity with the citrus leaf blotch virus (CLBV), the single species within genus *Citrivirus*, family *Betaflexiviridae*. Hence, DMV should be considered a CLBV isolate (Vives et al., 2005; Hajeri et al., 2010).

C. Phantom citrus virus-like disorders reported in Japan

15. Bark pitting (inverse pitting)

This disorder of Satsuma mandarin is a rare disease of unknown etiology that was reported in a 1961 publication from Japan (Tanaka & Yamada, 1961). Pits were observed on the cambial side of the bark, with protruding small pegs on the opposing side of the wood. The affected trees suffered from severe defoliation. However, similar symptoms have been observed in different citrus varieties suffering from stress and poor rootstock-scion compatibility (Pedro Moreno, personal communication). No other work has been reported for this disorder, and, to the best of our knowledge, no type isolate is available.

Therefore, further characterization and comparison to known graft-transmissible diseases of citrus is not possible. Since citrus trees in Japan are widely infected with CTV, it is possible that the symptom results from a combination of a CTV isolate, host interaction and specific climatic condition.

16. Citrus yellow mottle

This disorder was first reported in the Kanagawa prefecture in Japan (Ushiyama et al., 1984). The affected Satsuma mandarin tree leaves exhibited distinct vein clearing with yellowish halos. Only three affected trees in two citrus orchards were found. The symptoms were consistently reproduced when multiple citrus species were graft-inoculated with the bark from the affected trees. Mechanical transmission using crude sap to citrus and herbaceous hosts was unsuccessful. However, mechanical transmission was achieved with back inoculation to Fukuhara sweet orange seedlings by stem-cut method with the 100,000xg sediment fraction prepared from symptomatic trees. Rod-shaped particles (690-740 nm X 12-14 nm) were consistently observed with the electron microscope from partially purified preparations and leaf-dips from symptomatic leaves. Ultra-thin leaf sections also revealed aggregates of similar rod-shaped particles. Consequently, the identified agent was termed citrus yellow mottle virus (CYMV) (Ushiyama et al., 1984). In more recent experiments, the researchers observed larger filamentous particles (about 900 X 14 nm in size), similar to those of CLBV, in citrus samples collected in Japan, but since the disease has not re-appeared in the Kanagawa prefecture and previously collected isolates were lost, these studies could not continue (Toru Iwanami, personal communication). In order to avoid confusion with the citrus yellow mottle-associated virus (CiYMaV, GenBank MK957246) recently reported from Pakistan (Wu et al., 2020), the disorder and the virus name, i.e., citrus yellow mottle and CYMV, should be retired from the literature. If these names need to be used, they should be accompanied with additional information such as the original host and origin (e.g., Satsuma citrus yellow mottle from Japan or CYMV originally reported from Satsuma in Japan) to ensure the differentiation from the CiYMaV. If the disease reemerges in Japan, more studies can be conducted to verify the identity of its causal agent as CLBV or a novel virus and rename the disease using any new information.

17. Oleocellosis-like symptoms of Satsuma orange, Summer orange dwarf (a.k.a. Satsuma dwarf-like disease on Citrus natsudaidai in Yamaguchi prefecture), citrus mosaic, and Natsudaidai dwarf

Oleocellosis-like symptoms of Satsuma orange and Summer orange dwarf were reported in a 1961 publication from Japan (Tanaka & Yamada 1961). Biological indexing experiments described in the publication indicate that these disorders were associated with single or mixed infections of CTV and satsuma dwarf virus (SDV). Oleocellosis-like symptoms of Satsuma mandarin and Summer orange dwarf apparently correspond to citrus mosaic and Natsudaidai dwarf, respectively (Toru Iwanami, personal communication). Citrus mosaic virus (CiMV) and Natsudaidai dwarf virus (NDV) share high amino acid identities (over 75%) with SDV, the only member of the genus *Sadwavirus*. Therefore, CiMV and NDV are classified as strains of SDV, and thus, they are no longer individual virus species (Le Gall et al., 2005; Iwanami, 2010; Iwanami, 2023). It has been proposed that CiMV and NDV should be referred to as “citrus mosaic strain” and “Natsudaidai dwarf strain” of SDV, respectively (Toru Iwanami, personal communication). This disorder should not be confused with the citrus mosaic disease reported from India (Ahlawat et al., 1985).

18. Hassaku dwarf

This disorder was described in Japan (Tanaka & Yamada, 1961). A careful examination of the symptoms described in the manuscript and the reactions of the bio-indexing indicators used in the study suggested that the disorder was caused by CTV. In a more recent report, Hassaku dwarf was recognized as a form of tristeza stem-pitting (Timmer et al., 2000). Therefore, Hassaku dwarf should not be considered a true phantom disease, but rather a disease syndrome used when describing CTV from Japan.

D. Phantom citrus virus-like disorders reported in Brazil

19. Leaf curl

A small number of citrus trees in Brazil showed leaf curling, dieback of branches, and decline (Salibe, 1959; 1965). The causal agent was proposed to be of viral origin because it was graft transmissible to other citrus species where it also induced those symptoms. However, since the original report, there have been no other reports of the disease, and, to the best of our knowledge, no type isolate is available.

20. Bahia bark scaling (BBS)

BBS disease received some attention in the affected Brazilian regions of Bahia and Sergipe after being identified in sweet orange, tangerine (*Citrus* × *aurantium* L. var. *chrysocarpa* (Hassk.) ined.), and grapefruit trees in the 1960s (Passos, 1965; Passos et al., 1974; Nickel et al., 2007). The disease symptoms were often more evident in grapefruit and were characterized by darkening, swelling, and cracking of the bark. Scaling, dieback, and gum exudation could be seen in older trees. However, while the typical bark symptoms were evident in up to 100% of the plants in certain areas, no young-leaf symptoms were observed in natural or experimental indicator hosts (Nickel et al., 2007). Due to the similarity of BBS symptoms with those associated with psorosis, the disease was named Bahia-type psorosis (Nickel et al., 2007). But since CPsV is not its etiological agent, that denomination has been disregarded (Moreno et al., 2015). In 2006, Laranjeira et al. suggested that BBS was transmitted by air-borne vectors with limited dispersion ability, strengthening the hypothesis of a viral causal agent. Attempts to graft or mechanically transmit the disease have been unsuccessful (Pedro Moreno, personal communication). It was recently shown that BBS is caused by different fungi, specifically, *Lasiodiplodia iraniensis*, which was identified as the causal agent of the disease in grapefruit trees (Gama, 2019, Gama et al., 2019). For this citrus species, Koch's postulates were fulfilled, although other species of *Lasiodiplodia* and, sometimes, other fungi were associated with BBS in other citrus genotypes (Cristiane Barbosa, personal communication). Therefore, BBS is not a citrus viral disease, and it should be referred to as a fungal disease of citrus.

21. Zonate chlorosis

This disorder was described in 1934 and is restricted to the coastal regions of Brazil (Bitancourt & Grillo, 1934; Ramos-González et al., 2023). The disorder appears in a wide range of citrus hosts and can produce irregular chlorotic lesions on the leaves and fruits (Ramos-González et al., 2023). Recently, hibiscus green spot virus 2 was shown to be the causal agent of this disorder (Rodrigues, 2022; Ramos-González et al., 2023). Consequently, zonate chlorosis should be considered a distinct viral disease in citrus.

E. Phantom citrus virus-like disorders reported in India

22. Rubbery wood (RW)

This disorder was reported in India in the 1970s, reaching high incidence on limes (*Citrus* × *aurantiifolia* (Christm.) Swingle) and lemons (Ahlawat & Chenulu, 1985). As the name implies, the limbs of the affected trees became atypically flexible and bent downwards (Ahlawat & Pant, 2003). The disease was transmitted by grafting and dodder, and the presence of a phytoplasma has been associated with RW. However, a different phytoplasma, which causes witches´ broom disease of limes (WBDL), was identified in Oman and later in India (Garnier et al., 1991; Ghosh et al., 1999, 2013; EPPO, 2006). While WBDL is still present in the field, RW has not been found for several years. According to Dilip Ghosh (personal communication), "after the first report, no further detailed work on the same [RW] was done as it is not a serious pathogen reported on citrus". Due to the lack of information, the absence of the disease in the field for many years, and the lack of a type isolate, further characterization of this disease does not seem possible.

23. Leathery leaf

Leathery leaf symptoms were described in India (Ahlawat et al., 1979). The agent could be transmitted by aphids, grafting, and mechanically to citrus and herbaceous hosts. Symptoms appeared to be similar to those of SDV and psorosis-like vein clearing. There have been no further reports of the disease, and, to the best of our knowledge, no type isolate is available.

F. Phantom citrus virus-like disorders reported from other citrus-growing regions

24. Brittle twig yellows, Iran

This disorder of sweet orange trees has been reported only in an abstract describing its graft-transmissible nature and some similarities and differences with stubborn and tristeza stem pitting (Samadi et al., 1977). The symptoms were described as thick, breakable, and abnormally branched shoots with the overall trees becoming dwarfed, bushy, and yellow. No other work on this disorder has been reported, and, to the best of our knowledge, no type isolate is available. Therefore, further characterization and comparison to known graft-transmissible diseases of citrus is not possible.

25. Tarocco pit, Italy

An atypical, severe form of the concave-gum disease was observed on Tarocco orange (*Citrus* × *aurantium* L. var. *sinensis* L.) trees in Sicily in 1963 (Russo & Klotz, 1963), which was characterized by significant concavities or inferior depressions in the surface of the tree trunk and limbs. Circular holes ranging from 1 mm to 2 cm in diameter harboring a cork-like material were seen in the center of these depressions. Similar to the concave-gum disease, the affected Tarocco trees also displayed oak-leaf pattern and leaf flecking. To test whether the pitting observed on these Tarocco trees was bud-perpetuated, sweet orange indicators were graft-inoculated with budwood from the affected Tarocco trees. The sweet orange trees developed leaf flecking and oak-leaf pattern. However, the pitting effect was not reproduced. The authors acknowledged the presence of concave-gum virus in the affected trees and postulated that a more virulent strain of the virus or another virus in combination with concave-gum virus was the cause of tarocco pit. Whether the citrus concave-gum virus mentioned in Klotz and Russo (1963) refers to the citrus concave gum-associated virus (Minutolo et al., 2021) or citrus virus A (Navarro et al., 2018) is not clear. There have been no further reports on the disease.

26. Multiple sprouting, South Africa

This disorder was identified in South Africa in 1970. The disease was graft-transmitted from a single symptomatic field tree and maintained in a greenhouse in Nelspruit (1974-79); however, the isolate has since been lost (John da Graça, personal communication). Multiple sprouting was associated with the putative citrus multiple sprouting virus (Majorana & Schwarz, 1972), although it was not proven to be the causal agent. Multiple sprouting has been also reported in Satsuma mandarin trees in Turkey as a symptom of satsuma dwarf virus (SDV) infection (Onelge & Çinar, 2010). However, SDV has not been associated with the multiple sprouting disorder observed in South Africa. The symptoms were described as a dense proliferation of shoots, somewhat similar to witches' broom. Additionally, according to a personal communication from Glynnis Cook, based on the information gathered with extension advisors and people who have worked with citrus for years in South Africa, “multiple sprouting is no longer found in the field”. Sometimes such symptoms were associated with high numbers of thrips infesting the buds or, otherwise, with herbicides like 2,4-D on young trees, neither of which are graft-transmissible conditions. It is possible that through proper crop management this disorder was eliminated, but there was never a specific agent that could be associated with the condition. It has been also suggested that multiple sprouting is sometimes a cultivar trait found in certain citrus cultivars, for example, early Clementine or ‘Mor’ type mandarin.

G. Other phantom citrus virus-like disorders

A series of suspected citrus virus and virus-like diseases and disorders appeared in the literature between 1913 and 1972 (summarized in Knorr 1973; Knorr 1968; Childs 1968; Klotz, 1973) (Table 1, #27-55). However, these diseases did not appear in the literature after 1978 when a book series “The Citrus Industry” presented a comprehensive review of virus, virus-like, and graft-transmissible diseases of citrus (Wallace, 1978). Even if some of these disorders were truly of viral nature, the wide use of citrus therapy techniques such as thermotherapy (first applied successfully in 1957) and shoot-tip grafting (first applied successfully in 1972) most likely eliminated any viral pathogens associated with these disorders from citrus propagative materials (Roistacher, 1995). Furthermore, the establishment of registration programs for citrus budwood sources as early as 1937 and the establishment of comprehensive therapy and indexing citrus germplasm programs in many citrus-producing areas in the world by the 1970s would have limited the proliferation of such diseases even further (Hiltebrand, 1957; Mather, 1968; Mather and McEachern, 1974; Calavan et al., 1978). Finally, the progressive replacement of old citrus with pathogen-free propagative material, primarily due to problems with tree productivity or death during the global tristeza quick decline epidemics (Moreno et al., 2008), most likely contributed further to the lack of any additional reports of these disorders in the field. Furthermore, many of these disorders were described in the early 20th century. Therefore, while it was unknown at the time of the original report, some of these disorders could possibly be identical to other presently known citrus disorders.

Table 1
Suspected citrus virus and virus-like diseases and disorders appearing in the literature between 1913 and 1972, summarized in Knorr (1973); Knorr (1968); Childs (1968); Klotz (1973).

Disorder (Region)	**Affected citrus varieties**	**Distinctive symptoms**	**References**
27. Bark rot (China, Indonesia, Japan, Philippines)	• Tangerine, mandarin, and Satsuma mandarins (*Citrus* × *aurantium* L. var. *chrysocarpa* (Hassk.) ined.) • Calamondin (*Citrus* × *microcarpa* Bunge) • Caburao (*Citrus hystrix* DC.)	• Longitudinal bark cracks, which exude white foam • Tissue in the affected areas dies, eventually killing the tree	Fawcett (1936); Lee (1923)
28. Bark rot of sour-orange rootstock (USA-California)	• Sour orange (*Citrus* × *aurantium* L.) rootstocks of sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.)	• Vitreous gummy or resinous infiltrated degeneration of bark that extends to the wood	Fawcett & Klotz (1948)
29. Branch blight of grapefruit (USA-California)	• Grapefruit (*Citrus* × *aurantium* L. var. *racemosa* (Risso) ined.)	• Dieback and death of entire branches • Gum exudates along the margins of dead areas • Dark brown bark extending beyond the affected areas	Fawcett & Klotz (1948)
30. Bud-union constriction disorder of grapefruit on sour orange (Israel)	• Grapefruit (*Citrus* × *aurantium* L. var. *racemosa* (Risso) ined.) on sour orange (*Citrus* × *aurantium* L.) rootstocks	• General tree decline associated with swelling of the scion at the bud-union and a constriction in the wood around the line of union	Reichert et al. (1965)
31. Cancroid spot (USA-Florida)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.)	• Fruit lesions • Leaf spots • Small leaves • Defoliation • Dieback	Knorr (1968)
32. Chronic decline (USA-California)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.) on sour orange (*Citrus* × *aurantium* L.) rootstock	• Tree decline with necrosis of sieve elements and bark thickening below the bud union • Excessive fibers and crystal idioblasts • Hyperplastic woody, elongated fusiform rays • Similar to tristeza decline	Schneider (1957)
33. Chrysosis (Brazil)	• Sour orange (*Citrus* × *aurantium* L.)	• Yellow or golden spots on leaves resembling ringspots	Fawcett (1937)
34. Convex gum (China)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.)	• Bark swelling, which produces convexities of the surface with gum underneath	Lin (1943)
35. Crinkle-scurf (Morocco, USA-Florida)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.)	• Leaf twisting • Corky banding on bark	Knorr (1968)
36. Crotch disease of tangerine (Argentina, Paraguay, Uruguay)	• Tangerine (*Citrus* × *aurantium* L. var. *chrysocarpa* (Hassk.) ined.)	• Water-soaked regions in the crotches of main branches where bark is injured or killed • These areas can split open and rot	Fawcett & Bitancourt (1940); Fawcett & Klotz (1948)

37. Cyclosis (Brazil)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.)	• Light-yellow circular or elongated spots or rings on leaves • Gum pustules around and over older spots	Fawcett & Klotz (1948)
38. Ellendale decline (Australia, South Africa)	• Ellendale and Imperial mandarins (*Citrus* × *aurantium* L. var. *chrysocarpa* (Hassk.) ined.) on rough lemon (*Citrus* × *granulata* Raf.) rootstock	• Decline	Levitt (1955); McClean (1956)
39. Epidemic dieback of lime (Trinidad)	• Limes (*Citrus* × *aurantiifolia* (Christm.) Swingle)	• Occurrence of dead wood • Leaf yellowing and defoliation • Fruit Shriveling • Tree death • Characteristic ascending dead strips running up the trunk and branches from the roots • Distinct from tristeza quick decline: No stem pitting or vein clearing, and no disease in the adjacent sweet orange on sour orange trees	Lucie-Smith (1951)
40. Eruptive bud-union crease (Egypt, India)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.) on rough lemon (*Citrus* × *granulata* Raf.) rootstock	• Crease or protrusion at the bud-union line with various possible wood/bark abnormalities • Can induce decline of tops	Bhutani et al. (1972); Chadha et al., (1970); Nour-Eldin (1957)
41. Eruptive gummosis (Argentina)	• Gum eruption certain in Marsh grapefruit (*Citrus* × *aurantium* L. var. *racemosa* (Risso) ined.), uncertain in Perrine lemon (*Citrus* × *limon* (L.) Osbeck)	• Eruption of gum from trunk and stems • Gum-filled fruit lesions • Lines of gum in the wood underneath the bark	Pujol (1968)
42. Infectious mottling (Petri's) (Italy)	• Sour orange (*Citrus* × *aurantium* L.)	• White, pale green, or yellow irregular areas on leaves • Blistered and wilted leaves • Gummy degeneration of palisade cells in the white-colored areas	Fawcett (1936); Fawcett & Klotz (1948)
43. Juvenile spot (USA-Florida, Argentina, South Africa, Syria, Japan)	• Grapefruit (*Citrus* × *aurantium* L. var. *racemosa* (Risso) ined.)	• Brown spots with yellow halos on leaves	Knorr (1968)
44. Knobby bark (USA-California)	• Orange (*Citrus* × *aurantium* L. Var. *Sinensis* L.) • Lemon (*Citrus* × *limon* (L.) Osbeck)	• Hard, gall-like projections originating in the wood below the bark	Fawcett & Klotz (1948)
45. Lemon tree collapse (USA-California)	• Lemon (*Citrus* × *limon* (L.) Osbeck), most commonly on sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.) and grapefruit (*Citrus* × *aurantium* L. var. *racemosa* (Risso) ined.) rootstocks	• Twisted, rolled leaves • Wilting, yellowing, and defoliation of leaves • Excessive fruiting • Small and prematurely colored fruit • Lack of growth • Sloughing of bark from rootlets • Trees may die or partially recover	Calavan (1949)
46. Lime ringspot (Argentina, Venezuela)	• Limes (*Citrus* × *aurantiifolia* (Christm.) Swingle)	• Light green, translucent spots on leaves formed of narrow rings with irregular margins	Fawcett & Klotz (1948); Knorr et al. (1964)

47. Narrow leaf "Stenofillia" (Italy)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.) and sour orange (*Citrus* × *aurantium* L.)	• Leaf narrowing that can be either mild or so severe that the leaf is almost reduced to just the midrib • Leaves are lobate, asymmetrical, curled, sometimes crinkled and twisted, thick, and very leathery • Symptoms more pronounced in spring • Shorter internodes and bushier trees	Marras (1960); Marras (1972)
48. Ombrosis (Brazil)	• Citron (*Citrus medica* L.)	• Circular, dark maroon spots on leaves • Dark dots within spots	Fawcett & Klotz (1948)
49. Podagra (USA-Florida)	• Rough lemon (*Citrus* × *granulata* Raf.) rootstocks	• Rootstock enlargement • Exocortis-like bark scaling	Bitters (2021); Knorr (1957)
50. Popped bark (USA-Florida)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.)	• Corky pustules on twigs and leaves	Knorr (1968)
51. Trabut's infectious chlorosis (Algeria)	• Not specified	• Clearing of main leaf veins, followed by eventual clearing of smaller veins	Trabut (1913)
52. Variola (Brazil)	• Tangerine (*Citrus* × *aurantium* L. var. *chrysocarpa* (Hassk.) ined.) • Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.) • Lemon (*Citrus* × *limon* (L.) Osbeck) • Lime (*Citrus* × *aurantiifolia* (Christm.) Swingle) • Grapefruit (*Citrus* × *aurantium* L. var. *racemosa* (Risso) ined.)	• Small blisters on twigs caused by gum pockets in the xylem • Blister may burst to form craters • Stem pitting • Heavily affected trees may exhibit stunting, decline, and defoliation	Salibe & Moreira (1965)
53. Vasudeva's viral die-back (India)	• Sour lemon (*Citrus* × *limon* (L.) Osbeck) • Sour orange (*Citrus* × *aurantium* L.) • West Indian lime (*Citrus* × *aurantiifolia* (Christm.) Swingle) • Key lime (*Citrus* × *aurantiifolia* (Christm.) Swingle)	• Pronounced veinal chlorosis • Chlorotic spots • Twig decline	Vasudeva (1957)
54. Wart (USA-Florida, Dominican Republic)	• Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.)	• Eruptive galls on the trunk with brown cavity underneath	Knorr (1968)
55. Watson's citrus tree decline (Iraq)	• Lemon (*Citrus* × *limon* (L.) Osbeck) • Sweet orange (*Citrus* × *aurantium* L. var. *sinensis* L.) • Sour orange (*Citrus* × *aurantium* L.) • Tangerine (*Citrus* × *aurantium* L. var. *chrysocarpa* (Hassk.) ined.)	• Leaves turn pale green and then yellow • Irregular defoliation leading to complete defoliation • Eventual death of large limbs • Irregular growth in the cambium area • Lack of new growth, yet even defoliated trees could bloom • Partially defoliated trees can produce fruit	Watson & Al-Adhami (1957)

Concluding remarks

In conclusion, we recommend that the citrus disorders or pathogens referred to in the literature as citrus dieback disease (#6), leaf mottle yellows disease (#6), citrus vein-phloem degeneration (#6), citrus crinkly leaf virus (#7), mechanically transmitted citrus ringspot virus (#12), Dweet mottle virus (#14), oleocellosis-like symptoms of satsuma orange, summer orange dwarf (a.k.a., satsuma dwarf-like disease on Citrus natsudaidai in Yamaguchi prefecture) (#17), citrus mosaic (#17), natsudaidai dwarf (#17), Hassaku dwarf (#18), Bahia bark scaling (#20), and zonate chlorosis (#21) no longer be considered distinct viral diseases. Rather, each of these diseases has been attributed to other known pathogens, viruses or not.

The other phantom disorders mentioned in this work remain disorders of unknown etiology. Furthermore, while a few of them continue to be occasionally observed in the field, most of them have been absent for many decades, and all known isolates have been lost. With the growing ease of access to high-throughput sequencing technologies, it is increasingly likely that the status of any of the graft-transmissible citrus disorders of unknown etiology still present in the field or in greenhouse disease collections will be characterized in the near future. As for the rest, without any new reports or type isolates, they may remain true phantoms and never pose a threat to the citrus industry again.

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