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SCIENTIFIC OPINION

Scientific Opinion on the pest categorisation of *Plenodomus tracheiphilus* (Petri) Gruyter, Aveskamp & Verkley [syn. *Phoma tracheiphila* (Petri) L.A. Kantschaveli & Gikashvili][1]

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ABSTRACT

The European Commission requested the EFSA Panel on Plant Health to perform a pest categorisation of *Phoma tracheiphila*, the fungal pathogen responsible for "mal secco" disease of citrus. This pathogen is listed in Annex IIAII of Directive 2000/29/EC. Recently, the pathogen has been reclassified as *Plenodomus tracheiphilus* (Petri) Gruyter, Aveskamp & Verkley, based on molecular phylogenetic analysis. *Plenodomus tracheiphilus* is a single taxonomic entity, and sensitive and specific methods are available for its differentiation from other related *Plenodomus* species. The main host is lemon (*Citrus limon* L.), but the pathogen has also been reported on other species of the genera *Citrus*, *Fortunella*, *Poncirus* and *Severinia* and on their hybrids. Host plants are widely grown in the southern EU Member States (MSs) and climatic conditions are conducive to disease development in both orchards and nurseries. The pathogen is present in part of the risk assessment area, being mainly reported on lemon grown in Italy, Greece, Cyprus and France, where it has a serious impact on the citrus industry. There are no obvious ecological/climatic factors limiting the potential establishment and spread of the pathogen in the, so far, non-infested citrus-producing EU MSs (i.e. Spain, Portugal, Malta and Croatia). Short-distance spread of the pathogen occurs via water splash and wind-driven rain, whereas movement of infected host plants for planting, particularly asymptomatic plants, is considered to be responsible for the introduction of the pathogen into new areas. Cultural practices and copper-based fungicide sprays may reduce inoculum sources and prevent new infections but they cannot eliminate the pathogen. *P. tracheiphilus* fulfils all of the pest categorisation criteria for having the potential to be a quarantine pest and a regulated non-quarantine pest, as those are defined in the International Standards for Phytosanitary Measures No 11 and 21, respectively.

KEY WORDS

biology, citrus, distribution, European Union, impacts, mal secco, regulated non-quarantine

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[1] On request from the European Commission, Question No EFSA-Q-2014-00264, adopted on 2 July 2014.

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BACKGROUND AS PROVIDED BY THE EUROPEAN COMMISSION

The current European Union plant health regime is established by Council Directive 2000/29/EC on protective measures against the introduction into the Community of organisms harmful to plants or plant products and against their spread within the Community (OJ L 169, 10.7.2000, p. 1).

The Directive lays down, amongst others, the technical phytosanitary provisions to be met by plants and plant products and the control checks to be carried out at the place of origin on plants and plant products destined for the Union or to be moved within the Union, the list of harmful organisms whose introduction into or spread within the Union is prohibited and the control measures to be carried out at the outer border of the Union on arrival of plants and plant products.

The Commission is currently carrying out a revision of the regulatory status of organisms listed in the Annexes of Directive 2000/29/EC. This revision targets mainly organisms which are already locally present in the EU territory and that in many cases are regulated in the EU since a long time. Therefore it is considered to be appropriate to evaluate whether these organisms still deserve to remain regulated under Council Directive 2000/29/EC, or whether, if appropriate, they should be regulated in the context of the marketing of plant propagation material, or be deregulated. The revision of the regulatory status of these organisms is also in line with the outcome of the recent evaluation of the EU Plant Health Regime, which called for a modernisation of the system through more focus on prevention and better risk targeting (prioritisation).

In order to carry out this evaluation, a recent pest risk analysis is needed which takes into account the latest scientific and technical knowledge on these organisms, including data on their agronomic and environmental impact, as well as their present distribution in the EU territory. In this context, EFSA has already been asked to prepare risk assessments for some organisms listed in Annex IIAII. The current request concerns 23 additional organisms listed in Annex II, Part A, Section II as well as five organisms listed in Annex I, Part A, Section I, one listed in Annex I, Part A, Section II and nine organisms listed in Annex II, Part A, Section I of Council Directive 2000/29/EC. The organisms in question are the following:

Organisms listed in Annex II, Part A, Section II:

- *Ditylenchus destructor* Thome
- *Circulifer haematoceps*
- *Circulifer tenellus*
- *Helicoverpa armigera* (Hübner)
- *Radopholus similis* (Cobb) Thome (could be addressed together with the HAI organism *Radopholus citrophilus* Huettel Dickson and Kaplan)
- *Paysandisia archon* (Burmeister)
- *Clavibacter michiganensis* spp. *insidiosus* (McCulloch) Davis *et al*.
- *Erwinia amylovora* (Burr.) Winsl. *et al*. (also listed in Annex IIB)
- *Pseudomonas syringae* pv. *persicae* (Prunier *et al*.) Young *et al*.
- *Xanthomonas campestris* pv. *phaseoli* (Smith) Dye
- *Xanthomonas campestris* pv. *pruni* (Smith) Dye
- *Xylophilus ampelinus* (Panagopoulos) Willems *et al*.
- *Ceratocystis fimbriata* f. sp. *platani* Walter (also listed in Annex IIB)
- *Cryphonectria parasitica* (Murrill) Barr (also listed in Annex IIB)
- *Phoma tracheiphila* (Petri) Kantschaveli and Gikashvili
- *Verticillium albo-atrum* Reinke and Berthold
- *Verticillium dahliae* Klebahn
- Beet leaf curl virus
- Citrus tristeza virus (European isolates) (also listed in Annex IIB)

- Grapevine flavescence dorée MLO (also listed in Annex IIB)
- Potato stolbur mycoplasma
- *Spiroplasma citri* Saglio *et al.*
- Tomato yellow leaf curl virus

Organisms listed in Annex I, Part A, Section I:

- *Rhagoletis cingulata* (Loew)
- *Rhagoletis ribicola* Doane
- Strawberry vein banding virus
- Strawberry latent C virus
- Elm phloem necrosis mycoplasm

Organisms listed in Annex I, Part A, Section II:

- *Spodoptera littoralis* (Boisd.)

Organisms listed in Annex II, Part A, Section I:

- *Aculops fuchsiae* Keifer
- *Aonidiella citrina* Coquillet
- Prunus necrotic ringspot virus
- Cherry leafroll virus
- *Radopholus citrophilus* Huettel Dickson and Kaplan (could be addressed together with IIAII organism *Radopholus similis* (Cobb) Thome)
- *Scirtothrips dorsalis* Hendel
- *Atropellis* spp.
- *Eotetranychus lewisi* McGregor
- *Diaporthe vaccinii* Shaer.

TERMS OF REFERENCE AS PROVIDED BY THE EUROPEAN COMMISSION

EFSA is requested, pursuant to Article 29(1) and Article 22(5) of Regulation (EC) No 178/2002, to provide a pest risk assessment of *Ditylenchus destructor* Thome, *Circulifer haematoceps*, *Circulifer tenellus*, *Helicoverpa armigera* (Hübner), *Radopholus similis* (Cobb) Thome, *Paysandisia archon* (Burmeister), *Clavibacter michiganensis* spp. *insidiosus* (McCulloch) Davis *et al*, *Erwinia amylovora* (Burr.) Winsl. *et al*, *Pseudomonas syringae pv. persicae* (Prunier *et al)* Young *et al*. *Xanthomonas campestris* pv. *phaseoli* (Smith) Dye, *Xanthomonas campestris* pv. *pruni* (Smith) Dye, *Xyîophilus ampelinus* (Panagopoulos) Willems *et al*, *Ceratocystis fimbriata* f. sp. *platani* Walter, *Cryphonectria parasitica* (Murrill) Barr, *Phoma tracheiphila* (Petri) Kanchaveli and Gikashvili, *Verticillium alboatrum* Reinke and Berthold, *Verticillium dahliae* Klebahn, Beet leaf curl virus, Citrus tristeza virus (European isolates), Grapevine flavescence dorée MLO, Potato stolbur mycoplasma, *Spiroplasma citri* Saglio *et al*, Tomato yellow leaf curl virus, *Rhagoletis cingulata* (Loew), *Rhagoletis ribicola* Doane, Strawberry vein banding virus, Strawberry latent C virus, Elm phloem necrosis mycoplasma, *Spodoptera littoralis* (Boisd.), *Aculops fuchsiae* Keifer, *Aonidiella citrina* Coquillet, Prunus necrotic ringspot virus, Cherry leafroll virus, *Radopholus citrophilus* Huettel Dickson and Kaplan (to address with the IIAII *Radopholus similis* (Cobb) Thome), *Scirtothrips dorsalis* Hendel, *Atropellis* spp., *Eotetranychus lewisi* McGregor *md Diaporthe vaccinii* Shaer., for the EU territory.

In line with the experience gained with the previous two batches of pest risk assessments of organisms listed in Annex II, Part A, Section II, requested to EFSA, and in order to further streamline the preparation of risk assessments for regulated pests, the work should be split in two stages, each with a specific output. EFSA is requested to prepare and deliver first a pest categorisation for each of these

38 regulated pests (step 1). Upon receipt and analysis of this output, the Commission will inform EFSA for which organisms it is necessary to complete the pest risk assessment, to identify risk reduction options and to provide an assessment of the effectiveness of current EU phytosanitary requirements (step 2). *Clavibacter michiganensis* spp. *michiganensis* (Smith) Davis *et al*. and *Xanthomonas campestris* pv. *vesicatoria* (Doidge) Dye, from the second batch of risk assessment requests for Annex IIAII organisms requested to EFSA (ARES(2012)880155), could be used as pilot cases for this approach, given that the working group for the preparation of their pest risk assessments has been constituted and it is currently dealing with the step 1 "pest categorisation". This proposed modification of previous request would allow a rapid delivery by EFSA by May 2014 of the first two outputs for step 1 "pest categorisation", that could be used as pilot case for this request and obtain a prompt feedback on its fitness for purpose from the risk manager's point of view.

As indicated in previous requests of risk assessments for regulated pests, in order to target its level of detail to the needs of the risk manager, and thereby to rationalise the resources used for their preparation and to speed up their delivery, for the preparation of the pest categorisations EFSA is requested, in order to define the potential for establishment, spread and impact in the risk assessment area, to concentrate in particular on the analysis of the present distribution of the organism in comparison with the distribution of the main hosts and on the analysis of the observed impacts of the organism in the risk assessment area.

ASSESSMENT

1. Introduction

1.1. Purpose

This document presents a pest categorisation prepared by the EFSA Scientific Panel on Plant Health (hereinafter referred to as the Panel) for the species *Plenodomus tracheiphilus* (Petri) Gruyter, Aveskamp & Verkley, syn. *Phoma tracheiphila* (Petri) Kantschaveli and Gikashvili, in response to a request from the European Commission.

1.2. Scope

This pest categorisation is for *Plenodomus tracheiphilus* (Petri) Gruyter, Aveskamp & Verkley, which was previously named *Phoma tracheiphila* (Petri) Kantschaveli and Gikashvili. The pest risk assessment (PRA) area is the territory of the European Union (hereinafter referred to as the EU) with 28 Member States (hereinafter referred to as EU MSs), restricted to the area of application of Council Directive 2000/29/EC, which excludes Ceuta and Melilla, the Canary Islands and the French overseas departments.

2. Methodology and data

2.1. Methodology

The Panel performed the pest categorisation for *Plenodomus tracheiphilus* (Petri) Gruyter, Aveskamp & Verkley, following guiding principles and steps presented in the EFSA Guidance on the harmonised framework for pest risk assessment (EFSA PLH Panel, 2010) and as defined in the International Standards for Phytosanitary Measures (ISPM) 11 (FAO, 2013) and ISPM 21 (FAO, 2004).

In accordance with the harmonised framework for pest risk assessment in the EU (EFSA PLH Panel, 2010), this work was initiated as a result of the review or revision of phytosanitary policies and priorities. As explained in the background of the European Commission request, the objective of this mandate is to provide updated scientific advice to the European risk managers to take into consideration when evaluating whether those organisms listed in the Annexes of Council Directive 2000/29/EC deserve to remain regulated under Council Directive 2000/29/EC, or whether they should be regulated in the context of the marketing of plant propagation material, or should be deregulated. Therefore, to facilitate the decision-making process, in the conclusions of the pest categorisation, the Panel addresses explicitly each criterion for a quarantine pest in accordance with ISPM 11 (FAO, 2013) but also for a regulated non-quarantine pest (RNQP) in accordance with ISPM 21 (FAO, 2004) and includes additional information required as per the specific terms of reference received by the European Commission. In addition, for each conclusion the Panel provides a short description of its associated uncertainty.

Table 1 below presents the ISPM 11 (FAO, 2013) and ISPM 21 (FAO, 2004) pest categorisation criteria on which the Panel bases its conclusions. It should be noted that the Panel's conclusions are formulated respecting its remit and particularly with regards to the principle of separation between risk assessment and risk management (EFSA founding regulation[1]); therefore, instead of determining whether the pest is likely to have an unacceptable impact, the Panel will present a summary of the observed pest impacts. Economic impacts are expressed in terms of yield and quality losses and not in monetary terms, in agreement with EFSA Guidance on the harmonised framework for pest risk assessment (EFSA PLH Panel, 2010).

Table 1: International Standards for Phytosanitary Measures (ISPM) 11 (FAO, 2013) and ISPM 21 (FAO, 2004) pest categorisation criteria under evaluation.

Pest categorisation criteria	ISPM 11 for being a potential quarantine pest	ISPM 21 for being a potential regulated non-quarantine pest
Identity of the pest	The identity of the pest should be clearly defined to ensure that the assessment is being performed on a distinct organism and that biological and other information used in the assessment is relevant to the organism in question. If this is not possible because the causal agent of particular symptoms has not yet been fully identified, then it should have been shown to produce consistent symptoms and to be transmissible.	The identity of the pest is clearly defined.
Presence or absence in the PRA area	The pest should be **absent from all or a defined part of the PRA area.**	The pest is **present** in the PRA area
Regulatory status	If the pest is present but not widely distributed in the PRA area, it should be under official control or expected to be under official control in the near future.	The pest is under official control (or being considered for official control) in the PRA area with respect to the specified plants for planting.
Potential for establishment and spread in PRA area	The PRA area should have ecological/climatic conditions including those in protected conditions suitable for the establishment and spread of the pest and, where relevant, host species (or near relatives), alternative hosts and vectors should be present in the PRA area.	-
Association of the pest with the plants for planting and the effect on their intended use	-	Plants for planting are a pathway for introduction and spread of this pest.
Potential for consequences (including environmental consequences) in the PRA area	There should be clear indications that the pest is likely to have an unacceptable economic impact (including environmental impact) in the PRA area.	-
Indication of impact(s) of the pest on the intended use of the plants for planting	-	The pest may cause unacceptable economic impact on the intended use of the plants for planting.
Conclusion	If it has been determined that the pest has the potential to be a quarantine pest, the PRA process should continue. If a pest does not fulfil all of the criteria for a quarantine pest, the PRA process for that pest may stop. In the absence of sufficient information, the uncertainties should be identified and the PRA process should continue.	If a pest does not fulfil all the criteria for a regulated non-quarantine pest, the PRA process may stop.

In addition, in order to reply to the specific questions listed in the terms of reference, three issues are specifically discussed only for pests already present in the EU: the analysis of the present EU distribution of the organism in comparison with the EU distribution of the main hosts; the analysis of the observed impact of the organism in the EU; and the pest control and cultural measures currently implemented in the EU.

The Panel will not indicate in the conclusions of the pest categorisation whether to continue the PRA process as it is clearly stated in the terms of reference that at the end of the pest categorisation the EC will indicate if further risk assessment work is required following its analysis of the Panel's scientific opinion.

2.2. Data

2.2.1. Literature search

An extensive literature search on *Phoma tracheiphila* (the pathogen has been reclassified as *Plenodomus tracheiphilus* (Petri) Gruyter, Aveskamp & Verkley) was conducted at the beginning of the mandate. Further references and information were obtained from experts and from citations within the references.

2.2.2. Data collection

To complement the information concerning the current situation of the pest provided by the literature and online databases on pest distribution, damage and management, the PLH Panel sent a short questionnaire on the current situation at country level based on the information available in the European and Mediterranean Plant Protection Organization (EPPO) Plant Quarantine Retrieval (PQR) to the National Plant Protection Organisation (NPPO) contacts of all the EU MSs. A summary of the pest status based on EPPO PQR and MSs replies is presented in Table 3.

Information on distribution of the main host plants was obtained from the EUROSTAT database.

3. Pest categorisation

3.1. Identity and biology of *Plenodomus tracheiphilus*

3.1.1. Taxonomy

Name:

Plenodomus tracheiphilus (Petri) Gruyter, Aveskamp & Verkley (de Gruyter et al., 2013)

Synonyms:

Phoma tracheiphila (Petri) L.A. Kantschaveli and Gikashvili (Kantschaveli and Gikashvili 1948);

Bakerophoma tracheiphila (Petri) Cif. (Petri, 1929);

Deuterophoma tracheiphila Petri (Petri, 1929; 1930)

Taxonomic position:

Eukaryota; Fungi; Ascomycota; Pezizomycotina; Dothideomycetes; Pleosporomycetidae; Pleosporales; Leptosphaeriaceae; *Plenodomus*; *Plenodomus tracheiphilus*

Common names:

The common names used in English-speaking countries are: mal secco, mal secco of citrus, citrus mal secco, citrus wilt, mal secco disease of citrus, wilt of citrus (EPPO PQR, 2014).

Originally, the pathogen was described as *Deuterophoma tracheiphila* by Petri (1929; 1930); later it was transferred to the genus *Phoma* (Kantschaveli and Gikashvili, 1948). It was classified in *Phoma* section *Plenodomus* owing to the formation of pycnidia with typical thick-walled, scleroplectenchymatous cells (Boerema et al., 1994). Many species classified in *Phoma* section *Plenodomus* have *Leptosphaeria* teleomorphs (Boerema et al., 2004). Molecular phylogenetic studies supported the status of *P. tracheiphila* in *Phoma* and showed a relationship to some *Leptosphaeria* species (Balmas et al., 2005). However, recently detailed molecular phylogenetic studies on the genus *Phoma* and allied genera demonstrated the polyphyly of *Phoma* (de Gruyter et al., 2009, Aveskamp et al., 2010). As a result, several species formerly classified in *Phoma* section *Plenodomus* have been redescribed in the genus *Plenodomus*, including *Phoma tracheiphila* as *Plenodomus tracheiphilus* (de Gruyter et al., 2013). *P. tracheiphylus* also produces hyphal conidia; this has been described as *Acremonium*-like (Petri, 1929), as *Cephalosporium*-to *Cadophora*-like (Goidanich and Ruggieri, 1947) and as a *Phialophora* sp. (Boerema et al., 2004).

3.1.2. Biology of *Plenodomus tracheiphilus*

Plenodomus tracheiphilus is a vascular pathogen of lemons (*Citrus limon* L.) and other species of the genera *Citrus*, *Fortunella*, *Poncirus* and *Severinia* and their hybrids. The typical symptoms consist of red discoloured strands in the xylem of stems, veinal chlorosis, wilt and shedding of leaves and ultimately dieback of twigs and branches. The disease symptoms are induced by a phytotoxin, “malseccin” (Nachmias et al., 1979).

Infection occurs by conidia that are produced in pycnidia on withered twigs or by phialoconidia produced by phialides formed on “free” hyphae grown on exposed wood surfaces (including wood debris on soil), wounded plant tissues and within the xylem elements (Migheli et al., 2009). Plant debris (twigs, branches, leaves, etc.) in the orchard can be a source of inoculum for several weeks not only for the infection of the aerial plant parts, but also for the infection of wounded roots (Traversa et al., 1992). Conidia are usually dispersed by water-splash, wind-blown rain, insects, birds, etc. to infect new host plant tissues (Perrotta and Graniti, 1988).

Conidial germination may occur at temperatures between 5 °C and 30 °C with an optimum at 25 °C (Migheli et al., 2009; Nigro et al., 2011). Warm moist conditions are conducive to infection and disease development. Conidia require 40 h of moisture at temperatures in the range of 14 °C to 28 °C to germinate (Perrotta and Graniti, 1988).

The pathogen enters susceptible host tissues (leaves, branches, twigs and roots) by both conidia and mycelium through wounds/injuries caused by cultivation practices (e.g. pruning, grafting, deep ripping, etc.), thorns, weather conditions (e.g. wind, frost, hail, etc.) or other organisms (insects, birds, etc.) (Bassi et al., 1980). Although penetration through stomata was hypothesised by Petri (1929), it has never been demonstrated under field conditions (Zucker and Catara, 1985; Perrotta and Graniti, 1988; Palm, 1996).

Once in the host the fungus reaches the lumen of the xylem and then spreads systemically, mostly upward. Conidia are also transported in the xylem sap (Liberato et al., 2011; Anonymous, 2014). As the optimum temperature for disease development is 20 °C to 25 °C, disease progress is most rapid in spring and autumn. Temperatures above 30 °C inhibit mycelial growth but do not kill the pathogen within the infected tissues (Reichert and Chorin, 1956; Perrotta and Graniti, 1988). In the citrus-producing Mediterranean areas, the infection period depends on local climatic and seasonal conditions. In Sicily, infections usually occur from September to April (Ruggieri, 1948; Goidanich, 1964; Somma and Sammarco, 1986; Somma and Scarito, 1986). In Israel, the period between mid-November and mid-April was the most conducive for infection, coinciding with the rainy period, although no correlation was found between the number of rainy days and the disease incidence (Solel, 1976). As no infection was observed after the rain ceased, Solel (1976) assumed that the rain affects inoculum dissemination rather than infection.

The length of the incubation period varies depending on the climatic conditions, the age of the host and the form of the disease. In young citrus trees, the incubation period for the “mal secco” form (see below) of the disease ranges between two and seven months (Grasso and Tirrò, 1984; Somma and Sammarco, 1986), whereas in the case of the “mal nero” form (see below), the incubation period may last for several years, because this chronic type of infection could remain confined to the heartwood over a long time (Cutuli, 1985).

Disease symptoms vary depending on whether the pathogen attacks the host via the roots or shoots. On lemon, the first symptoms of “mal secco” usually appear in spring as shoot and leaf vein chlorosis followed by a premature leaf shedding and a dieback of twigs and branches (Migheli et al., 2009). Although initially the disease affects individual twigs and branches, gradually it affects the whole tree, which eventually dies. On the symptomatic twigs, immersed, flask-shaped or globose pycnidia appear as black points within lead-grey or ash-grey areas. On cutting into the twigs or after peeling off the bark of the branches and/or the trunk of the infected trees, characteristic salmon-pink or orange-reddish discoloration of the wood can be observed. This internal symptom is associated with gum production within the xylem vessels. The growth of sprouts from the base of the affected branches and of suckers from the rootstock is a common response of the tree to the “mal secco” form of the disease.

In addition to “mal secco”, which is the most common form of the disease, chronic infections of mature trees, most likely attributed to the entry of the pathogen via the roots, may cause a brown discoloration of the heartwood without any external symptoms at first. However, when the pathogen invades the outer functional xylem, infected trees collapse suddenly. This form of the disease is known as “mal nero” (Perrotta and Graniti, 1988; Solel and Salerno, 2000; Hajlaoui et al., 2008). Finally, a third form of the disease called “mal fulminante” causes a rapid wilting of the branches or of the whole tree as a result of the systemic invasion of the functional xylem by the pathogen.

The optimum temperature for symptom expression and xylem colonisation ranges between 20 °C and 25 °C (Perrotta and Graniti, 1988). At temperatures above 30 °C, fungal growth ceases and symptoms are not expressed (Perrotta and Graniti, 1988). Disease progress is temporarily inhibited by hot and cold temperature extremes. At temperatures ranging from 10 °C to 25 °C, the pathogen is able to sporulate on infected symptomatic leaves that fall on the orchard floor during autumn and spring (Anonymous, 2013). A relative humidity near saturation and temperatures between 20 °C and 25 °C are favourable for the production of phialides on hyphae present in wounds and leaf scars (De Cicco et al., 1986).

P. tracheiphilus can survive as mycelium and conidia within pycnidia on infected leaves, twigs and branches of susceptible citrus hosts (Nigro et al., 2011). The pathogen has also been demonstrated to survive as mycelium in lemon seeds, but developing seedlings do not become infected (Ippolito et al., 1987). The survival potential of the pathogen in infected host plant debris lying on the orchard floor varies between 30 days and one year depending on the soil type (De Cicco et al., 1987).

No teleomorph of *P. tracheiphilus* is known, and the absence of genetic variability observed among isolates of the pathogen in Italy (Balmas et al., 2005), Israel (Ezra et al., 2007) and Tunisia (Kalai et al., 2010) suggests that *P. tracheiphilus* has a low rate of mutation and reproduces clonally under Mediterranean conditions (Migheli et al., 2009). The pathogen is considered to have been introduced into the Mediterranean basin from other regions of the world (Migheli et al., 2009).

3.1.3. Detection and identification of *Plenodomus tracheiphilus*

Citrus leaves should be inspected for the presence of vein chlorosis, which is an early symptom of the disease (Perrotta and Graniti, 1988). Twigs of affected trees become chlorotic with a salmon-pink to orange-red discoloration of the xylem (Migheli et al., 2009). Dead twigs are often ash coloured owing to the lifting of the epidermis by the pycnidia produced underneath (Perrotta and Graniti, 1988). However, diagnosis of the disease based only on the above-mentioned symptoms (and/or signs) is not reliable, as (i) symptoms may vary depending on the form of the disease (see section 3.1.2), (ii) similar symptoms are caused by other citrus diseases and disorders (see section 3.1.3.1), and (iii) symptomless (latent) infections may occur owing to the rapid basipetal translocation of the pathogen prior to symptom development (Kalai et al., 2010).

Therefore, for a reliable detection and identification of the pathogen, laboratory testing of the affected plant tissues should be performed.

The pathogen can be isolated on various culture media (potato dextrose agar, carrot agar, malt extract agar + 1 µg/mL chloramphenicol, etc.) and can be identified on the basis of its cultural and morphological characters (EPPO/OEPP, 2007). In the absence of pycnidia, detection and identification of the pathogen can be made by using molecular methods. Conventional polymerase chain reaction (PCR) assays have been developed for the detection and identification of the pathogen on pure cultures and infected plant tissues (leaves, twigs, fruit) and for its differentiation from other *Phoma* species or other citrus pathogens (Balmas et al., 2005; Ezra et al., 2007). More recently, fast and sensitive real-time PCR protocols were developed for the quantification of the pathogen in the host as well as for its detection in latently infected (asymptomatic) plant tissues (Licciardella et al., 2006; Demontis et al., 2008). It should be noted that, although the enzyme-linked immunosorbent assay developed by Nachmias et al. (1979) can detect *P. tracheiphilus* antigens in crude plant extracts, it has not been widely used because of high levels of non-specific reactions.

A detailed description of the cultural and morphological characteristics of *P. tracheiphilus*, as well as of the methods recommended for its detection and identification, is included in EPPO Standard PM 7/48 (EPPO/OEPP, 2007).

3.1.3.1. Similarities to other citrus diseases and disorders

Other citrus diseases with symptoms similar to those caused by *P. tracheiphilus* are (a) citrus tristeza virus (CTV), especially in cases in which the rootstock is sour orange (*Citrus* × *aurantium*), (b) *Phytophthora* crown rot, if the rootstock is tristeza tolerant, (c) anthracnose (*Colletotrichum gloeosporioides*), (d) citrus blast (*Pseudomonas syringae* pv. *syringae*), (e) Asiatic citrus canker (*Xanthomonas axonopodis* pv. *citri*), (f) citrus variegated chlorosis (*Xylella fastidiosa*) (Timmer et al., 2000), and (g) Huanglongbing (*Candidatus* Liberibacter spp.), a systemic disease causing slow decline of citrus trees, with yellowing of leaves and dieback of branches (Garnier and Bove, 2000). Moreover, drought, mechanical wounding, frost injury and other stresses can weaken trees and allow infections by weakly pathogenic fungi that result in branch cankers and dieback (Graham and Menge, 2000).

3.2. Current distribution of *Plenodomus tracheiphilus*

3.2.1. Global distribution of *Plenodomus tracheiphilus*

According to the EPPO PQR (2014), *P. tracheiphilus* occurs in most citrus-growing countries of the Mediterranean and Black Sea basins, but it has not been reported from Spain, Portugal, Malta and Morocco (Duran-Vila and Moreno, 2000; Licciardello et al., 2006; Migheli et al., 2009). In Turkey, the disease was restricted to a certain area when discovered in 1933, but later it spread with the expansion of citrus plantations (Tuzcu et al., 1989). The presence of *P. tracheiphilus* in Colombia, Uganda and Queensland (Australia) has been reported, but these records were considered doubtful and, thus, they have been excluded from the distribution map of the pathogen published by EPPO PQR (2014). The global distribution of *P. tracheiphilus* is shown in Figure 1. Details of the pest status in non-EU countries are provided in Table 2.

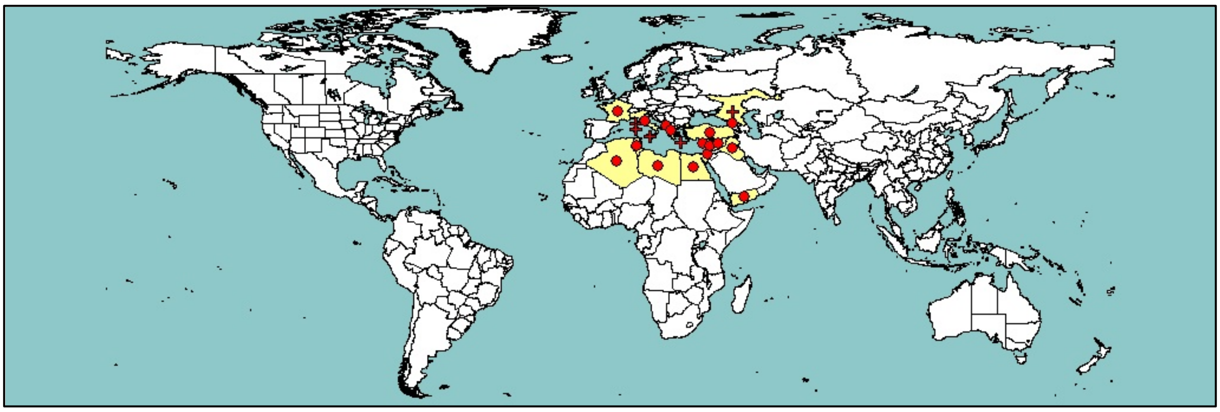

Figure 1: Global distribution map of *Plenodomus tracheiphilus* (syn. *Phoma tracheiphila*), as extracted from EPPO PQR (2014), version 5.3.1, accessed on 17 June 2014. Red circles and crosses represent national and sub-national pest records, respectively.

Table 2: The status of *Plenodomus tracheiphilus* (syn. *Phoma tracheiphila*) in non-EU countries (CABI, 2010; EPPO PQR, 2014).

Country	Pest status	References
Europe		
Albania	Present	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Russian Federation	Restricted distribution (Caucasus region)	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Asia		
Armenia	Present	Duran-Vila and Moreno, 2000
Georgia (Republic of)	Present	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Iraq	Present	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Israel	Restricted distribution	CABI/EPPO, 2004; CMI, 1989; Ezra et al., 2007; EPPO PQR, 2014
Lebanon	Present	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Syria	Present	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Turkey	Restricted distribution	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Yemen	Restricted distribution	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Africa		
Algeria	Restricted distribution	CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Egypt	Present	Punithalingam and Holliday, 1973; CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Libya	Present	Punithalingam and Holliday, 1973; CMI, 1989; CABI/EPPO, 2004; EPPO PQR, 2014
Tunisia	Present	CMI, 1989; CABI/EPPO, 2004; Hajlaoui et al., 2008; EPPO PQR, 2014
South America		
Colombia	Absent, unreliable record	EPPO PQR, 2014

3.2.2. Distribution in the EU of *Plenodomus tracheiphilus*

As indicated by EPPO PQR (2014) and the answers to the EFSA questionnaire received from the EU MSs, Iceland and Norway, the presence of *P. tracheiphilus* is reported in Italy (islands of Sicily and

Sardinia included), Greece (island of Crete included), Cyprus and France (island of Corsica included) (Table 3). There is only one interception record of *P. tracheipilus* (as *Deuterophoma tracheiphila*) on lemon (*C. limon*) plants for planting in the Europhyt database.

Table 3: Current distribution of *Plenodomus tracheiphilus* (syn. *Phoma tracheiphila*) in the risk assessment area, based on the EPPO PQR database (2014) (version 5.3.1, accessed on 17 June 2014) and the answers received from the NPPOs(a) of the EU Member States, Iceland and Norway up to 26 June 2014.

Member State	Pest status according to EPPO PQR (2014)	Pest status according to the responses to the EFSA questionnaire received from the NPPOs of the EU Member States
Austria		**Absent**, no pest records
Belgium	**Absent**, confirmed by surveys	**Absent**, no pest records
Bulgaria	**Absent**	Confirmed
Croatia		**Absent**
Cyprus	**Present**, widespread	–(b)
Czech Republic		**Absent**, no records
Denmark		–
Estonia		**Absent**, no pest records
Finland		**Absent**, no pest records
France Corsica	**Present**, restricted distribution **Present**, few occurrences	–
Germany		**Absent**, no pest records
Greece	**Present**, widespread	–
Hungary		**Absent**, no pest records
Ireland		**Absent**, no pest record
Italy Sicily & Sardinia	**Present**, no details **Present**	**Present**, restricted distribution
Latvia		–
Lithuania		–
Luxembourg		–
Malta		**Absent**, no pest records
Poland		**Absent**, no pest records
Portugal		No records
Romania		–
Slovak Republic		**Absent**, no pest records
Slovenia		**Absent**, no pest records
Spain		**Absent**
Sweden		**Absent**
The Netherlands	**Absent**, confirmed by surveys	Confirmed
United Kingdom		**Absent**
Iceland		–
Norway		–

(a) National Plant Protection Organisations
(b) No information is available

3.3. Regulatory status

3.3.1. Legislation addressing *Plenodomus tracheiphilus* (Directive 2000/29/EC)

P. tracheiphilus is regulated as a harmful organism in the EU and is listed as *P. tracheiphila* in Council Directive 2000/29/EC (Table 4) in the following sections:

Table 4: *Plenodomus tracheiphilus* in Council Directive 2000/29/EC

Annex II, Part A	Harmful organisms whose introduction into, and spread within, all Member States shall be banned if they are present on certain plants or plant products
Section II	Harmful organisms known to occur in the Community and relevant for the entire Community
(c)	Fungi
Subject of contamination	Plants of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf., and their hybrids, other than seeds

3.3.2. Legislation addressing hosts of *Plenodomus tracheiphilus*

Host plants of *P. tracheiphilus* are mentioned in the Council Directive 2000/29/EC showed in the Table 5.

Table 5: Host plants of *Plenodomus tracheiphilus* in Council Directive 2000/29/EC

Annex III Point 16	A general prohibition of the introduction in all MSs of plants of *Citrus*, *Fortunella*, *Poncirus* and their hybrids, other than fruit and seeds, from non-EU countries are mentioned in this part.
Annex IV **Part A** **Section I** **16.1**	There are no special requirements with respect to *P. tracheiphila* in Council Directive 2000/29/EC for import into the EU of fruit of *Citrus*, *Fortunella*, *Poncirus* and their hybrids. However, according to this part of the Directive, such fruit, originating in non-EU countries, shall be free from peduncles and leaves and the packaging shall bear an appropriate origin mark. Other special requirements for import into the EU of fruit of *Citrus*, *Fortunella*, *Poncirus* and their hybrids inthis part of the Directive, are targeted at harmful organisms other than *P. tracheiphila*.
Section II **(10)**	Concerning movement within the EU of plants of *Citrus*, *Fortunella*, *Poncirus* and their hybrids, other than fruit and seeds, an official statement is required to the effect that: (a) the plants originate in areas known to be free from *Spiroplasma citri* Saglio et al. (1973), *Phoma tracheiphila* (Petri), Kantschaveli and Gikashvili, *Citrus* vein enation woody gall and *Citrus* tristeza virus (European strains); or (b) the plants derive from a certification scheme requiring them to be derived in direct line from material that has been maintained under appropriate conditions and has been subjected to official individual testing for, at least, *Citrus tristeza* virus (European strains) and *Citrus* vein enation woody gall, using appropriate indicators or equivalent methods, in accordance with the procedure referred to in Article 18(2), and have been growing permanently in an insect-proof glasshouse or in an isolated cage on which no symptoms of *Spiroplasma citri* Saglio et al., *Phoma tracheiphila* (Pandri) Kantschaveli and Gikashvili, *Citrus tristeza* virus (European strains) and *Citrus* vein enation woody gall have been observed; or

	(c) the plants: • have been derived from a certification scheme requiring them to be derived in direct line from material that has been maintained under appropriate conditions and has been subjected to official individual testing for, at least, *Citrus* vein enation woody gall and *Citrus tristeza* virus (European strains), using appropriate indicators or equivalent methods, approved in accordance with the procedure referred to in Article 18(2), and has been found in these tests to be free from *Citrus tristeza* virus (European strains) and certified free from, at least, *Citrus tristeza* virus (European strains) in official individuals tests carried out according to the methods mentioned in this indent; and • have been inspected and no symptoms of *Spiroplasma citri* Saglio et al. (1973), *Phoma tracheiphila* (Pandri) Kantschaveli and Gikashvili and *Citrus* vein enation woody gall and *Citrus tristeza* virus have been observed since the beginning of the last complete cycle of vegetation.
Part B **Art. 31**	Special requirements are requested for fruit of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf. and their hybrids originating in Spain, France (except Corsica) Cyprus and Italy during transport through Greece, Corsica (France), Malta and Portugal (excepting Madeira): • the fruit shall be free from leaves and peduncles; or • in the case of fruit with leaves or peduncles, an official statement that the fruits are packed in closed containers that have been officially sealed and shall remain sealed during their transport through a protected zone, recognised for these fruit, and shall bear a distinguishing mark to be reported on the passport.
Annex V **Part A** **Section I**	This Annex specifies plants, plant products and other objects, originating in the EU, which must be subject to a plant health inspection at the place of production before being moved within the EU and which must be accompanied by a plant passport. This section includes (point 1.4) plants of *Fortunella* and *Poncirus* and their hybrids, other than fruit and seeds, (point 1.5) plants of *Citrus* and their hybrids other than fruit and seeds and (point 1.6) fruits of *Citrus*, *Fortunella*, *Poncirus* and their hybrids with leaves and peduncles.
Part B **Section I**	This part of the Annex specifies plants, plant products and other objects, originating outside the EU which must be subject to a plant health inspection in the country of origin or the consignor country before being permitted to enter the EU. This section includes (point 3) fruits of *Citrus*, *Fortunella*, *Poncirus* and their hybrids. To summarise, the pathway "plants for planting" is regulated by prohibition of import, and the pathway "fruit" is regulated by the requirement of a general plant health inspection, but not by special requirements with respect to *P. tracheiphila*.

3.3.3. Marketing Directives

Some of the host plants of *P. tracheiphilus* are also regulated under Marketing Directives of the EU (Table 6).

Table 6: *Plenodomus tracheiphilus* host plants in EU Marketing Directives

Plant propagation material	Marketing Directive	Comment
Citrus L. *Fortunella* Swingle *Poncirus* Raf.	Council Directive 2008/90/EC of 29 September 2008 on the marketing of fruit plant propagating material and fruit plants intended for fruit production	Official inspections check if the material meets criteria for: Identity; Quality; Plant health; The rules also cover batch separation and marking, identification of varieties and labelling.

3.4. Elements to assess the potential for establishment and spread in the EU

3.4.1. Host range

The main host of *P. tracheiphilus* is lemon (*C. limon*), but the pathogen has also been reported on many other citrus species belonging to the genera *Citrus*, *Fortunella*, *Poncirus* and *Severinia*, as well as on their interspecific and intergeneric hybrids (EPPO/CABI, 1997; Migheli et al., 2009). The following plant species are also hosts of *P. tracheiphilus*: *C. madurensis* (syn. *Citrofortunella microcarpa;* calamondin), *C. aurantiifolia* (lime), *C. aurantium* (sour orange), *C. bergamia* (bergamot), *C. jambhiri* (rough lemon), *C. latifolia* (Tahiti lime), *C. limonia* (mandarin lime), *C. macrophylla* (alemow), *C. medica* (citron), *C. reticulate* (common mandarin), *C. sinensis* (sweet orange), *C. unshiu* (satsuma mandarin), *Citrus* × *paradisi* (grapefruit), *Fortunella* spp. (kumquat), and *Poncirus trifoliata* (trifoliate orange) (CABI, 2010).

Citrus species and cultivars differ in their susceptibility to infection by *P. tracheiphilus* (Table 7). Most of the available information on the relative susceptibility of citrus selections to infection by the pathogen is based on field observations and it has not always been evaluated in comparative pathogenicity tests (Migheli et al., 2009; Nigro et al., 2011). Perrotta and Graniti (1988), Tuzcu et al. (1989) and Migheli et al. (2009) reported that most of the citrus species are susceptible to *P. tracheiphilus* when artificially inoculated by wounding.

Citrus rootstocks also show different reactions to infection by the pathogen. The most widespread lemon rootstocks in Italy, Greece and Turkey, sour orange (*C. aurantium*), rough lemon (*C. jambiri*), volkamer lemon (*C. volkameriana*) and alemow (*C. macrophilla*), are very susceptible to the disease (Table 7) (Solel and Oren, 1975; De Cicco and Ippolito, 1987; Migheli et al., 2009; Nigro et al., 2011). Serious infection by the pathogen has been observed in Italian orchards of sweet orange “Tarocco nucellar line” and clementine mandarin grafted onto sour orange rootstock, as well as on “Fortune” mandarin and “Tacle” hybrid (*C. sinensis* “Tarocco” × clementine) grafted onto alemow rootstock (Balmas et al., 2005; Magnano di San Lio et al., 2005). On the other hand, some of the rootstocks are considered to be tolerant, such as Cleopatra mandarin (*C. reshni* Hort. ex Tanaka), trifoliate orange (*Poncirus trifoliata*), and, to a lesser extent, Troyer citrange (*C. sinensis* × *P. trifoliata*) (Table 7) (EPPO/CABI, 1997; Nigro et al., 2011). There is evidence indicating that the rootstock influences the susceptibility of the scion to *P. tracheiplila* (Solel and Spiegel-Roy, 1978; Nigro et al., 1996).

Evaluations of the susceptibility level of citrus to infection by the pathogen are sometimes contradictory, especially for rootstocks. For instance, in Tunisia, the citrus industry is based on the use of the lemon cultivars Eureka and Lunari grafted onto sour orange (*C. aurantium*), which is the oldest rootstock used in citriculture for its resistance to several diseases (Ziadi et al., 2014). On the other hand, sour orange is considered to be highly susceptible to the infection by *P. tracheiphilus* in Italy, while it is moderately susceptible in Israel (Palm, 1996). In pathogenicity tests, the susceptibility of sour orange was higher in young plants than in older ones (Demontis et al., 2008). Owing to the above contradictions, Khanchouch and Hajlaoui (2012) proposed a mathematical model to conduct a rapid and efficient resistance screening test.

Table 7: Susceptibility to *Plenodomus tracheiphilus* (syn. *Phoma tracheiphila*) infection of some citrus species, allied genera and hybrids, other than lemon [adapted and updated by Nigro et al. (2011) from Cutuli et al. (1984)]

Latin binomial name	Common name	Susceptibility [(a)]
Citrus jambhiri	Rough lemon	+++
C. volkameriana	Volkamer lemon	++
C. meyeri	Meyer lemon	+
C. webberii	Kalpi papeda	+++
C. karna	Karna lemon	+++
C. medica	Citron	+++
C. limettioides	Sweet lime	+
C. limonia	Rangpur lime	+++
C. aurantifolia	Key lime	+++
C. bergamia	Bergamot	+++
C. deliciosa	Mediterranean mandarin	+
C. reticulata	Common mandarin	++
C. reshni	Cleopatra mandarin	+
C. clementina	Common clementine	++
C. macrophylla	Alemow	+++
C. junos	Yuzu orange	+++
C. sinensis	Sweet orange	+/++ [(b)]
C. aurantium	Sour orange	+++
C. paradisi	Grapefruit	+
C. myrtifolia	Chinotto	+++
C. madurensis	Calamondin	++
C. taiwanica	Taiwan orange	+++
C. tangelo	Orlando tangelo	+
C. ichangensis × *C. grandis*	Ichang lemon	+
Poncirus trifoliata	Trifoliate orange	+
Fortunella spp.	Kumquat	+
Severinia buxifolia	Box orange	++
Poncirus trifoliata × *C. sinensis*	“Washinghton Navel” citrange carrizo	++
C. sinensis “Washinghton Navel” × *C. trifoliata*	Troyer citrange	++

(a): Susceptibility: + low; ++ medium; +++ high.
(b): (+) according to Ruggieri (1948); (++) according to Crescimanno et al. (1973)

3.4.2. EU distribution of main host plants

Citrus are widely available as commercial crops in southern EU MSs. The area cultivated with sweet orange, lemon and small fruited citrus varieties in the EU by MS and NUTS2 (Nomenclature of Territorial Units for Statistics) region is given in Table 8.

The citrus production area in the EU-28, estimated at 494 913 ha in 2007, is located in the following eight MSs: Spain (314 908 ha), Italy (112 417 ha), Greece (44 252 ha), Portugal (16 145 ha), Cyprus (3 985 ha), France (1 705 ha), Croatia (1 500 ha), and Malta (193 ha) (Table 8). Approximately 13 % of the total area grown with citrus in the southern EU MSs is cultivated with lemon, which is considered to be the citrus species most severely affected by *P. tracheiphilus*.

Table 8: The citrus production area (in hectares) in the EU in 2007. Data extracted from Eurostat (online) on 21 February 2013 (EFSA PLH Panel, 2014a).

Member State/region	Orange varieties	Lemon varieties	Small-fruited citrus varieties	All citrus varieties [(a)]
European Union (27 Member States)	**279 048**	**62 854**	**151 510**	**493 413**
Croatia	**200**	**100**	**1 200**	**1 500**
Cyprus	**1 554**	**665**	**1 766**	**3 985**
France	**28**	**22**	**1 654**	**1 705**
Provence-Alpes-Côte d'Azur	1	5	1	8
Corsica	27	17	1 648	1 692
France, not allocated	0	0	3	4
Greece	**32 439**	**5 180**	**6 631**	**44 252**
Kentriki Ellada, Evoia	6 531	1 969	0	8 500
Ipeiros	3 993	0	0	3 993
Peloponnisos	17 347	1 730	3 379	22 458
Nisia Aigaiou	883	308	213	1 405
Crete	3 410	277	356	4 044
Other Greek regions	266	885	2 598	3 750
Malta [(b)]				**193**
Italy	**73 785**	**16 633**	**21 997**	**112 417**
Piemonte	0	0	0	0
Liguria	7	17	3	28
Tuscany (NUTS 2006)	6	0	0	6
Lazio (NUTS 2006)	399	82	178	660
Abruzzo	178	0	0	178
Molise	9	0	9	18
Campania	689	954	634	2 278
Puglia	3 462	146	4 059	7 668
Basilicata	4 640	39	2 093	6 774
Calabria	17 273	967	10 774	29 015
Sicily	43 731	14 338	3 106	61 176
Sardinia	3 387	86	1 138	4 612
Portugal	**12 416**	**494**	**3 235**	**16 145**
Norte	734	52	133	920
Centro (PT) (NUTS 95)	401	27	54	482
Lisboa e Vale do Tejo (NUTS 95)	256	196	37	490
Alentejo (NUTS 95)	1 585	11	247	1 844
Algarve	9 437	206	2 763	12 407
Spain	**158 824**	**39 859**	**116 225**	**314 908**
Principado de Asturias	0		0	1.00
Extremadura	278	0	38	317
Cataluña	2 080	20	10 777	12 877
Comunidad Valenciana	76 593	9 127	90 878	176 599
Îles Balears	660	397	98	1 156
Andalucía	64 158	5 646	9 999	79 804
Región de Murcia	14 514	24	4.433	43 509
Canary Islands (ES)	538	104	0	643

(a): Calculated by summing the area grown with orange, lemon and small-fruited citrus varieties.

(b): Data for the citrus production area in Malta are provided according to FAOSTAT (online) for the year 2011. The detailed production figures are as follows: tangerines, mandarins, clementines (6 ha); grapefruit including pomelo (1 ha); lemons and limes (38 ha); oranges (95 ha); other citrus fruits (53 ha).

A map showing the EU NUTS3 citrus-growing regions is shown in Figure 2, based on the total area cultivated with citrus species in the EU NUTS3 regions, as extracted from the national statistical databases of the EU citrus-growing MSs (Spain, Italy, Greece, Portugal, Cyprus, France, Croatia and Malta).

Figures on citrus propagating material for fruit production grown in EU nurseries are not easily available. Therefore, they were mostly estimated as number of plants based on a 7.5 % rate of tree renewal (Aubert and Vullin, 1997):

- Greece: 825 813 plants in 2006 and 542 300 in 2007 (EFSA PLH Panel, 2014b)
- France: 818 568 plants in 2005 (EFSA PLH Panel, 2014b)
- Portugal: 844 000 plants (Aubert and Vullin, 1997)
- Spain: 10 665 000 plants (Aubert and Vullin, 1997)
- Italy: 5 771 000 plants (Aubert and Vullin, 1997).

In addition to commercial orchards and nurseries, citrus species susceptible to *P. tracheiphilus* are commonly grown in city streets and public/private gardens, in both rural and urban regions of the southern EU MSs.

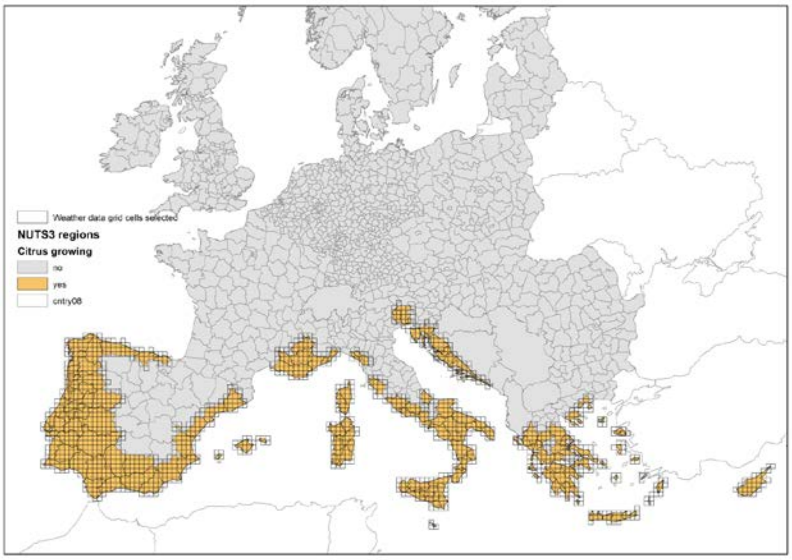

Figure 2: Map of EU NUTS3 citrus-growing regions based on citrus production data extracted from national statistical databases of Spain, Italy, Greece, Portugal, Cyprus, France, Croatia and Malta (EFSA PLH Panel, 2014a).

3.4.3. Analysis of the potential distribution of *Plenodomus tracheiphilus*

P. tracheiphilus is not known to be present in some citrus-growing areas of the risk assessment area, particularly those in the Iberian Peninsula (i.e. Spain and Portugal), Croatia and Malta (Table 3).

On the contrary, the pest is present in other Mediterranean citrus-growing areas of the EU, such as Italy, France, Greece and Cyprus (Table 3).

Lemon, which is the citrus species most severely affected by the pathogen, is widely grown in all citrus-producing EU MSs (Table 8). The total EU area grown with lemon is 62 854 ha, covering approximately 13 % of the total area cultivated with citrus in the EU. Currently the pathogen is reported to be present in four MSs, namely Italy, Greece, Cyprus and France, where it is mainly restricted to lemon. The area cultivated with lemon in those four infested EU MSs (i.e. 22 500 ha) accounts for approximately one-third of the total area grown with lemon in the EU (Table 8).

The climate types of the infested by the pathogen citrus-growing EU MSs (Italy, Greece, Cyprus and France) and non-EU countries (Tunisia, Algeria, Egypt, Libya, Israel, Lebanon, Syria and Turkey) are similar to those present in the citrus-producing areas of Spain and Portugal, with a prevalence of the Csa climate (C = temperate, s = dry summer, a = hot summer) (Figure 3).

Based on the above and the biology of *P. tracheiphilus* (see section 3.1.2), the Panel concludes that there are no obvious ecological or climatic factors limiting the potential establishment of the pathogen in the so far non-infested citrus-producing EU MSs.

3.4.4. Spread capacity

3.4.4.1. Spread by natural means

Conidia of *P. tracheiphilus*, produced within pycnidia on infected plant tissues (twigs, branches, peduncles, leaves, etc.) still attached to the tree or lying on the orchard floor, are usually dispersed over relatively short distances by rain-splash, overhead irrigation, surface flow of water or wind-blown rain (Solel and Salerno, 2000; Migheli et al., 2009; Nigro et al., 2011). According to Balmas et al. (2005), some conidia may also become airborne. Laviola and Scarito's (1989) field studies showed that the pathogen spread up to 16 m from the inoculum source, which was the maximum distance tested. Although it has not been demonstrated so far, birds and insects may act as carriers of the pathogen between trees (Perrotta and Graniti, 1986).

3.4.4.2. Spread by human assistance

The pathogen can spread over long distances via the movement of infected host plants for planting (rootstocks, grafted plants, scions, budwood, etc.), fruit peduncles and leaves, particularly latently infected (asymptomatic). It was assumed that the pathogen was introduced into Sicily accidentally with lemon plants imported from Greece (Ruggieri and Goidanich, 1953). Based on the analysis of Tunisian and Italian isolates of the pathogen, Kalai et al. (2010) assumed that the pathogen was most likely introduced into Tunisia in 1960 with infected citrus propagating material imported from southern Italy. Soil containing infected plant debris, particularly twigs, may also be a potential pathway for the introduction of the disease into new areas, as the pathogen can survive on those plant parts for up to one year, depending on the soil type (De Cicco et al., 1987).

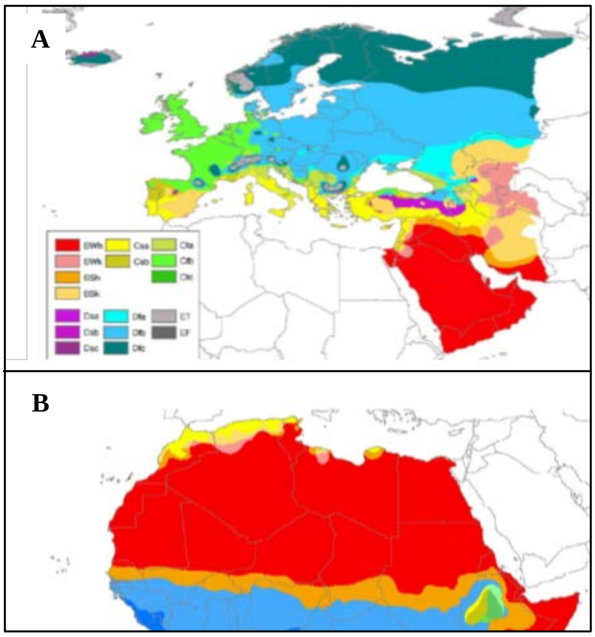

Figure 3: Köppen–Geiger climate maps of Europe and eastern Asia (A) and northern Africa (B) (from Peel et al., 2007).

P. tracheiphilus has also been detected in lemon fruit and seeds (Stepanov and Shaluishkina, 1952). The role of lemon seeds in the spread of the pathogen has been investigated by Ippolito et al. (1987, 1992), who concluded that this dissemination mechanism is unlikely because, although the fungus survives as mycelium on the seed coat, emerging seedlings do not become infected. According to Ippolito et al. (1987, 1992), fruit and seeds of susceptible citrus species, other than lemon, may also become infected by the pathogen. However, there is no evidence that the pathogen can spread via infected citrus fruit or seeds (CABI, 2010).

Although it has not been demonstrated, *P. tracheiphilus* can spread on contaminated pruning tools and agricultural machinery (CABI, 2010).

3.5. Elements to assess the potential for consequences in the EU

3.5.1. Potential effects of *Plenodomus tracheiphilus*

P. tracheiphilus is a seriously destructive pathogen, with a highly significant impact on the citrus industry in areas where the host plants, particularly lemon, are widely grown and the pathogen is present (Migheli et al., 2009; Nigro et al., 2011). Reports on pest effects from non-EU infested areas are available from Turkey and Tunisia. A long-term negative effect of *P. tracheiphilus* was reported from Turkey (in the district of Mersin) in 1956, where approximately 20 000 lemon trees were killed by the disease within 15 years (Cutuli et al., 1984). In a recent study from Turkey, it was reported that effective control of the disease would double production in lemon-producing countries (Gulsen et al., 2007). In addition, Ziadi et al. (2012, 2014) recently reported that the disease causes serious damage in Tunisia having affected up to 100 % of trees of susceptible lemon cultivars.

3.5.2. Observed impacts of *Plenodomus tracheiphilus*

3.5.2.1. Direct pest effects

There are several reports from Italy and Greece on the effects of the pest on host plants. The disease can cause a great yield loss and can also lead to twig death, thus seriously reducing the volume of the citrus tree canopy. In a final stage the disease can lead to the death of the whole tree (Migheli et al., 2009, Nigro et al., 2011). Destructive outbreaks of the disease have occurred after frost spells and hail storms in spring (Perrotta and Graniti, 1988).

In early studies it was reported that approximately 3 000 ha of lemon orchards were destroyed by "mal secco" in Sicily within 15 years of first being noticed (Savastano, 1923; Casella, 1935). Later in the 1980s it was reported that the mean fruit yield of Sicilian lemon orchards (the region consists of more than 90 % of Italian lemon production) was approximately 20 t/ha, whereas in disease-free lemon orchards yield reached 60 to 80 t/ha (Cutuli, 1985). In the same study it was estimated that, each year, 5 % of dead lemon trees and 50 % of diseased trees were attributable to infection with *P. tracheiphilus* (Cutuli, 1985). The estimated yield losses due to *P. tracheiphilus* were up to 50 % in years with highly conducive weather conditions (Cutuli, 1982). Negative effects of the disease have been also reported from Greece, where 60 to 100 % of lemon trees were affected by the disease within 20 to 25 years of planting (Thanassoulopoulos and Manos, 1992). Disease incidence was about 30 to 40 %, causing a yield loss between 20 and 30 %, but in more conducive conditions yield losses were more than 60 % in some lemon and citrus orchards (Thanassoulopoulos and Manos, 1992). Magnano di San Lio (1992) showed that in Syracuse province (Sicily) the level of the disease differed between young and older trees. The author showed that the incidence of "mal secco" was 36 to 38 % in trees under 20 years old and up to 49 % in older trees. The mean reduction in tree volume was 34 % in the older trees and varied from 24 to 29 % in younger trees (Magnano di San Lio, 1992). Magnano di San Lio (1992) also showed that the lethality index was higher in trees under eight years old.

3.5.2.2. Indirect effects of *Plenodomus tracheiphilus*

Damage caused by *P. tracheilphila* includes not only direct but also indirect negative effects, such as the additional cost of pruning dead branches and twigs and of removal of dead trees, the reduction in fruit quality owing to the use of resistant cultivars with low fruit quality (e.g. cv. Monachello) and the cost of additional fungicide sprays (Migheli et al., 2009). The disease also limits the use of susceptible, but very productive citrus species or cultivars with high fruit quality, particularly lemons (Migheli et al., 2009). Moreover, the presence of *P. tracheiphilus* in some areas could be an obstacle for the development of international breeding programmes for the genetic improvement of citrus owing to restrictions in the exchange of plant propagation material. Finally, new, promising highly productive cultivars, such as the virus-free nuclear clones, cannot be introduced in infested areas, because they are highly susceptible to the disease (Migheli et al., 2009, Nigro et al., 2011).

3.5.2.3. Environmental consequences

Environmental consequences are envisaged as a result of the additional fungicide treatments required to reduce disease incidence and severity once the pathogen is established in a new area. Copper compounds and mancozeb, which are the only plant protection products currently registered for citrus in the EU (Directive 91/414/CEE), have been associated with environmental concerns (Alva et al., 1993; Houeto et al., 1995).

3.6. Currently applied control methods in the EU

Once it enters the host, the pathogen invades the plant vessels and, thus, it is impossible to eliminate it by cultural practices and/or chemical measures. Nevertheless, any cultural practices and chemical measures applied in the infested EU MSs are aimed mainly at reducing inoculum sources and protecting the wounded/injured aerial plant parts from infection.

3.6.1. Cultural practices

Among cultural practices applied in the infested EU MSs for the control of *P. tracheiphilus*, careful pruning of diseased twigs/branches, timely removal of suckers and/or dead citrus trees are the most common practices (Salerno and Cutuli, 1981; Migheli et al., 2009). In some citrus-growing EU MSs (e.g. Spain, France and Italy), citrus plants are produced under certification programmes (Navarro et al., 2002). Such programmes prevent the introduction and further spread of *P. tracheiphilus* in new areas through the use of citrus planting material produced in certified nurseries. However, in other citrus-producing EU regions, such programmes are not fully operational (EFSA PLH Panel, 2014a).

3.6.2. Chemical control

It is considered that any fungicide sprays currently applied in the EU citrus-growing areas cannot control the disease in infested areas, particularly the "mal nero" and "mal fulminante" forms, or prevent the establishment of *P. tracheiphilus* in new areas. Some late-maturing mandarin hybrids are routinely sprayed in spring and autumn with copper-based fungicides or mancozeb for the control of Alternaria brown spot (Vicent et al., 2007, 2009). These chemicals could to some extent prevent the infection of wounded/injured aerial plant parts by the pathogen. However, the areas grown with cultivars susceptible to Alternaria brown spot represent a very minor proportion of the EU citrus-growing area, and in most of the EU citrus-growing areas no fungicide sprays are usually applied (EFSA PLH Panel, 2014a).

3.6.3. Host genetic resistance

The most effective control method against *P. tracheiphilus* would be the use of resistant citrus cultivars or clones grafted onto resistant rootstocks. In Italy and Greece, susceptible lemon cultivars (e.g. Femminello, Maglini, etc.) have been replaced in some areas by cultivars resistant/tolerant to *P. tracheiphilus* infection (e.g. Monachello, Femminello Ovale, Ermioni, etc.) (Russo, 1977; Thanassoulopoulos, 1991; Migheli et al., 2009). Nevertheless, the latter proved to have lower yield and/or fruit quality or inferior agronomic characteristics compared with the susceptible ones. *In vitro* selection was originally claimed as an innovative and promising technique to obtain lemon clones tolerant to the disease (Gentile et al., 2000). However, no commercial lemon cultivar obtained with this breeding method is presently available and no detailed information on yield, fruit quality, or agronomic characteristics has been reported (La Malfa and Gentile, 2005).

Currently, no citrus cultivars/clones or rootstocks resistant/tolerant to *P. tracheiphilus* infection that have competitive yields and satisfactory bioagronomic features are available for use on a large scale to control the disease in infested citrus-producing areas (Migheli et al., 2009).

3.6.4. Biological control

Although several *in vitro* and *in planta* studies have shown promising results on the potential control of *P. tracheiphilus* using non-pathogenic/*Agrobacterium*-mediated mutants of *P. tracheiphilus* or citrus bacterial and fungal endophytes, none of them is currently used in practice for the control of the disease in citrus orchards (De Cicco et al., 1986; Coco et al., 2004; Migheli et al., 2009).

3.7. Uncertainty

The main sources of uncertainties of this pest categorisation are listed below:

- Uncertainty on spread distance: The maximum distance, over which conidia of *P. tracheiphilus* could be dispersed by weather-related events, particularly wind-driven rain, is not known. There is only one scientific paper available on the distance of dispersal by natural means, according to which *P. tracheiphilus* conidia were trapped up to 16 m from the inoculum source. However, this distance was the maximum tested in that study.

- Uncertainty on pathways of entry and/or spread, other than host plants for planting: No information/evidence is available for citrus fruit and seed as a pathway of entry and spread of the pathogen.

- Uncertainty on factors affecting establishment: The fact that the pest is not established in some citrus-producing MSs, although host plants are present and climate is suitable, may imply that not all the factors affecting establishment are well known to date.

CONCLUSIONS

The Panel summarises in Table 9 below its conclusions on the key elements addressed in this scientific opinion in consideration of the pest categorisation criteria defined in ISPM 11 and ISPM 21 and of the additional questions formulated in the terms of reference.

Table. 9. Panel's conclusions on the pest categorisation criteria defined in the International Standards for Phytosanitary Measures No 11 and No 21 and on the additional questions formulated in the terms of references.

<table>
<tr><th>Criterion of pest categorisation</th><th>Panel's conclusions against ISPM 11 criterion
Yes/ No</th><th>Panel's conclusions against ISPM 21 criterion
Yes/ No</th><th>List of main uncertainties</th></tr>
<tr><td>Identity of the pest</td><td colspan="2">Is the identity of the pest clearly defined? Do clearly discriminative detection methods exist for the pest?
Yes, the pest satisfies this criterion.
Phoma tracheiphila has been recently reclassified as Plenodomus tracheiphilus (Petri) Gruyter, Aveskamp & Verkley. It is a single taxonomic entity and sensitive methods are available for its reliable detection and identification as well as for its differentiation from other related Plenodomus species.</td><td>-</td></tr>
<tr><td>Absence/ presence of the pest in the PRA area</td><td>Is the pest absent from all or a defined part of the PRA area?
Yes, P. tracheiphilus satisfies this criterion.
The pest is present in part of the risk assessment area, i.e. in four out of the eight citrus-producing MSs, namely Italy, Greece, Cyprus and France, where it is mainly restricted to lemon (Citrus limon). The pest is absent in other relevant EU citrus-growing areas, namely Spain, Portugal, Malta and Croatia.</td><td>Is the pest present in the PRA area?
Yes, P. tracheiphilus satisfies this criterion.
The pest is present in the risk assessment area.</td><td>-</td></tr>
<tr><td>Regulatory status</td><td colspan="2">In consideration that the pest under scrutiny is already regulated just mention in which annexes of 2000/29/EC and the marketing directives the pest and associated hosts are listed without further analysis. (the RM will have to consider the relevance of the regulation against official control)
Plenodomus tracheiphilus, as Phoma tracheiphila, and/or some of its hosts (i.e. Citrus spp., Poncirus spp., Fortunella spp.) are listed in Annexes IIAII, IVAI, IVAII, IVB, VAI and VBI of Council Directive 2000/29/EC (see section 3.3).
Host plants of the genus Severinia are not included in Council Directive 2000/29/EC</td><td>-</td></tr>
</table>

Potential establishment and spread	*Does the PRA area have ecological conditions (including climate and those in protected conditions) suitable for the establishment and spread of the pest?* *And, where relevant, are host species (or near relatives), alternate hosts and vectors present in the PRA area?* **Yes**, *P. tracheiphilus* satisfies this criterion. The main host, lemon (*C. limon*), as well as other susceptible hosts of the genera *Citrus*, *Fortunella*, *Poncirus* and *Severinia*, and their hybrids, are widely grown in the citrus-producing EU MSs. The ecoclimatic conditions in this part of the risk assessment area are suitable for establishment as well as for spread of the pathogen by both natural means and human assistance.	*Are plants for planting a pathway for introduction and spread of the pest?* **Yes**, *P. tracheiphilus* satisfies this criterion. *P. tracheiphilus* can spread via the movement of infected host plants for planting (rootstocks, grafted plants, scions, budwood, etc.), fruit peduncles and leaves, particularly latently infected (asymptomatic).	The pest is not known to occur in Portugal, Spain, Malta and Croatia, although susceptible hosts are present and the climatic conditions are suitable for its establishment and spread. The reasons for the absence of the pest in those areas are unknown.
Potential for consequences in the PRA area	*What are the potential for consequences in the PRA area?* *Provide a summary of impact in terms of yield and quality losses and environmental consequences.* In the infested citrus-growing EU MSs, *P. tracheiphilus* is causing damage in the form of twig and tree death, serious reduction in the volume of tree canopy and yield and quality losses. Indirect negative impact is associated with the additional cost of pruning dead branches/twigs and of removal of dead trees, reduction in fruit quality owing to the use of resistant/tolerant cultivars with low fruit quality and the cost of additional fungicide sprays. Environmental consequences are also envisaged as a result of the additional fungicide sprays required to reduce disease incidence and severity.	*If applicable is there indication of impact(s) of the pest as a result of the intended use of the plants for planting?* It was assumed that *P. tracheiphilus* spread from Greece to Italy and then to Tunisia through the movement of host plants for planting. In all three countries there are severe consequences for citrus production, particularly lemon.	-

Conclusion on pest categorisation	***P. tracheiphilus* has the potential to be a quarantine pest,** as it fulfils all of the pest categorisation criteria as those are defined in ISPM 11	***P. tracheiphilus* has the potential to be a regulated non-quarantine pest**, as it fulfils all of the pest categorisation criteria as those are defined in ISPM 21	-

Conclusion on specific ToR questions	*If the pest is already present in the EU, provide a brief summary of* - *the analysis of the present distribution of the organism in comparison with the distribution of the main hosts, and the distribution of hardiness/climate zones, indicating in particular if in the PRA area, the pest is absent from areas where host plants are present and where the ecological conditions (including climate and those in protected conditions) are suitable for its establishment,* *P. tracheiphilus* is present in part of the EU citrus-growing areas, namely Italy, Greece, Cyprus and France, but not in Portugal, Spain Croatia and Malta. The pest is also absent in other EU MSs, where citrus plants may be grown in gardens and/or greenhouses. There are no obvious ecoclimatic factors limiting the potential establishment of the pathogen in the so far non-infested citrus-producing EU MSs. - *the analysis of the observed impacts of the organism in the risk assessment area.* The disease is causing twig and plant death, a serious reduction in the tree canopy and yield and quality losses. Indirect pest effects are associated with additional costs of pruning dead branches/twigs and of removal of dead trees, reduction in fruit quality owing to the use of resistant cultivars with low fruit quality and the cost of additional fungicide sprays. Environmental consequences are also envisaged as a result of the additional fungicide sprays required to reduce disease incidence and severity.	-

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ABBREVIATIONS

EFSA:	European Food Safety Authority
EPPO:	European and Mediterranean Plant Protection Organization
EPPO-PQR:	European and Mediterranean Plant Protection Organization Plant Quarantine Retrieval System
EU:	European Union
ISPM:	International Standard for Phytosanitary Measures
MS(s):	Member State(s)
NPPO:	National Plant Protection Organisation
PLH Panel:	Plant Health Panel
RNQP:	Regulated Non Quarantine Pest