

EPPO Datasheet: *Geosmithia morbida*

Last updated: 2020-11-10

The fungus *Geosmithia morbida* and its insect vector *Pityophthorus juglandis* are responsible for the thousand cankers disease of walnut.

IDENTITY

Preferred name: *Geosmithia morbida*

Authority: Kolarík, Freeland, Utley & Tisserat

Taxonomic position: Fungi: Ascomycota: Pezizomycotina: Sordariomycetes: Hypocreomycetidae: Hypocreales: Bionectriaceae

Common names: thousand cankers disease

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EPPO Categorization: A2 list, Alert list (formerly)

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EU Categorization: Quarantine pest ((EU) 2019/2072 Annex II B)

EPPO Code: GEOHMO



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Notes on taxonomy and nomenclature

This species was described by Kolarík, Freeland, Utley & Tisserat in 2011 (Kolarík *et al.*, 2011). The classification was modified in 2020 (EFSA, 2020), the previous classification included *Geosmithia morbida* within the *Trichocomaceae* family (Ascomycota, Eurotiomycetes; Eurotiales).

HOSTS

The EPPO pest risk analysis (EPPO, 2015) gives a comprehensive overview of the host plants. The host plants of *Geosmithia morbida* belong to the genera *Juglans* and *Pterocarya* (Juglandaceae). Although they belong to the same family, the species *Carya illinoinensis*, *C. aquatica*, and *C. ovata* were found to be immune to the fungus (Utley *et al.*, 2013). To date, *Juglans nigra* (black walnut) has been recorded as the most severely affected host plant (EFSA, 2020). In the USA, the disease has also been observed on many other walnut species, such as *J. californica* (Southern California black walnut), *J. hindsii* (Northern California black walnut), *Juglans* hybrids (e.g. *J. hindsii* × *J. regia*), and occasionally on *J. cinerea* (butter nut). On *J. major* (Arizona walnut), *G. morbida* causes small, superficial cankers but no extensive dieback. Finally, *J. regia* (English walnut) has exhibited symptoms only in rare cases. Susceptibility studies carried out in the USA have shown that all tested walnut species (*J. ailanthifolia*, *J. californica*, *J. cinerea*, *J. hindsii*, *J. major*, *J. mandshurica*, *J. microcarpa*, *J. nigra*, *J. regia*) were susceptible to the disease but at different levels. In these experiments, *J. nigra* was the most susceptible species, and results obtained for other *Juglans* spp. corroborated many of the field observations made so far in the USA. In these experiments, inoculated *J. regia* developed cankers but susceptibility varied between experiments (EPPO, 2015). In Europe, the disease was recorded in Italy only on *J. nigra* and *J. regia*, the former being more severely affected (Montecchio and Faccioli, 2014; Montecchio *et al.*, 2014).

Host list: *Juglans ailanthifolia*, *Juglans californica*, *Juglans cinerea*, *Juglans hindsii*, *Juglans major*, *Juglans mandshurica*, *Juglans microcarpa*, *Juglans mollis*, *Juglans nigra*, *Juglans regia*, *Juglans*, *Pterocarya fraxinifolia*, *Pterocarya rhoifolia*, *Pterocarya stenoptera*, *Pterocarya*

GEOGRAPHICAL DISTRIBUTION

The origin of this newly recognized pathogen is uncertain, but isolates from walnut species in the Western United States show high genetic diversity, suggesting the fungus may be native to that region. The known distribution of *Geosmithia morbida* overlaps with those of its vector, the walnut twig beetle, *Pityophthorus juglandis* (Coleoptera:

Curculionidae, Scolytinae), native to the south-western United States and Mexico and then introduced to the North-Western and Eastern USA. However, *P. juglandis* has additionally been reported from Chihuahua State, Mexico, where the fungus was not detected (Wood, 1982; Wood and Bright, 1992). In 2013 both the thousand cankers disease, the disease caused by *G. morbida*, and its vector *P. juglandis* were first recorded in the EPPO region in North-Eastern Italy, in Veneto region, Vicenza province on symptomatic black walnuts (*Juglans nigra*). In 2014 both pests were also found on English walnut (*Juglans regia*) (Montecchio and Faccoli, 2014; Montecchio *et al.*, 2014). The disease then spread within Northern Italy into Piedmont, the rest of Veneto, Lombardy, Tuscany and Emilia-Romagna regions (EPPO, 2019), and the vector *P. juglandis* was also detected in Friuli Venezia Giulia (EPPO, 2016).



EPPO Region: France (mainland), Italy (mainland)

North America: United States of America (Arizona, California, Colorado, Idaho, Indiana, Maryland, Nevada, New Mexico, North Carolina, Ohio, Oregon, Pennsylvania, Tennessee, Utah, Virginia, Washington)

BIOLOGY

Thousand cankers disease is the result of the combined activity of the fungus *G. morbida* and its insect vector *P. juglandis*. Only asexual reproduction of *G. morbida* is known and no sexual stage has been identified (EPPO, 2015). Reproduction occurs by conidia (fungal spores) produced in conidiophores (CABI, 2019). Adults of *P. juglandis* carry the pathogen conidia passively and introduce them into the host trees when the insects bore penetration holes and galleries into the bark to reproduce in the phloem. The insect can also introduce the fungus into the tree via short galleries excavated in the bark, which are then abandoned (tasting holes) (Audley *et al.*, 2017). Unlike most bark beetles, *P. juglandis* has no mycangium (specific structures to transport symbiotic fungi) however, the elytras of emerging adult beetles from infected host trees are heavily contaminated externally with spores of *G. morbida* (Newton *et al.*, 2009; Cranshaw and Tisserat, 2012). Small, round to oval, and dark-brown cankers develop around the penetration holes and the maternal galleries bored by the adults of *P. juglandis* during the bark colonisation of the host. The fungus develops within and around the insect galleries in the phloem but it does not spread systemically within the host tree (EFSA, 2020). The mating systems of *P. juglandis* are short, perpendicular to the wood fibres, 2.5–5 cm long and composed of 2-3 egg galleries (Graves *et al.*, 2009; Faccoli, 2015). The cankers are often scattered every 2–5 cm and initially limited to the phloem and the outer bark.

DETECTION AND IDENTIFICATION

Symptoms

The external symptoms of thousand cankers disease are not usually visible until the tree is significantly damaged. The period between the first bark infection and the development of visible external symptoms of the tree may be several years, depending on many factors. In the most susceptible walnut species and genotypes, tree death occurs 3–5 years after the first symptoms appear (Tisserat *et al.*, 2009). Macroscopic symptoms of thousand cankers disease are detectable at an early stage on small-diameter twigs (up to 15 mm) (Seybold *et al.*, 2012). These symptoms consist of feeding wounds or penetration and emergence holes produced by infected walnut twig beetles which release mycelial propagules, allowing fungal colonization of the neighbouring subcortical tissues. This gives rise to numerous (hence the name thousand cankers) single small subcortical dark-brown to black cankers, from a few millimetres up to 10–20 cm in size, elongated lengthwise along the stem. Initially, the cankers are only visible under the bark, but in a few weeks symptoms become visible also externally (Tisserat *et al.*, 2009). These are usually the result of many infestation points on the same branch resulting in multiple, coalescing cankers girdling the branch, and spreading downward often causing the death of the tree (Montecchio *et al.*, 2016). Non-merging cankers, equidistant from neighbouring ones and frequently exhibiting darker edges (due to incompatibility reactions between isolates, which are probably associated with the presence of a mycovirus) can be also observed (Montecchio *et al.*, 2015). As the cankers develop deeply reaching the cambium, the infected plant tissues macerate and the cankers can merge as a result of extensive bark colonization of the beetle due to multiple attacks and may finally cause dieback of the whole branches and twigs. Following merging, in the advanced stages of decline trunk cankers can exceed 2 m vertically and cover half or more of the trunk circumference (Tisserat *et al.*, 2009; EPPO, 2020).

Stressed trees show earlier external symptoms than vigorous trees. The main external symptoms of thousand cankers disease include crown dieback, flag-like wilting, subcortical cankers along trunk and branches, abnormally thin crowns, holes on the bark with emerging mycelium, and wilted individual branches with yellowing or wilted leaves that remain on the tree. Relatively large numbers of walnut twig beetles are needed to cause enough cankers to kill trees. The pathogen does not affect the plant root system, and stumps or the lowest part of trunk, which may produce many new vigorous suckers. The best season for external examination is summer, when wilting symptoms will be most noticeable.

Morphology

Morphological identification of *G. morbida* by colony characteristics and microscopy is not easy and should be performed by expert mycologists. The EFSA (2020) pest survey card dealing with *G. morbida* and its vector *P. juglandis* provides a clear description of the main protocols required for correct isolation, and morphological or molecular identification of *G. morbida*. Similar information is also reported in the thousand cankers disease guidelines from the US Department of Agriculture (Seybold *et al.*, 2019). In this respect, EFSA (2020) underlines that *G. morbida* can be confused with *Penicillium*, although colonies of *G. morbida* are lobated and various shades of yellow, whereas *Penicillium* has green to grey colonies. Moreover, *G. morbida* produces conidia linked in cylindrical to ellipsoid chains (Seybold *et al.*, 2019), distinguishable from the globose conidia of *Penicillium* (EFSA, 2020). Pictures of the colony appearance and the fungus are shown in Kola?ík *et al.* (2011). Molecular identification carried out with the *G. morbida*-specific primers GmF3 and GmR13 resulted in 80–90% or higher accuracy in detecting the fungus from infected insects (Moore *et al.*, 2019; Seybold *et al.*, 2019 in EFSA 2020). Molecular identification of *G. morbida* from colonies in culture is also very reliable, as fungal DNA is highly concentrated (EFSA, 2020). A rapid molecular method to identify *G. morbida* with the species-specific GS 004 microsatellite locus is also available (Oren *et al.* 2018; EFSA, 2020).

Detection and inspection methods

Usually, the fungus and its vector are not easily detectable by visual examination of an infected tree. The symptoms include the penetration and exit holes of the insect vector (larvae and beetles are under the bark), as well as external symptoms on the tree due to fungal proliferation (EFSA, 2020).

Early-detection of *G. morbida* may be carried out in EPPO countries by a specific and intensive survey program, which should be set up especially in those countries importing large quantities of walnut wood from the USA. In

particular, surveys could be performed at points of entry (e.g. ports) and facilities (e.g. sawmills and nurseries) receiving *Juglans* wood and plants, and in nearby areas where *Juglans* trees grow. Moreover, extensive surveys should also be conducted in the regions where the pest has been already found. The survey can be based on the following points:

Detection of infected plants. A specific survey to detect infected plants should be carried out in walnut plantations growing close to the potential points of entry. The presence of symptoms should be looked for on the walnut trees. Detection of symptoms is very difficult in the early stages of the infestation, and surveys need to be performed each year during the vegetative season, looking at the upper part of the canopy which may show the disease symptoms (Montecchio *et al.*, 2016). As visible symptoms on damaged trees may look similar to symptoms caused by other biological and abiotic stress factors (EPPO, 2015; Oren *et al.*, 2018), these symptoms are not specific to this pathogen but they are important features for a possible early-detection.

Low insect population densities at the beginning of an infestation are difficult to detect. *P. juglandis* is one of only a few species of this genus that infests hardwood in North America, and the only one in Europe (Tisserat *et al.*, 2009; EPPO, 2015; EPPO, 2020). A detection of small bark beetles on *Juglans* spp. or *Pterocarya* spp. may indicate an infestation with *P. juglandis*.

Material inspection. Careful inspection of potentially infected material and the most relevant walnut commodities (i.e. round wood, firewood, bark, plants for planting) should be carried out at points of entry into the EPPO region to prevent or reduce further introductions and pathogen dispersal. Bark branches and logs may be inspected looking for entrance holes. Penetration and exit holes created by the vector are easy to detect. The shape of the mating system (egg and larval galleries) may also help in identifying the beetle species (Faccoli, 2015). The typical oval cankers caused by *G. morbida* are located in the phloem around *P. juglandis* galleries (Tisserat *et al.*, 2009), and they are usually not visible until a thin layer of the outer bark is removed (Tisserat *et al.*, 2009; Grant *et al.*, 2011) making their detection difficult (Newton *et al.*, 2009). In this respect, debarking allows an inspector to check for the presence of phloem degradation and occurrence of cankers. Dark stains of old plant sap may be visible on the bark (Graves *et al.*, 2009).

Tree sampling. Trees showing symptoms of thousand cankers disease should be sampled to check for the presence of *G. morbida*. Heavily symptomatic branches or branches with small and round penetration holes must be debarked carefully with a knife until reaching the phloem. Only branches showing holes, insects (beetles or larvae) and/or cankers should be collected. Seybold *et al.* (2019) suggest cutting branches with a diameter of 5–10 and 15–30 cm length including both healthy and damaged tissues. Beetles or larvae extracted from the plant tissues can be stored singly in vials with 70% ethanol. Sampling tools should be sterilised with 70% ethanol before carrying out further sampling on different trees (EFSA, 2020).

PATHWAYS FOR MOVEMENT

The reasons why walnut twig beetle populations have recently increased to levels causing major walnut mortality is unknown, but is likely to be linked to its invasion of new geographic areas where new species of walnut are found, some of which are highly susceptible.

According to EFSA (2020) and to therein reported literature, the active flight capacity of the walnut twig beetle adults is limited and their natural spread capacity is rather low (EPPO, 2020), although wind may play a key role in the natural passive dispersal (Kees *et al.*, 2017). The potential spread of *P. juglandis* is expected to be correlated to the population density (EPPO, 2015). However, field observations carried out in infested areas in Italy suggest a very high dispersal capacity of the disease also at very low population density, with a very rapid spread in the whole Northern and Central Italy in a few years (Faccoli *et al.*, 2016). Concerning the natural spread of the fungus without its vector, preliminary investigations through spore trapping and laboratory trials demonstrated that *G. morbida* can produce and release conidia from walnut twig beetle exit holes, which could spread at least over short distances (Montecchio *et al.*, 2016). Air-dispersed conidia from infected walnuts to recently pruned fruit orchards growing close by cannot be excluded as a spread pathway. However, a successful colonisation of new hosts by air-dispersed conidia from infected walnuts is not expected without the vector (EPPO, 2015; EFSA, 2020).

Human-assisted movement of infested bark, wood with bark (i.e. round wood, logs, processing wood residues, firewood) and mature plants for planting of the genera *Juglans* and *Pterocarya* can easily contribute to the spread of

thousand cankers disease over long distances and across geographical barriers. This could explain the presence of a strain from the United States in the first European outbreak located in North Italy a few kilometres from a sawmill importing walnut logs from North America (Montecchio and Faccoli, 2014). For the fungus, wood chips may also be a pathway but it cannot spread or become further established in other host plants without the presence of the vector. Roots, nuts and seeds of these genera are not pathways for the pest or its vector (Newton *et al.*, 2009; EPPO, 2015; EFSA, 2020).

PEST SIGNIFICANCE

Economic impact

Since the mid-1990s the disease has been responsible for widespread mortality of walnut species in the United States (Zerillo *et al.*, 2014), and since 2013 also in Italy (Montecchio and Faccoli, 2014; Montecchio *et al.*, 2014)

Walnuts are trees of primary importance in Europe; they are naturally present in forests, cultivated for fruit or timber production, and grown as ornamental trees, and the dispersal of thousand cankers disease may have significant ecological, environmental and economic impacts (Montecchio *et al.* 2016). In this respect, the economic impacts of thousand cankers disease would occur in four main areas: 1) loss to the wood products industry as trees die, 2) loss to forest landowners with black walnut trees on their property, 3) loss to the nut industry, and 4) the loss to local communities as street trees die.

Control

No specific control methods (chemical, cultural, biological, resistant varieties) are currently available against *G. morbida* (and *P. juglandis*). A successful control procedure for thousand cankers disease has not yet been identified and the development of specific control strategies will require better understanding of the biology and ecology of *G. morbida* and its vector. Because of the extended period when adult beetles are active during spring and summer (Faccoli *et al.*, 2016), and the (usually) large size of the susceptible trees, insecticide spray applications will likely have limited effectiveness. Furthermore, colonization of the bark and cambium by the fungus may continue even if adult beetles or larvae are killed by insecticides. Moreover, bark colonization will likely limit the effectiveness of systemic insecticides and fungicides to control pest and pathogen in already infested trees.

Various chemical control methods (sprays of fungicides or applications through soil irrigation or trunk injections) have been investigated, but to date none have been reported to adequately control the pathogen (Hasey and Seybold, 2010; Tisserat and Cranshaw, 2012, Cranshaw and Tisserat, 2012). However, although no registered control protocols are available at present for preventing or treating trees infected by thousand cankers disease, preliminary tests performed - both in vitro and on naturally infected black walnut trees - with an injectable blend of pesticides killed more than 90% of the insects and the fungus for at least 12 months (Montecchio *et al.*, 2016). Heat treatment of infected logs at 48°C (measured 1 cm below the cambium) for 40 minutes was also demonstrated to be effective against *G. morbida* (Mayfield *et al.*, 2014).

Use of walnut resistant cultivars is a possibility. Differences of susceptibility to *G. morbida* infections have been observed between walnut species and between trees within the same species. Studies are in progress to evaluate whether differences occur between different populations of *J. nigra*. Surviving trees in affected areas may be particularly promising sources of genetic material to develop resistant cultivars. Identifying resistant or less susceptible cultivars would provide a very promising and sustainable control method for the long term.

Rapid detection and removal of infected trees currently remains the primary means of managing thousand cankers disease. Reducing or slowing fungus spread from infested areas relies on quarantines of wood products and public education.

Phytosanitary risk

In the EPPO region, the most widely grown *Juglans* species is *J. regia* which has long been cultivated for nut production, amenity purposes and wood production. In this respect, the EPPO Pest Risk Analysis (EPPO, 2015)

notes that *G. morbida* (and *P. juglandis*) have the potential to establish throughout the EPPO region where *Juglans* species occur. They are likely to be more damaging in the Southern and Eastern parts of the EPPO region, according to the higher number of generations of *P. juglandis* per year and where walnuts are also grown more widely. However, the susceptibility of these EPPO regions to thousand cankers disease remains to be further studied. The most susceptible species, *J. nigra*, has been introduced during the 17th Century into the EPPO region, firstly for amenity purposes and later for the production of high-quality wood. Although more data is needed on its distribution and economic importance, this insect-fungus association probably has the potential to establish and spread if no measures are taken.

The introduction of the walnut twig beetle and the associated thousand cankers disease clearly represents a threat to the cultivation of *Juglans* species in the EPPO region, and it is desirable that measures are taken to prevent or reduce any further spread.

Identification of risk factors and their relative risk estimation is essential for performing a risk-based survey. For the identification of risk areas, it is first necessary to identify the activities that could contribute to the introduction or spread of *G. morbida* and its vector *P. juglandis*. In this respect, thousand cankers disease is considered to be a serious threat to *Juglans* spp. in the whole area in which they are grown within the EPPO region, as the distribution of susceptible walnut species and hybrids frequently overlaps in a geographical continuum. thousand cankers disease can thus potentially spread in the whole EPPO regions where walnut trees occur and, due to the thermophilic character of the fungus, this disease is expected to move mainly southward from the current outbreaks.

PHYTOSANITARY MEASURES

EPPO Standard PM 8/12 on *Juglans* (EPPO, 2020) reports a detailed list of recommended phytosanitary measures and the requirements needed for the *Juglans* protection and the containment of *Juglans* pests, including thousand cankers disease and walnut twig beetle. The measures are based mainly on the movement restriction of products potentially infected and destruction of those already infected, as follows:

Movement restriction. In the USA and Italy, phytosanitary measures have been taken to protect healthy walnut trees from extensive mortality. The directly applied phytosanitary measures mainly consist of restrictions on the movement of walnut wood and plants for planting to prevent fungus spread. Since 2014 a specific thousand cankers disease survey was implemented in North-Eastern Italy to monitor the infested areas, periodically updated according to new records. In these areas specific phytosanitary measures were implemented, including the prohibition of movement outside the infected areas of plants for planting of *Juglans* and *Pterocarya* with a diameter over 10 mm and wood products of the same genera (including felling and pruning residuals and bark), except for (a) wood squared to entirely remove bark, phloem and external xylem rings, and (b) wood thermally treated to reach at least 60°C at the external xylem rings for at least 45 min (higher than in Mayfield *et al.*, 2014). The survey was carried out in walnut plantations and in nurseries producing plants for planting of *Juglans* and *Pterocarya*; nurseries occurring in the infested areas had to keep a register of plant movements (Montecchio *et al.*, 2016).

Sanitation felling. The main phytosanitary measures against thousand cankers disease and walnut twig beetle are based on the prompt cut, harvesting and destruction of infected trees aimed to reduce population density of the vector and the fungus inoculum in the environment. In particular, walnut wood may allow development of *G. morbida* and *P. juglandis* until it has thoroughly dried, and infected wood must be destroyed. To be effective this phytosanitary practice needs to be done over a wide area. Infested trees and wood must be disposed of in a way that will reduce further insect emergence and not allow dispersal of beetles, or movement of fungus infected trees. Effective sanitation felling is often complicated by the long time lag between tree infestation and symptom expression, and difficulties in detecting *G. morbida* and *P. juglandis* when their populations are low. Nevertheless, some measures may be useful as part of containment plans to dispose of infested trees and wood or to reduce populations within an infested area, such as, for example, isolation, storage, debarking, chipping and appropriate disposal of the wood by grinding or burning of the infested trees and woody material.

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Datasheet history

This datasheet was first published in 2020. It is maintained in an electronic format in the EPPO Global Database.

The sections on 'Identity', 'Hosts', and 'Geographical distribution' are automatically updated from the database. For other sections, the date of last revision is indicated on the right.



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