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Pest categorisation of *Diplodia bulgarica*

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Abstract

The EFSA Plant Health Panel performed a pest categorisation of *Diplodia bulgarica,* a clearly defined plant pathogenic fungus of the family Botryosphaeriaceae. The pathogen affects *Malus domestica, M. sylvestris* and *Pyrus communis* causing various symptoms such as canker, twig blight, gummosis, pre- and post-harvest fruit rot, dieback and tree decline. The pathogen is present in Asia (India, Iran, Türkiye) and in non-EU Europe (Serbia). Concerning the EU, the pathogen is present in Bulgaria and widespread in Germany. There is a key uncertainty on the geographical distribution of *D. bulgarica* worldwide and in the EU, because in the past, when molecular tools were not available, the pathogen might have been misidentified as other *Diplodia* species (e.g. *D. intermedia, D. malorum, D. mutila, D. seriata*) or other members of the Botryosphaeriaceae family affecting apple and pear based only on morphology and pathogenicity tests. *Diplodia bulgarica* is not included in Commission Implementing Regulation (EU) 2019/2072. Plants for planting, other than seeds, fresh fruits, and bark and wood of host plants as well as soil and other plant-growing media carrying plant debris are the main pathways for the further entry of the pathogen into the EU. Host availability and climate suitability factors are favourable for the further establishment of the pathogen in the EU. In the areas of its present distribution, including Germany, the pathogen has a direct impact on cultivated hosts. Phytosanitary measures are available to prevent the further introduction and spread of the pathogen into the EU. *Diplodia bulgarica* satisfies the criteria that are within the remit of EFSA to assess for this species to be regarded as potential Union quarantine pest.

Keywords: black canker of apple and pear, *Malus* spp. *Pyrus communis*, pest risk, plant health, plant pest, quarantine

Requestor: European Commission

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1. Introduction

1.1. Background and terms of reference as provided by the requestor

1.1.1. Background

The new Plant Health Regulation (EU) 2016/2031, on the protective measures against pests of plants, is applying from 14 December 2019. Conditions are laid down in this legislation in order for pests to qualify for listing as Union quarantine pests, protected zone quarantine pests or Union regulated non-quarantine pests. The lists of the EU regulated pests together with the associated import or internal movement requirements of commodities are included in Commission Implementing Regulation (EU) 2019/2072. Additionally, as stipulated in the Commission Implementing Regulation 2018/2019, certain commodities are provisionally prohibited to enter in the EU (high risk plants, HRP). EFSA is performing the risk assessment of the dossiers submitted by exporting to the EU countries of the HRP commodities, as stipulated in Commission Implementing Regulation 2018/2018. Furthermore, EFSA has evaluated a number of requests from exporting to the EU countries for derogations from specific EU import requirements.

In line with the principles of the new plant health law, the European Commission with the Member States are discussing monthly the reports of the interceptions and the outbreaks of pests notified by the Member States. Notifications of an imminent danger from pests that may fulfil the conditions for inclusion in the list of the Union quarantine pest are included. Furthermore, EFSA has been performing horizon scanning of media and literature.

As a follow-up of the above-mentioned activities (reporting of interceptions and outbreaks, HRP, derogation requests and horizon scanning), a number of pests of concern have been identified. EFSA is requested to provide scientific opinions for these pests, in view of their potential inclusion by the risk manager in the lists of Commission Implementing Regulation (EU) 2019/2072 and the inclusion of specific import requirements for relevant host commodities, when deemed necessary by the risk manager.

1.1.2. Terms of reference

EFSA is requested, pursuant to Article 29(1) of Regulation (EC) No 178/2002, to provide scientific opinions in the field of plant health.

EFSA is requested to deliver 53 pest categorisations for the pests listed in Annex 1A, 1B, 1D and 1E (for more details see mandate M-2021-00027 on the Open.EFSA portal). Additionally, EFSA is requested to perform pest categorisations for the pests so far not regulated in the EU, identified as pests potentially associated with a commodity in the commodity risk assessments of the HRP dossiers (Annex 1C; for more details see mandate M-2021-00027 on the Open.EFSA portal). Such pest categorisations are needed in the case where there are not available risk assessments for the EU.

When the pests of Annex 1A are qualifying as potential Union quarantine pests, EFSA should proceed to phase 2 risk assessment. The opinions should address entry pathways, spread, establishment, impact and include a risk reduction options analysis.

Additionally, EFSA is requested to develop further the quantitative methodology currently followed for risk assessment, in order to have the possibility to deliver an express risk assessment methodology. Such methodological development should take into account the EFSA Plant Health Panel Guidance on quantitative pest risk assessment and the experience obtained during its implementation for the Union candidate priority pests and for the likelihood of pest freedom at entry for the commodity risk assessment of High Risk Plants.

1.2. Interpretation of the terms of reference

Diplodia bulgarica is one of a number of pests listed in Annex 1C to the Terms of Reference (ToR) to be subject to pest categorisation to determine whether it fulfils the criteria of a potential Union quarantine pest for the area of the EU excluding Ceuta, Melilla and the outermost regions of Member States referred to in Article 355(1) of the Treaty on the Functioning of the European Union (TFEU), other than Madeira and the Azores, and so inform EU decision-making as to its appropriateness for potential inclusion in the lists of pests of Commission Implementing Regulation (EU) 2019/ 2072. If a pest fulfils the criteria to be potentially listed as a Union quarantine pest, risk reduction options will be identified.

1.3. Additional information

This pest categorisation was initiated following the commodity risk assessment of *Malus domestica* plants from Türkiye performed by EFSA (EFSA PLH Panel, 2022), in which *D. bulgarica* was identified as a relevant non-regulated EU pest which could potentially enter the EU on *M. domestica*.

2. Data and methodologies

2.1. Data

2.1.1. Information on pest status from NPPOs

In the context of the current mandate, EFSA is preparing pest categorisations for new/emerging pests that are not yet regulated in the EU. When official pest status is not available in the European and Mediterranean Plant Protection Organization (EPPO) Global Database (EPPO, online), EFSA consults the NPPOs of the relevant MSs. To obtain information on the official pest status for *Diplodia bulgarica*, EFSA has consulted the NPPOs of Bulgaria and Germany. The results of this consultation are presented in Section 3.2.2.

2.1.2. Literature search

A literature search on *Diplodia bulgarica* was conducted at the beginning of the categorisation in the ISI Web of Science bibliographic database, using the scientific name of the pest as search term. Papers relevant for the pest categorisation were reviewed, and further references and information were obtained from experts, as well as from citations within the references and grey literature.

2.1.3. Database search

Pest information, on host(s) and distribution, was retrieved from the European and Mediterranean Plant Protection Organization (EPPO) Global Database (EPPO, online), the CABI databases and scientific literature databases as referred above in Section 2.1.1.

Data about the import of commodity types that could potentially provide a pathway for the pest to enter the EU and about the area of hosts grown in the EU were obtained from EUROSTAT (Statistical Office of the European Communities).

The Europhyt and TRACES databases were consulted for pest-specific notifications on interceptions and outbreaks. Europhyt is a web-based network run by the Directorate General for Health and Food Safety (DG SANTÉ) of the European Commission as a subproject of PHYSAN (Phyto-Sanitary Controls) specifically concerned with plant health information. TRACES is the European Commission's multilingual online platform for sanitary and phytosanitary certification required for the importation of animals, animal products, food and feed of non-animal origin and plants into the European Union, and the intra-EU trade and EU exports of animals and certain animal products. Up until May 2020, the Europhyt database managed notifications of interceptions of plants or plant products that do not comply with EU legislation, as well as notifications of plant pests detected in the territory of the Member States and the phytosanitary measures taken to eradicate or avoid their spread. The recording of interceptions switched from Europhyt to TRACES in May 2020.

GenBank was searched to determine whether it contained any nucleotide sequences for *D. bulgarica* which could be used as reference material for molecular diagnosis. GenBank® (www.ncbi.nlm.nih.gov/genbank/) is a comprehensive publicly available database that as of August 2019 (release version 227) contained over 6.25 trillion base pairs from over 1.6 billion nucleotide sequences for 450,000 formally described species (Sayers et al., 2020).

2.2. Methodologies

The Panel performed the pest categorisation for *D. bulgarica*, following guiding principles and steps presented in the EFSA guidance on quantitative pest risk assessment (EFSA PLH Panel, 2018), the EFSA guidance on the use of the weight of evidence approach in scientific assessments (EFSA Scientific Committee, 2017) and the International Standards for Phytosanitary Measures No. 11 (FAO, 2013).

The criteria to be considered when categorising a pest as a potential Union quarantine pest (QP) is given in Regulation (EU) 2016/2031 Article 3 and Annex I, Section 1 of the Regulation. Table 1

presents the Regulation (EU) 2016/2031 pest categorisation criteria on which the Panel bases its conclusions. In judging whether a criterion is met, the Panel uses its best professional judgement (EFSA Scientific Committee, 2017) by integrating a range of evidence from a variety of sources (as presented above in Section 2.1) to reach an informed conclusion as to whether or not a criterion is satisfied.

The Panel's conclusions are formulated respecting its remit and particularly with regard to the principle of separation between risk assessment and risk management (EFSA founding regulation (EU) No 178/2002); therefore, instead of determining whether the pest is likely to have an unacceptable impact, deemed to be a risk management decision, the Panel will present a summary of the observed impacts in the areas where the pest occurs, and make a judgement about potential likely impacts in the EU. Whilst the Panel may quote impacts reported from areas where the pest occurs in monetary terms, the Panel will seek to express potential EU impacts in terms of yield and quality losses and not in monetary terms, in agreement with the EFSA guidance on quantitative pest risk assessment (EFSA PLH Panel, 2018). Article 3 (d) of Regulation (EU) 2016/2031 refers to unacceptable social impact as a criterion for quarantine pest status. Assessing social impact is outside the remit of the Panel.

Table 1: Pest categorisation criteria under evaluation, as derived from Regulation (EU) 2016/2031 on protective measures against pests of plants (the number of the relevant sections of the pest categorisation is shown in brackets in the first column)

Criterion of pest categorisation	Criterion in Regulation (EU) 2016/2031 regarding Union quarantine pest (article 3)
Identity of the pest (Section 3.1)	Is the identity of the pest clearly defined, or has it been shown to produce consistent symptoms and to be transmissible?
Absence/presence of the pest in the EU territory (Section 3.2)	Is the pest present in the EU territory? If present, is the pest in a limited part of the EU or is it scarce, irregular, isolated or present infrequently? If so, the pest is considered to be not widely distributed.
Pest potential for entry, establishment and spread in the EU territory (Section 3.4)	Is the pest able to enter into, become established in, and spread within, the EU territory? If yes, briefly list the pathways for entry and spread.
Potential for consequences in the EU territory (Section 3.5)	Would the pests' introduction have an economic or environmental impact on the EU territory?
Available measures (Section 3.6)	Are there measures available to prevent pest entry, establishment, spread or impacts? If already present in the EU are measures available to slow spread or facilitate eradication?
Conclusion of pest categorisation (Section 4)	A statement as to whether (1) all criteria assessed by EFSA above for consideration as a potential quarantine pest were met and (2) if not, which one(s) were not met.

3. Pest categorisation

3.1. Identity and biology of the pest

3.1.1. Identity and taxonomy

Is the identity of the pest clearly defined, or has it been shown to produce consistent symptoms and/or to be transmissible?

Yes, the identity of *Diplodia bulgarica* is clearly defined and the pathogen has been shown to produce consistent symptoms and to be transmissible.

Diplodia bulgarica A.J.L. Phillips, J. Lopes & S.G. Bobev is a plant pathogenic fungus of the family Botryospheriaceae. The pathogen was first described in 2012 on *Malus sylvestris* in Bulgaria and on *Malus domestica* in Iran (Phillips et al., 2012).

The EPPO Global Database (EPPO, online) provides the following taxonomic identification for *D. bulgarica*:

Preferred name: *Diplodia bulgarica* Phillips AJL, Lopes J and Bobev SG
Order: Botryosphaeriales
Family: Botryosphaeriaceae
Genus: *Diplodia*
Species: *Diplodia bulgarica*
Common names: black canker of apple and pear, apple canker.

The EPPO code[1] (Griessinger and Roy, 2015; EPPO, 2019) for this species is: DIPBBU (EPPO, online).

3.1.2. Biology of the pest

Diplodia is an important genus of the family Botryosphaeriaceae, with more than 1000 species being pathogenic, endophytic or saprophytic on a wide range of mainly woody plants (Phillips et al., 2012). As pathogens, they have been reported to cause different disease symptoms on their hosts, such as canker, twig blight, gummosis, pre- and post-harvest fruit rot, dieback and tree decline (Crous et al., 2006; Phillips et al., 2007, 2012; Slippers and Wingfield, 2007; Lazzizera et al., 2008; Abdollahzadeh, 2015; Hanifeh et al., 2017). *Diplodia bulgarica* is one of the several *Diplodia* species (e.g. *D. intermedia, D. malorum, D. mutila, D. seriata, D. pseudoseriata*) described on apple worldwide (Hanifeh et al., 2013, 2014; Delgado-Cerrone et al., 2016; Sessa et al., 2016; Vučković et al., 2022).

Although no specific information is available on the biology and epidemiology of the pathogen, it may be assumed that its life cycle is similar to that of other species of the genus *Diplodia* such as *D. seriata,* a widely distributed pathogen that causes canker, dieback, fruit rot and leaf spot diseases on several economically important forest and horticultural species (Farr and Rossman, 2023). The pathogen is most likely capable to overwinter on dead plant organs (twigs, branches, mummified fruit) and on plant debris in the soil mainly in the form of mycelium and pycnidia. The conidia released from pycnidia during wet weather are dispersed by water (overhead irrigation, rain, windblown rain) over relatively short distances to infect susceptible hosts. Although there is no reported evidence, conidia of *D. bulgarica* could potentially be passively dispersed between hosts by insects (mainly of the Buprestidae and Cerambycidae families) similarly to other *Diplodia* species (Epstein et al., 2008; Panzavolta et al., 2018). The pathogen infects primarily through wounds, although entry through natural openings (lenticels, stomata) or direct penetration of the host tissues is also possible (CABI, 2022). Similarly to other members of the family Botryosphaeriaceae, *Diplodia* spp. can live endophytically inside their host plants (Crous et al., 2006). Symptoms on wood and fruits may not become visible for several weeks after infection (CABI, 2022). The conidia germinate at temperatures between 15°C and 37°C. Infection is favoured by conditions that can stress the plant, such as drought, frost or hail damage, poor nutrition and improper pruning practices (CABI, 2022). In Iran, black canker disease caused by *D. bulgarica* has been reported to be prevalent in apple trees more than 15-year-old suffering from environmental stress, such as drought and nutrient deficiency (Hanifeh et al., 2017). According to Nourian et al. (2021), the optimal temperature for the *in vitro* mycelial growth of *D. bulgarica* is 25°C; the pathogen could still grow at 10°C but not at 35°C. There is no evidence that *D. bulgarica* is seed-borne, although other *Diplodia* species or other members of the *Botryosphaeriaceae* have been shown to be seed-borne (Gure et al., 2005).

No sexual stage for *D. bulgarica* has been reported so far. Similarly, the sexual stage has rarely been observed in other species of *Diplodia,* and hence, these have previously been considered as mainly reproducing asexually (Zeynali Bari et al., 2021). However, recent identification of mating type (MAT) idiomorphs in the genome of several *Diplodia* species provided indirect evidence for occurrence of a cryptic, heterothallic sexual reproduction in their life cycle (Bihon et al., 2014; Lopes et al., 2018). Although there is no information either on the presence of MAT idiomorphs in the genome or occurrence of sexual reproduction in *D. bulgarica,* Zeynali Bari et al. (2021) assumed that the pathogen probably has a cryptic sexual stage that contributed to the high number of genotypes detected in the West Azerbaijan province of Iran.

[1] An EPPO code, formerly known as a Bayer code, is a unique identifier linked to the name of a plant or plant pest important in agriculture and plant protection. Codes are based on genus and species names. However, if a scientific name is changed, the EPPO code remains the same. This provides a harmonised system to facilitate the management of plant and pest names in computerised databases, as well as data exchange between IT systems (Griessinger and Roy, 2015; EPPO, 2019).

3.1.3. Host range/species affected

Diplodia bulgarica has a very limited natural host range in the family Rosaceae; the pathogen has been reported so far on *Malus domestica* (Phillips et al., 2012; Abdollahzadeh, 2015; Hanifeh et al., 2017; Nabi et al., 2020; Eken, 2021; Hinrichs-Berger et al., 2021; Nourian et al., 2021; Zeynali Bari et al., 2021), *M. sylvestris* (Phillips et al., 2012, 2013) and *Pyrus communis* (Hinrichs-Berger et al., 2021).

According to Ketabchi et al. (2015), 1- to 2-year-old detached shoots of *Corylus avellana* (hazelnut), *Pistacia vera* (pistachio), *Prunus domestica* (plum) and *Salix* spp. (willow and pussy willow) trees showed bark and wood discoloration and drying 4 months after their artificial wound inoculation with isolates of *D. bulgarica*. However, only the abstract of that study is available in which no detailed information is provided on the experimental materials and methods, including the origin of the isolates and the method used for their identification. Therefore, there is uncertainty on whether the above-mentioned plant species could be considered as experimental hosts of *D. bulgarica* and if these species could be infected by the pathogen under natural conditions.

A detailed list of *D. bulgarica* hosts reported so far in the literature is included in Appendix A (last updated 27 March 2023).

3.1.4. Intraspecific diversity

In 2017 and 2018, a study was conducted on vegetative compatibility and aggressiveness diversity of 101 isolates of *D. bulgarica* recovered from apple trees (more than 10-year-old) exhibiting symptoms of canker, dieback and decline in the major apple production areas of West Azerbaijan province of Iran (Zeynali Bari et al., 2021). Inter-simple sequence repeat (ISSR) marker analyses revealed high diversity within populations, low genetic differentiation, high gene flow and sharing of multilocus genotypes (MLGs) among geographic populations. Vegetative compatibility analyses revealed the occurrence of anastomosis between non-self pairings and high vegetative compatibility group diversity within populations. All studied MLGs produced necrotic lesions on artificially inoculated detached shoots of apple (cv. Red Delicious) but differed in their aggressiveness (Zeynali Bari et al., 2021).

Although no sexual stage for *D. bulgarica* has been reported so far, the potential ability of the pathogen to differentiate sexual reproductive stages (see Section 3.1.2) may enhance its genomic plasticity and adaptation to various adverse environmental conditions, including fungicide exposure.

3.1.5. Detection and identification of the pest

Are detection and identification methods available for the pest?

Yes, there are methods available for the detection and identification of *Diplodia bulgarica* and its discrimination from other *Diplodia* species or other fungi of the family Botryosphaeriaceae.

Diplodia bulgarica causes cankers on trunks and limbs, twig blight, gummosis, pre- and post-harvest fruit rot, bark and vascular discoloration of infected shoots, dieback and tree decline (Abdollahzadeh, 2015; Hinrichs-Berger et al., 2021; Nourian et al., 2021). As the infection develops, the bark separates from the underlying wood and falls to the ground, whereas the wood beneath it appears blackened and looks like charcoal. In some cases, the trees are girdled by the cankers and die (Hinrichs-Berger et al., 2021). The above-mentioned symptoms are similar to those caused by other *Diplodia* species or other biotic (fungi, bacteria, etc.) or abiotic agents. Pycnidia of the pathogen may be detected using a magnifying lens on the affected plant tissues, particularly on older cankers (Hinrichs-Berger et al., 2021). However, they are similar in morphology to those of other *Diplodia* species or other fungal species of the family Botryosphaeriaceae affecting apple and pear (Slippers et al., 2007; Cloete et al., 2011; Phillips et al., 2012). In addition, the pathogen may remain quiescent or latent within its asymptomatic hosts (see section 3.1.2). Based on the above, it is unlikely that *D. bulgarica* could be detected only by visual inspection of its host plants.

Diplodia bulgarica can be readily isolated on culture media and description of its cultural and morphological characteristics is available in the literature (Phillips et al., 2012; Nourian et al., 2021) (Figure 1). In the past, the identification of species of the family Botryosphaeriaceae, including species of the genus *Diplodia,* was based on cultural and morphological characteristics (Slippers et al., 2013). However, morphological features alone are inadequate to define genera or identify species within Botryosphaeriaceae, since conidial dimensions are overlapping, whereas their septation and pigmentation is influenced by the cultural conditions (Phillips et al., 2013; Slippers et al., 2013).

Morphology combined with multilocus sequence analysis of the internal transcribed spacer (ITS) region and the elongation factor 1-α (*EF1-α*) is currently used to reliably identify *D. bulgarica* in culture and discriminate it from other morphologically similar *Diplodia* species affecting *Malus* spp. (e.g. *D. malorum, D. intermedia*) or other phylogenetically related *Diplodia* species, such as *D. cupressi* and *D. tsugae* (Phillips et al., 2012; Nabi et al., 2020; Nourian et al., 2021; Zeynali Bari et al., 2021). There are no species-specific primers available for the in planta identification of the pathogen.

Nucleotide sequences of *D. bulgarica* are available in GenBank (https://www.ncbi.nlm.nih.gov/genbank) and could be used as reference material for molecular diagnosis.

No EPPO Standard is available for the detection and identification of *D. bulgarica*.

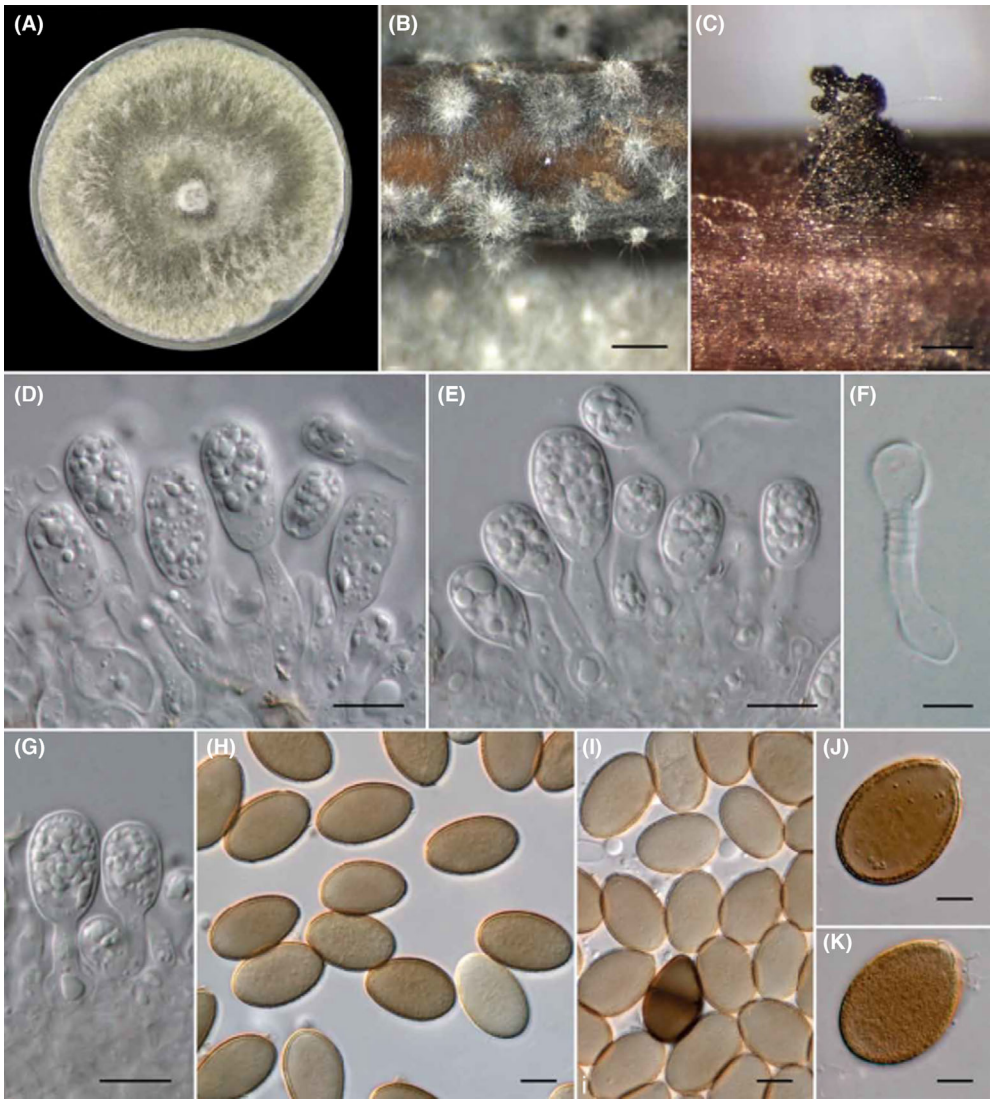

Figure 1: *Diplodia bulgarica*. (a) Culture growing on PDA; (b) pycnidia developing on pine needles in culture; (c) pycnidium on pine needle exuding conidia; (d–g) conidiogenous cells with developing conidia; (h) brown, aseptate conidia; (i) brown aseptate conidia and a two-celled conidium; (j, k) conidium in two levels of focus showing finely verruculose inner surface of the conidium wall. Scale bars: b = 500 μm; c = 200 μm; d–i = 10 μm; j, k = 5 μm (from Phillips et al., 2012)

3.2. Pest distribution

3.2.1. Pest distribution outside the EU

Diplodia bulgarica has been reported to be present in Asia (India, Iran, Türkiye) and in non-EU Europe (Serbia) (Abdollahzadeh, 2015; Hanifeh et al., 2017; Nabi et al., 2020; Eken, 2022; Vučković

et al., 2022). The current geographical distribution of *D. bulgarica* is shown in Figure 2. A complete list of the countries and states/provinces from where the pathogen has been reported is included in Appendix B.

Figure 2: Global distribution of *Diplodia bulgarica* [Data Source: CABI, 2022 (last accessed on 15 March 2023) and other literature sources]

There is uncertainty about the geographical distribution of *D. bulgarica* outside the EU, because in the past, when molecular tools were not available, the pathogen might have been misidentified as other *Diplodia* species (e.g. *D. intermedia, D. malorum, D. mutila, D. seriata*) or other members of the Botryosphaeriaceae family affecting apples and pears based only on morphology and pathogenicity tests. Therefore, the geographical distribution of the pathogen might be wider than the one reported so far.

3.2.2. Pest distribution in the EU

Is the pest present in the EU territory? If present, is the pest in a limited part of the EU or is it scarce, irregular, isolated or present infrequently? If so, the pest is considered to be not widely distributed.

Yes, *Diplodia bulgarica* has been reported from Bulgaria and Germany.

Diplodia bulgarica has been reported from Bulgaria (Phillips et al., 2012) and Germany (Hinrichs-Berger et al., 2021; Koch et al., 2022).

The report from Bulgaria is based on a phylogenetic study conducted by Phillips et al. (2012), in which three *D. bulgarica* strains (i.e. CBS 124135, CBS 124254, CBS 124136) isolated from *M. sylvestris* in Bulgaria (Plovdiv) and maintained in the Fungal Collection of the Westerdijk Fungal Biodiversity Institute (The Netherlands; https://wi.knaw.nl/) were used. There are no other reports available in the literature on the occurrence of the pathogen in Bulgaria.

According to the Bulgarian NPPO (May 2023), *D. bulgarica* is not well studied in Bulgaria. The available information is insufficient, apart from the report in the region of Plovdiv in 2012 (Phillips et al., 2012). Till now, there are no data on further spread in Bulgaria. Although surveys for quarantine pests on fruit species have been carried out for more than 20 years in Bulgaria, no samples suspected of the presence of this fungal pathogen have been received in the Bulgarian Central Laboratory for Plant Quarantine. Considering the available data, the NPPO concludes that *D. bulgarica* should be considered as 'Present, no details' in Bulgaria.

According to the German NPPO (March 2023), the pathogen was first reported causing black rot canker on apple (*M. domestica*) and pear (*P. communis*) in organic pome orchards in Baden-Wuerttemberg (Hinrichs-Berger et al., 2021). A large survey carried out during November 2021–

September 2022 focusing on black rot canker symptoms in apple and pear orchards in 16 Federal States resulted in the detection of *D. bulgarica* in 137 out of 445 samples collected (31%). Of the above-mentioned positive samples, 135 derived from *M. domestica* trees and two from *P. communis* trees (unpublished data). These data show distribution of *D. bulgarica* in 12 out of the 16 Federal States, thus indicating that the pathogen is widespread in Germany. The NPPO also mentioned that phytosanitary measures (curative and preventive) are applied in the infested orchards. These measures include pruning of symptomatic branches, removal of severely infected trees, application of fungicides, protection of wounds, white tree trunk painting for preventing sunscald that causes cracks to the bark, etc.

A key uncertainty concerns the current geographical distribution of *D. bulgarica* in the EU, which might be wider than that reported, for the reasons listed in Section 3.2.1. In addition, given that the pathogen is widely distributed in Germany and there is a lot of trade of host planting material from Germany to other pome-growing EU MSs, *D. bulgarica* could also be present in other EU MSs.

3.3. Regulatory status

3.3.1. Commission implementing regulation 2019/2072

Diplodia bulgarica is not listed in Annex II of Commission Implementing Regulation (EU) 2019/2072, an implementing act of Regulation (EU) 2016/2031, or in any emergency plant health legislation.

3.3.2. Hosts or species affected that are prohibited from entering the union from third countries

A list of hosts included in Annex VI of Commission Implementing Regulation (EU) 2019/2072 is provided in Table 2. Hosts of the genera *Malus* are included in the Commission Implementing Regulation (EU) 2018/2019 on high-risk plants.

Table 2: List of plants, plant products and other objects that are *Diplodia bulgarica* hosts whose introduction into the Union from certain third countries is prohibited (Source: Commission Implementing Regulation (EU) 2019/2072, Annex VI)

List of plants, plant products and other objects whose introduction into the Union from certain third countries is prohibited			
	Description	**CN code**	**Third country, group of third countries or specific area of third country**
8.	Plants for planting of [. . .] *Malus* Mill., [. . .] *Pyrus* L. [. . .] other than dormant plants free from leaves, flowers and fruits	ex 0602 10 90 ex 0602 20 20 ex 0602 20 80 ex 0602 40 00 ex 0602 90 41 ex 0602 90 45 ex 0602 90 46 ex 0602 90 47 ex 0602 90 48 ex 0602 90 50 ex 0602 90 70 ex 0602 90 91 ex 0602 90 99	Third countries other than Albania, Andorra, Armenia, Azerbaijan, Belarus, Bosnia and Herzegovina, Canary Islands, Faroe Islands, Georgia, Iceland, Liechtenstein, Moldova, Monaco, Montenegro, North Macedonia, Norway, Russia (only the following parts: Central Federal District (Tsentralny federalny okrug), Northwestern Federal District (Severo-Zapadny federalny okrug), Southern Federal District (Yuzhny federalny okrug), North Caucasian Federal District (Severo-Kavkazsky federalny okrug) and Volga Federal District (Privolzhsky federalny okrug)), San Marino, Serbia, Switzerland, Türkiye, Ukraine and the United Kingdom
9.	Plants for planting of [. . .] *Malus* Mill., [. . .]. and *Pyrus* L. and their hybrids, and *Fragaria* L., other than seeds	ex 0602 10 90 ex 0602 20 20 ex 0602 90 30 ex 0602 90 41 ex 0602 90 45 ex 0602 90 46 ex 0602 90 48 ex 0602 90 50 ex 0602 90 70 ex 0602 90 91	Third countries other than Albania, Algeria, Andorra, Armenia, Australia, Azerbaijan, Belarus, Bosnia and Herzegovina, Canada, Canary Islands, Egypt, Faroe Islands, Georgia, Iceland, Israel, Jordan, Lebanon, Libya, Liechtenstein, Moldova, Monaco, Montenegro, Morocco, New Zealand, North Macedonia, Norway, Russia (only the following parts: Central Federal District (Tsentralny federalny okrug), Northwestern Federal District (Severo- Zapadny federalny okrug), Southern Federal District (Yuzhny federalny okrug), North Caucasian Federal District (Severo- Kavkazsky federalny okrug) and

List of plants, plant products and other objects whose introduction into the Union from certain third countries is prohibited			
	Description	**CN code**	**Third country, group of third countries or specific area of third country**
		ex 0602 90 99	Volga Federal District (Privolzhsky federalny okrug)), San Marino, Serbia, Switzerland, Syria, Tunisia, Türkiye, Ukraine, the United Kingdom (1) and United States other than Hawaii
19.	Soil as such consisting in part of solid organic substances	ex 2530 90 00 ex 3824 99 93	Third countries other than Switzerland
20.	Growing medium as such, other than soil, consisting in whole or in part of solid organic substances, other than that composed entirely of peat or fibre of *Cocos nucifera* L., previously not used for growing of plants or for any agricultural purposes	ex 2530 10 00 ex 2530 90 00 ex 2703 00 00 ex 3101 00 00 ex 3824 99 93	Third countries other than Switzerland

3.4. Entry, establishment and spread in the EU

3.4.1. Entry

Is the pest able to enter into the EU territory? If yes, identify and list the pathways.

Yes, *Diplodia bulgarica* can further enter into the EU territory via host plants for planting, fruits, parts of host plants (e.g. branches, bark, wood) and soil/plant growing media carrying infected host plant debris.

Comment on plants for planting as a pathway.

Plants for planting is a main pathway of the further entry of the pathogen into the EU.

The Panel identified the following main pathways for the further entry of *D. bulgarica* into the EU:

1) host plants for planting, other than seeds,
2) fresh fruits of host plants,
3) wood and bark of host plants and
4) soil and other plant-growing media carrying infected host plant debris,

all originating in infested third countries.

Similarly to other *Diplodia* species or other members of the Botryosphaeriaceae family (Tattar, 1989; Gure et al., 2005; Decourcelle et al., 2014), *D. bulgarica* could be potentially transmitted via seeds, although so far there has been no evidence for the pathogen being seedborne. However, this is considered a minor pathway of entry, as apples and pears are mainly propagated by grafting.

Diplodia bulgarica could potentially further enter the EU by natural means (particularly wind-driven rain, insects, birds, etc.) from infested third countries such as Serbia and Türkiye. In Serbia, the pathogen has been reported from apple orchards and fruit storage facilities in Radmilovac region (Vučković et al., 2022), which is located near Belgrade. In Türkiye, *D. bulgarica* has been detected in apple fruit storage facilities in Eğirdir district of Isparta province (Eken, 2022). Although both of those regions are far from the EU borders, there is uncertainty about the presence of the pathogen in other apple and pear production areas of Serbia and Türkiye neighbouring EU MS.

Although there are no quantitative data available, viable conidia of the pathogen may also be present as contaminants on other substrates or objects (e.g. non-host plants, second-hand agricultural

machinery and equipment, crates, etc.) imported into the EU. Nevertheless, these are considered minor pathways for the further entry of the pathogen into the EU.

A list of all the potential pathways for the further entry of *D. bulgarica* into the EU is included in Table 3.

Table 3: Potential pathways for the further entry of *Diplodia bulgarica* into the EU 27

Pathways (e.g. host/ intended use/source)	Life stage	Relevant mitigations [e.g. prohibitions (Annex VI), special requirements (Annex VII) or phytosanitary certificates (Annex XI) within Implementing Regulation 2019/2072]
Host plants for planting, other than seeds	Mycelium, pycnidia	Plants for planting, other than seeds, that are hosts of *D. bulgarica* and are prohibited from being imported from third countries (Regulation 2019/2072, Annex VI) are listed in Table 2. There is a temporary prohibition for high-risk plants (Regulation 2018/2019).
Seeds of host plants for sowing	Mycelium	No relevant mitigations exist within Commission Implementing Regulation (EU) 2019/2072.
Fresh fruits of host plants	Mycelium, pycnidia	A phytosanitary certificate is required for the introduction into the Union from third countries, other than Switzerland, of fruits (fresh or chilled) of *Malus* and *Pyrus* [Annex XI, Part A (5) of Commission Implementing Regulation (EU) 2019/2072]
Parts of host plants, other than fruits and seeds	Mycelium, pycnidia	A phytosanitary certificate is required for the introduction into the Union from third countries, other than Switzerland, of foliage, branches and other parts of plants of *Malus* and *Pyrus*, without flowers or flower buds [Annex XI, Part A (3) of Commission Implementing Regulation (EU) 2019/2072].
Wood with bark of host plants	Mycelium and pycnidia (the latter formed only in the bark)	A phytosanitary certificate is required for the introduction into the Union from Canada and United States of *Malus* and *Pyrus* wood in the rough, not stripped of bark or sapwood [Annex XI, Part A (12) of Commission Implementing Regulation (EU) 2019/ 2072].
Soil as such not attached or associated with plants for planting carrying infected plant debris	Mycelium, pycnidia	The introduction into the Union from third countries, other than Switzerland, of soil as such consisting in part of solid organic substances is banned [Annex VI (19) of Commission Implementing Regulation (EU) 2019/2072].
Growing medium, attached to or associated with host and non-host plants for planting, carrying infected plant debris, with the exception of sterile medium of *in vitro* plants	Mycelium, pycnidia	A phytosanitary certificate and is required for the introduction into the Union from third countries, other than Switzerland, of growing medium attached to or associated with plants, intended to sustain the vitality of the plants [Annex XI, Part A (1) of Commission Implementing Regulation (EU) 2019/2072]. Special requirements also exist for this commodity [Annex VII (1) of Commission Implementing Regulation (EU) 2019/2072].
Machinery and vehicles with contaminated soil and/or infected debris of host plants.	Mycelium, pycnidia	A phytosanitary certificate is required for the introduction into the Union of machinery and vehicles from third countries, other than Switzerland [Annex XI, Part A (1) of Commission Implementing Regulation (EU) 2019/2072]. Special requirements also exist for this commodity [Annex VII (2) of Commission Implementing Regulation (EU) 2019/2072]

The quantity of fresh produce of hosts imported into the EU from countries where *D. bulgarica* is present is provided in Table 4 and Appendix C.

Table 4: EU 27 annual imports of fresh produce of hosts from countries where *Diplodia bulgarica* is present, 2016–2020 (in 100 kg) Source: Eurostat accessed on 16 March 2023

Commodity	HS code	2016	2017	2018	2019	2020
Fresh apples	0808 10	259,659.56	229,426.57	125,394.25	342,757.25	233,821.87
Fresh pears	0808 30	20,526.96	46,304.06	81,496.69	76,970.73	133,214.89
	Sum	**280,186.52**	**275,730.63**	**206,890.94**	**419,727.98**	**367,036.76**

Notifications of interceptions of harmful organisms began to be compiled in Europhyt in May 1994 and in TRACES in May 2020. As of April 2023, there were no records of interception of *D. bulgarica* in the Europhyt and TRACES databases. However, since *D. bulgarica* is not a quarantine pest, the EU MS have no obligation to notify interceptions of the pathogen via Europhyt.

3.4.2. Establishment

Is the pest able to become established in the EU territory?

Yes, *Diplodia bulgarica* has been reported from Bulgaria and has established in Germany (see section 3.2.2). Both biotic (host availability) and abiotic (climate suitability) factors occurring in the EU suggest that the pathogen could further establish in other parts of the EU territory where apples and pears are grown, similarly to other *Diplodia* species (e.g. *D. intermedia, D. malorum, D. mutila, D. seriata*) affecting these hosts in the EU.

Diplodia bulgarica could potentially be transferred from the pathways of entry to the host plants grown in the EU via splash-dispersed conidia, contaminated soil or other plant-growing media associated with plants for planting, surface (rain or irrigation) water and possibly insects, birds and small animals (see Section 3.4.3). The frequency of this transfer depends on the volume and frequency of the imported commodities, their destination (e.g. nurseries, retailers, packinghouses) and its proximity to the hosts grown in the EU territory, as well as on the management of plant debris and fruit waste.

3.4.2.1. EU distribution of main host plants

Diplodia bulgarica natural hosts (i.e. apples, pears) are widely distributed in the EU territory, in commercial production (orchards, nurseries) and in home gardens. The harvested area of apples and pears cultivated in the EU in recent years is shown in Tables 5 and 6, respectively.

Table 5: Harvested area of apples (code: F1110) in the EU, 2016–2020 (1,000 ha). Source EUROSTAT (accessed 21 March 2023)

Apples	2016	2017	2018	2019	2020
EU	505.66	504.61	506.27	491.08	484.63
Poland	164.76	162.53	166.15	155.62	152.6
Italy	56.16	57.26	57.44	55	54.91
Romania	55.53	55.6	53.94	52.74	52.34
France	49.65	50.31	50.54	50.37	50.15
Germany	31.74	33.98	33.98	33.98	33.98
Spain	30.87	30.55	29.93	29.64	29.49
Hungary	32.49	32.17	31.84	30.97	25.97
Greece	10.04	9.6	10.35	9.82	14.38
Portugal	14.16	13.85	13.61	14.31	14.31
Lithuania	9.7	9.82	10.13	10.18	10.5
Czechia	7.49	7.35	7.25	7.32	7.19
Austria	6.67	6.67	6.74	6.59	6.43
Netherlands	7.3	7	6.6	6.42	6.2
Belgium	6.49	6.16	5.99	5.79	5.48
Croatia	5.89	4.84	4.73	4.95	4.36
Bulgaria	4.11	3.97	3.98	4.14	3.56
Latvia	2.4	3.3	3.2	3.44	3.5
Latvia	2.4	3.3	3.2	3.44	3.5
Slovenia	2.42	2.36	2.33	2.27	2.16
Slovakia	2.31	2.18	2.14	2.06	1.8
Sweden	1.54	1.4	1.41	1.52	1.44
Denmark	1.35	1.28	1.42	1.39	1.38

Apples	2016	2017	2018	2019	2020
Ireland	0.7	0.7	0.71	0.71	0.71
Finland	0.62	0.63	0.63	0.65	0.67
Estonia	0.51	0.48	0.6	0.57	0.62
Cyprus	0.53	0.37	0.37	0.37	0.41
Luxembourg	0.26	0.27	0.27	0.27	0.08

Table 6: Harvested area of pears (code: F1120) in the EU, 2016–2020 (1,000 ha). Source EUROSTAT (accessed 21 March 2023)

Pears	2016	2017	2018	2019	2020
EU	115.13	113.81	113.54	110.66	107.76
Italy	32.29	31.73	31.34	28.71	26.6
Spain	22.55	21.89	21.33	20.62	20.22
Portugal	11.99	11.54	11.21	11.33	11.33
Belgium	9.69	10.02	10.15	10.37	10.66
Netherlands	9.4	9.7	10	10.09	10
Poland	7.49	7.26	7.3	7.22	5.8
Greece	4.08	4.07	4.41	4.34	5.42
France	5.3	5.25	5.24	5.25	5.38
Romania	3.15	3.12	3.1	3.08	3.09
Hungary	2.87	2.9	2.84	2.81	2.62
Germany	1.93	2.14	2.14	2.14	2.14
Lithuania	0.8	0.82	0.82	0.82	0.85
Czechia	0.74	0.71	0.75	0.8	0.83
Croatia	0.93	0.71	0.8	0.86	0.73
Austria	0.46	0.46	0.49	0.5	0.54
Bulgaria	0.41	0.45	0.57	0.7	0.5
Denmark	0.3	0.3	0.29	0.3	0.3
Slovenia	0.2	0.2	0.21	0.21	0.22
Latvia	0.2	0.2	0.2	0.2	0.2
Sweden	0.12	0.12	0.11	0.1	0.11
Slovakia	0.11	0.11	0.12	0.11	0.1
Cyprus	0.07	0.07	0.06	0.06	0.07
Finland	0.04	0.04	0.05	0.04	0.05
Luxembourg	0.02	0.02	0.02	0.02	0.01

3.4.2.2. Climatic conditions affecting establishment

Based on the data available in the literature on the geographic coordinates of the locations from where *D. bulgarica* has been reported, the pathogen is present in non-EU areas with BSh, BSk, Cfa, Cfb, Cfc, Csa, Csb, Dfb and Dfc Köppen–Geiger climate zones. These climate zones also occur in the EU, where susceptible hosts of *D. bulgarica* (apples and pears) are widely grown (Figure 3).

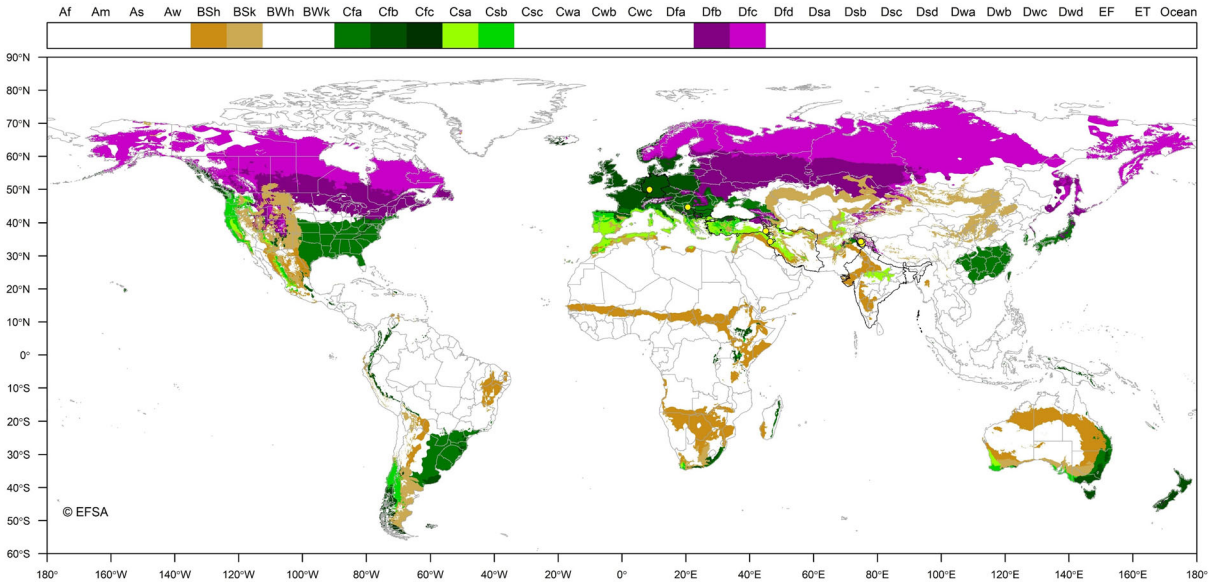

Figure 3: Distribution of nine Köppen–Geiger climate types, i.e. Bsh, Bsk, Cfa, Cfb, Cfc, Csa, Csb, Dfb and Dfc that occur in the EU and in third countries where *Diplodia bulgarica* has been reported. The legend shows the list of Köppen–Geiger climates (Kottek et al., 2006). Yellow dots indicate point locations where *D. bulgarica* was reported

3.4.3. Spread

Describe how the pest would be able to spread within the EU territory following establishment?

Diplodia bulgarica could potentially spread further within the EU by both natural and human-assisted means.

Comment on plants for planting as a mechanism of spread.

Host plants for planting is a main means of spread of the pathogen within the EU.

Diplodia bulgarica could potentially spread further within the EU via natural and human-assisted means.

Spread by natural means. Conidia of the pathogen, like those of other species of the genus *Diplodia* or other members of the family Botryosphaeriaceae, can spread over relatively short distances by water splash (rain, overhead irrigation) (Fullerton et al., 2018). Although it has not been studied in the case of *D. bulgarica*, wind may increase the dispersal distance of water-splashed conidia. Similarly to other *Diplodia* species, conidia of *D. bulgarica* could potentially be passively dispersed between hosts by arthropods (Epstein et al., 2008; Panzavolta et al., 2018). Birds, rodents and other small animals could also potentially disperse the pathogen via infected fruits and seeds (Corlett, 2017).

Spread by human-assisted means. The pathogen can spread over long distances via the movement of infected host plants for planting (rootstocks, grafted plants, scions, etc.), including dormant plants, as well as fresh fruits, contaminated soil/plant-growing media and agricultural machinery, tools, etc.

Although there is no evidence of *D. bulgarica* being seedborne, the pathogen could be potentially further spread within the EU via the seeds of its host plants, similarly to other *Diplodia* species or other members of the family Botryosphaeriaceae (Gure et al., 2005).

3.5. Impacts

Would the pests' introduction have an economic or environmental impact on the EU territory?

Yes, the further introduction into and/or spread within the EU of *Diplodia bulgarica* is expected to have yield and quality impacts in parts of the EU territory where hosts (apples and pears) are grown.

Although no quantitative data are available, in the non-EU areas of its present distribution, *D. bulgarica* has been reported to cause cankers, gummosis, pre- and post-harvest fruit rot, dieback and decline in apple and pear trees. Pathogenicity tests have indicated that *D. bulgarica* is one of the most aggressive *Diplodia* species affecting apple trees (Abdollahzadeh, 2015; Nabi et al., 2020). Based on Abdollahzadeh (2015) study, 2-year-old apple trees (cv. Golden Delicious) died 6 and 8 weeks following their artificial inoculation with the pathogen under greenhouse and field conditions, respectively. In Iran, *D. bulgarica* is the main pathogen of commercial apple orchards in Kermanshah and West Azerbaijan provinces and a serious threat to the apple industry (Abdollahzadeh, 2015). Canker disease caused by members of the Botryosphaeriaceae family, including *D. bulgarica,* is one of the main factors for Iran's ranking drop from the 5th in 2014 to the 7th in 2016 place among apple-producing countries worldwide (Nourian et al., 2021). *Diplodia bulgarica* is reported as the predominant pathogen causing cankers and dieback on apple trees in Jammu and Kashmir in India (Nabi et al., 2020).

In Germany, the pathogen was reported to have a direct impact on organically grown apple and pear trees, some of which were girdled by the cankers and died (Hinrichs-Berger et al., 2021).

Based on the above, it is expected that further introduction into and/or spread within the EU of *D. bulgarica* would potentially cause yield and quality losses in parts of the territory where apples and pears are grown. However, there is uncertainty on the magnitude of this impact as it depends on the susceptibility level of apple and pear cultivars grown in the EU and the physiological condition of the trees (the disease is more severe in trees suffering from environmental stresses). Moreover, it is not known whether the agricultural practices and chemical control measures currently applied in the EU could potentially reduce this impact.

3.6. Available measures and their limitations

Are there measures available to prevent pest entry, establishment, spread or impacts such that the risk becomes mitigated?

Yes. Although not specifically targeted against *Diplodia bulgarica*, existing phytosanitary measures (see Sections 3.3.2 and 3.4.1) mitigate the likelihood of the pathogen's further entry into the EU territory on host plants. Potential additional measures are also available to further mitigate the risk of further entry, establishment, spread and impacts of the pathogen in the EU (see Section 3.6.1).

3.6.1. Identification of potential additional measures

Phytosanitary measures (prohibitions) are currently applied to some host plants for planting (see Section 3.3.2).

Additional potential risk reduction options and supporting measures are shown in Sections 3.6.1.1 and 3.6.1.2.

3.6.1.1. Additional potential risk reduction options

Potential additional control measures are listed in Table 7.

Table 7: Selected control measures (a full list is available in EFSA PLH Panel, 2018) for pest entry/establishment/spread/impact in relation to currently unregulated hosts and pathways. Control measures are measures that have a direct effect on pest abundance

Control measure/ risk reduction option (Blue underline = Zenodo doc, Blue = WIP)	RRO summary	Risk element targeted (entry/establishment/ spread/impact)
Require pest freedom	Plants, plant products and other objects come from a pest-free country or a pest-free area or a pest-free place of production	Entry/spread

Control measure/ risk reduction option (Blue underline = Zenodo doc, Blue = WIP)	RRO summary	Risk element targeted (entry/establishment/ spread/impact)
Growing plants in isolation	Description of possible exclusion conditions that could be implemented to isolate the crop from pests and if applicable relevant vectors. E.g. a dedicated structure such as glass or plastic greenhouses. Growing nursery plants in isolation may represent an effective control measure.	Entry/establishment/spread
Managed growing conditions	Proper field drainage, plant distancing, use of pathogen-free agricultural tools (e.g. pruning scissors, saws and grafting blades), and removal of infected plants and plant debris in the field could potentially mitigate the likelihood of infection at origin as well as the spread of the pathogen.	Entry/spread/impact
Crop rotation, associations and density, weed/ volunteer control	Crop rotation, associations and density, weed/ volunteer control are used to prevent problems related to pests and are usually applied in various combinations to make the habitat less favourable for pests. The measures deal with (1) allocation of crops to field (over time and space) (multi-crop, diversity cropping) and (2) to control weeds and volunteers as hosts of pests/vectors. Although weeds have not been reported as hosts for *D. bulgarica*, their control could potentially make the microclimatic conditions less favourable (e.g. by reducing moisture) to pathogen infection and spread.	Establishment/spread/impact
Use of resistant and tolerant plant species/ varieties	Resistant plants are used to restrict the growth and development of a specified pest and/or the damage they cause when compared to susceptible plant varieties under similar environmental conditions and pest pressure. It is important to distinguish resistant from tolerant species/varieties. Although no information was found on resistant or tolerant varieties, this could be a perspective for the future management of the disease.	Entry/establishment/impact
Roguing and pruning	Roguing is defined as the removal of infested plants and/or uninfested host plants in a delimited area, whereas pruning is defined as the removal of infested plant parts only without affecting the viability of the plant. *Diplodia bulgarica* overwinters on infected attached plant organs which can act as inoculum sources. Thus, pruning of the symptomatic plant organs or removal of severely infected or dead trees may be important in reducing the sources of inoculum and spread capacity	Spread/impact

Control measure/ risk reduction option (Blue underline = Zenodo doc, Blue = WIP)	RRO summary	Risk element targeted (entry/establishment/ spread/impact)
Biological control and behavioural manipulation	Pest control such as: a) Biological control b) Sterile Insect Technique (SIT) c) Mating disruption d) Mass trapping Some microbial antagonists (e.g. *Trichoderma harzianum, T. longibrachiatum*) have shown *in vitro* promising results against *D. bulgarica* (Alijani et al., 2016), but none of them has been tested under field conditions.	Entry/establishment/spread/ impact
Chemical treatments on crops including reproductive material	There is no information on the effectiveness of available fungicides against *D. bulgarica*, since the pathogen was only recently identified. However, the fact that the disease is more prevalent in organically grown apples and pears compared to conventional and integrated cultivation indicates that fungicides commonly used in apple and pear orchards might be effective against *D. bulgarica* too.	Entry/establishment/spread/ impact
Chemical treatments on consignments or during processing	Use of chemical compounds that may be applied to plants or to plant products after harvest, during process or packaging operations and storage. The treatments addressed in this information sheet are: a) fumigation; b) spraying/dipping pesticides; c) surface disinfectants; d) process additives; e) protective compounds The application of fungicides to plants or plant products after harvest, during process or packaging operations and storage may contribute to mitigate the likelihood of further entry or spread of *D. bulgarica,* but no specific information is available on the efficacy of such applications against *D. bulgarica* on apple and pear plants or fruits.	Entry/spread
Physical treatments on consignments or during processing	This information sheet deals with the following categories of physical treatments: irradiation/ ionisation; mechanical cleaning (brushing, washing); sorting and grading; and removal of plant parts (e.g. debarking wood). This information sheet does not address: heat and cold treatment (information sheet 1.14); roguing and pruning (information sheet 1.12). Physical treatments (irradiation, mechanical cleaning, sorting, etc.) may reduce or mitigate the risk of entry and spread, but no specific information for *D. bulgarica* is available.	Entry/spread
Cleaning and disinfection of facilities, tools and machinery	The physical and chemical cleaning and disinfection of facilities, tools, machinery, transport means, facilities and other accessories (e.g. boxes, pots, pallets, palox, supports, hand tools). The measures addressed in this information sheet are washing, sweeping and fumigation.	Entry/spread

Control measure/ risk reduction option (Blue underline = Zenodo doc, Blue = WIP)	RRO summary	Risk element targeted (entry/establishment/ spread/impact)
	Diplodia bulgarica commonly enters its host plants through wounds created by pruning or grafting. Therefore, cleaning and surface sterilisation of pruning and grafting tools as well as of equipment and facilities (including premises, storage areas) are good cultural and handling practices employed in the production and marketing of any commodity and may mitigate the likelihood of further entry or spread of the pathogen.	
Limits on soil	*Diplodia bulgarica* survives in infected plant debris in soil. Therefore, plants, plant products and other objects (e.g. used farm machinery) should be free from soil to ensure freedom from the pathogen	Entry/spread
Soil treatment	The control of soil organisms by chemical and physical methods listed below: (a) Fumigation; (b) heating; (c) solarisation; (d) flooding; (e) soil suppression; (f) augmentative biological control; (g) biofumigation Given that *D. bulgarica* survives in infected plant debris in soil and despite the lack of specific studies for this pathogen, it is likely that soil and substrate disinfestation with chemical, biological or physical (heat, soil solarisation) means could potentially reduce the persistence and availability of inoculum sources.	Entry/establishment/spread/ impact
Use of non-contaminated water	Chemical and physical treatment of water to eliminate waterborne microorganisms. The measures addressed in this information sheet are chemical treatments (e.g. chlorine, chlorine dioxide, ozone); physical treatments (e.g. membrane filters, ultraviolet radiation, heat); ecological treatments (e.g. slow sand filtration). Although *D. bulgarica* could potentially spread via contaminated irrigation water, physical or chemical treatment of irrigation water is likely not to be feasible under field conditions but may be applied in nurseries and greenhouses.	Entry/ Spread/Impact
Waste management	Waste management (incineration, production of bioenergy) that takes place in authorised facilities and official restriction on the movement of infected plant material is in force to prevent the pest from escaping. On-site proper management of pruning residues is recommended as an efficient measure	Entry/establishment/spread
Heat and cold treatments	Controlled temperature treatments aimed to kill or inactivate pests without causing any unacceptable prejudice to the treated material itself. The measures addressed in this information sheet are: autoclaving; steam; hot water; hot air; cold treatment Although no specific studies are available for *D. bulgarica,* cold storage may delay fruit rot caused by *D. bulgarica*.	Entry/spread

Control measure/ risk reduction option (Blue underline = Zenodo doc, Blue = WIP)	RRO summary	Risk element targeted (entry/establishment/ spread/impact)
Conditions of transport	Specific requirements for mode and timing of transport of commodities to prevent escape of the pest and/or contamination. a) physical protection of consignment b) timing of transport/trade If plant material, potentially infected or contaminated with *D. bulgarica* (including waste material) must be transported, specific transport conditions (type of packaging/protection, transport means) should be defined to prevent the pathogen from escaping. These may include, albeit not exclusively: physical protection, sorting prior to transport, sealed packaging, etc.	Entry/spread
Controlled atmosphere	Storing fruit in a modified atmosphere (including modified humidity, O_2, CO_2, temperature, pressure) could delay post-harvest rot, but no specific information for *D. bulgarica* is available.	Entry/spread (via commodity)
Post-entry quarantine and other restrictions of movement in the importing country	This information sheet covers post-entry quarantine (PEQ) of relevant commodities; temporal, spatial and end-use restrictions in the importing country for import of relevant commodities; Prohibition of import of relevant commodities into the domestic country. 'Relevant commodities' are plants, plant parts and other materials that may carry pests, either as infection, infestation or contamination. Recommended for plant species known to be host of *D. bulgarica*. Nevertheless, this measure does not apply to fruits of host plants.	Establishment/spread

3.6.1.2. Additional supporting measures

Potential additional supporting measures are listed in Table 8.

Table 8: Selected supporting measures (a full list is available in EFSA PLH Panel, 2018) in relation to currently unregulated hosts and pathways. Supporting measures are organisational measures or procedures supporting the choice of appropriate risk reduction options that do not directly affect pest abundance

Supporting measure (Blue underline = Zenodo doc, Blue = WIP)	Summary	Risk element targeted (entry/establishment/ spread/impact)
Inspection and trapping	Inspection is defined as the official visual examination of plants, plant products or other regulated articles to determine if pests are present or to determine compliance with phytosanitary regulations (ISPM 5). The effectiveness of sampling and subsequent inspection to detect pests may be enhanced by including trapping and luring techniques.	Entry/establishment/spread

Supporting measure (Blue underline = Zenodo doc, Blue = WIP)	Summary	Risk element targeted (entry/establishment/ spread/impact)
	The symptoms caused by *D. bulgarica* on host plants are similar to those caused by other *Diplodia* species or other members of the family Botryosphaeriaceae or by other biotic agents or abiotic agents. Moreover, the pathogen may remain quiescent or latent within the asymptomatic host tissues. Therefore, it is unlikely that the pathogen could be detected based on visual inspection only.	
Laboratory testing	Examination, other than visual, to determine if pests are present using official diagnostic protocols. Diagnostic protocols describe the minimum requirements for reliable diagnosis of regulated pests. Multilocus gene sequencing analysis combined with cultural and morphological characteristics of fungal colonies and pycnidia with conidia is required for the reliable detection and identification of *D. bulgarica* (see Section 3.1.5)	Entry/establishment/spread
Sampling	According to ISPM 31, it is usually not feasible to inspect entire consignments, so phytosanitary inspection is performed mainly on samples obtained from a consignment. It is noted that the sampling concepts presented in this standard may also apply to other phytosanitary procedures, notably selection of units for testing. For inspection, testing and/or surveillance purposes, the sample may be taken according to a statistically based or a non-statistical sampling methodology. Necessary as part of other risk reduction options.	Entry/establishment/spread
Phytosanitary certificate and plant passport	An official paper document or its official electronic equivalent, consistent with the model certificates of the IPPC, attesting that a consignment meets phytosanitary import requirements (ISPM 5) a) export certificate (import) b) plant passport (EU internal trade) Recommended for plant species known to be hosts of *D. bulgarica*.	Entry/spread
Certified and approved premises	Mandatory/voluntary certification/approval of premises is a process including a set of procedures and of actions implemented by producers, conditioners and traders contributing to ensure the phytosanitary compliance of consignments. It can be a part of a larger system maintained by the NPPO in order to guarantee the fulfilment of plant health requirements of plants and plant products intended for trade. Key property of certified or approved premises is the traceability of activities and tasks (and their components) inherent the pursued phytosanitary objective. Traceability aims to provide access to all trustful pieces of information that may help to prove the compliance of consignments with phytosanitary requirements of importing countries. Certified and approved premises may reduce the likelihood of the plants and plant products originating in those premises to be infected by *D. bulgarica*.	Entry/Spread

Supporting measure (Blue underline = Zenodo doc, Blue = WIP)	Summary	Risk element targeted (entry/establishment/ spread/impact)
Certification of reproductive material (voluntary/official)	Plants come from within an approved propagation scheme and are certified pest free (level of infestation) following testing; Used to mitigate against pests that are included in a certification scheme. The risk of entry and/or spread of *D. bulgarica* is reduced if host plants for planting are produced under an approved certification scheme and tested free of the pathogen.	Entry/spread
Delimitation of Buffer zones	ISPM 5 defines a buffer zone as 'an area surrounding or adjacent to an area officially delimited for phytosanitary purposes in order to minimise the probability of spread of the target pest into or out of the delimited area, and subject to phytosanitary or other control measures, if appropriate' (ISPM 5). The objectives for delimiting a buffer zone can be to prevent spread from the outbreak area and to maintain a pest-free production place (PFPP), site (PFPS) or area (PFA). Delimitation of a buffer zone around an outbreak area can prevent spread of the pathogen and maintain a pest-free area, site or place of production.	Spread
Surveillance	Surveillance to guarantee that plants and products originate from a pest-free area could be an option. *Diplodia bulgarica* has been reported to be present in the EU. Therefore, surveillance would be an efficient supporting measure to define pest-free areas or pest-free places of production as well as to prevent further spread of the pathogen.	Spread

3.6.1.3. Biological or technical factors limiting the effectiveness of measures

- Latently infected (asymptomatic) host plants and plant products are unlikely to be detected by visual inspection.
- The similarity of disease symptoms and signs (e.g. pycnidia with conidia) of *D. bulgarica* with those of other *Diplodia* species or other fungi of the family Botryosphaeriaceae makes impossible the detection and identification of the pathogen based only on visual inspection. Moreover, symptoms caused by the pathogen may be similar to those caused by abiotic factors.
- The lack of rapid diagnostic methods based on molecular approaches does not allow proper *in planta* identification of the pathogen at entry. In addition, thorough post-entry laboratory analyses may not be feasible for certain commodities as isolation in pure culture is needed prior to DNA extraction as well as molecular identification based on multigene sequencing.

3.7. Uncertainty

There is a key uncertainty with respect to the geographical distribution of *D. bulgarica* (worldwide and in the EU), because in the past, when molecular tools were not available, the pathogen might have been misidentified as other *Diplodia* species (e.g. *D. intermedia, D. malorum, D. mutila, D. seriata*) or other members of the Botryosphaeriaceae family affecting apples and pears based only on morphology and pathogenicity tests.

4. Conclusions

Diplodia bulgarica has been reported from a limited part of the EU (Bulgaria, Germany). Therefore, *D. bulgarica* satisfies the criteria that are within the remit of EFSA to assess for this species to be regarded as potential Union quarantine pest (Table 9).

Table 9: The Panel's conclusions on the pest categorisation criteria defined in Regulation (EU) 2016/2031 on protective measures against pests of plants (the number of the relevant sections of the pest categorisation is shown in brackets in the first column)

Criterion of pest categorisation	Panel's conclusions against criterion in Regulation (EU) 2016/2031 regarding Union quarantine pest	Key uncertainties
Identity of the pest (Section 3.1)	The identity of *D. bulgarica* is clearly defined. The pathogen has been shown to produce consistent symptoms and to be transmissible.	None
Absence/presence of the pest in the EU (Section 3.2)	*Diplodia bulgarica* has been reported from Bulgaria and Germany.	The geographical distribution of *D. bulgarica* in the EU, particularly where other *Diplodia* species or other members of the family Botryosphaeriaceae were identified on apples and pears based only on morphology and pathogenicity tests.
Pest potential for entry, establishment and spread in the EU (Section 3.4)	*Diplodia bulgarica* has already entered the EU and it may be further introduced and spread within the EU territory. The main pathways for the further entry of the pathogen into and spread within the EU are: (i) host plants for planting, including seeds, (ii) fresh fruits of host plants, (iii) bark and wood of host plants and (iv) soil and other plant-growing media, originating in infested third countries. The pathogen has established in Germany and is present in Bulgaria, which indicates that both the biotic (host availability) and abiotic (climate suitability) factors occurring in parts of the EU are favourable for its establishment. *Diplodia bulgarica* could potentially further spread within the EU by both natural and human-assisted means.	None
Potential for consequences in the EU (Section 3.5)	The further introduction and spread of *D. bulgarica* in the EU are expected to have yield and quality impacts in parts of the territory where susceptible hosts are grown.	None
Available measures (Section 3.6)	Although not specifically targeted against *D. bulgarica*, existing phytosanitary measures mitigate the likelihood of the pathogen's further introduction and spread in the EU. Potential additional measures also exist to further mitigate the risk of further introduction and spread of the pathogen in the EU.	None
Conclusion (Section 4)	*Diplodia bulgarica* satisfies all the criteria that are within the remit of EFSA to assess for this species to be regarded as potential Union quarantine pest.	None
Aspects of assessment to focus on scenarios to address in future if appropriate:	The main knowledge gap concerns the present distribution of *D. bulgarica* worldwide and in the EU. To reduce this uncertainty, systematic surveys would need to be carried out and isolates of *Diplodia* species originated from apples and pears and identified based on morphology and pathogenicity tests would need to be re-evaluated using multilocus gene sequencing analysis.	

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Abbreviations

EPPO	European and Mediterranean Plant Protection Organisation
FAO	Food and Agriculture Organisation

IPPC	International Plant Protection Convention
ISPM	International Standards for Phytosanitary Measures
MS	Member State
PLH	EFSA Panel on Plant Health
PZ	Protected Zone
TFEU	Treaty on the Functioning of the European Union
ToR	Terms of Reference

Glossary

Containment (of a pest)	Application of phytosanitary measures in and around an infested area to prevent spread of a pest (FAO, 2022)
Control (of a pest)	Suppression, containment or eradication of a pest population (FAO, 2022)
Entry (of a pest)	Movement of a pest into an area where it is not yet present, or present but not widely distributed and being officially controlled (FAO, 2022)
Eradication (of a pest)	Application of phytosanitary measures to eliminate a pest from an area (FAO, 2022)
Establishment (of a pest)	Perpetuation, for the foreseeable future, of a pest within an area after entry (FAO, 2022)
Greenhouse	A walk-in, static, closed place of crop production with a usually translucent outer shell, which allows controlled exchange of material and energy with the surroundings and prevents release of plant protection products (PPPs) into the environment.
Hitchhiker	An organism sheltering or transported accidentally via inanimate pathways including with machinery, shipping containers and vehicles; such organisms are also known as contaminating pests or stowaways (Toy and Newfield, 2010).
Impact (of a pest)	The impact of the pest on the crop output and quality and on the environment in the occupied spatial units
Introduction (of a pest)	The entry of a pest resulting in its establishment (FAO, 2022)
Pathway	Any means that allows the entry or spread of a pest (FAO, 2022)
Phytosanitary measures	Any legislation, regulation or official procedure having the purpose to prevent the introduction or spread of quarantine pests, or to limit the economic impact of regulated non-quarantine pests (FAO, 2022)
Quarantine pest	A pest of potential economic importance to the area endangered thereby and not yet present there, or present but not widely distributed and being officially controlled (FAO, 2022)
Risk reduction option (RRO)	A measure acting on pest introduction and/or pest spread and/or the magnitude of the biological impact of the pest should the pest be present. A RRO may become a phytosanitary measure, action or procedure according to the decision of the risk manager
Spread (of a pest)	Expansion of the geographical distribution of a pest within an area (FAO, 2022)

Appendix A – *Diplodia bulgarica* host plants/species affected

Source: CABI Compendium (CABI, 2022) and other literature sources.

Host status	Host name	Plant family	Common name	Reference[A]
Cultivated hosts	*Malus domestica*	Rosaceae	Apple	Phillips et al. (2012), Abdollahzadeh (2015), Hanifeh et al. (2017), Nabi et al. (2020), Eken (2021), Hinrichs-Berger et al. (2021), Zeynali Bari et al. (2021)
	Malus sylvestris	Rosaceae	Wild apple	Phillips et al. (2012, 2013)
	Pyrus communis	Rosaceae	European pear	Hinrichs-Berger et al. (2021)
Artificial/ experimental hosts*	*Corylus avellana*	Betulaceae	Hazelnut	Ketabchi et al. (2015)
	Pistacia vera	Anacardiaceae	Pistachio	Ketabchi et al. (2015)
	Prunus domestica	Rosaceae	Plum	Ketabchi et al. (2015)
	Salix spp.	Salicaceae	Willow	Ketabchi et al. (2015)

*: With high uncertainty (see Section 3.1.3 Host range/Species affected).

Appendix B – Distribution of *Diplodia bulgarica*

Distribution records based on CABI Compendium (CABI, 2022) and other literature sources.

Region	Country	Subnational (e.g. State)	Status	References
EU	Bulgaria	Plovdiv	Present, no details	Phillips et al. (2012), pers. comm. from NPPO (2023)
	Germany		Present, widespread	Hinrichs-Berger et al. (2021), pers. comm. from NPPO (2023)
Other Europe	Serbia	Radmilovac	Present	Vučković et al. (2022), CABI (2022)
Asia	India	Anantnag	Present	Nabi et al. (2020), CABI (2022)
		Bandipora		
		Baramulla		
		Gaenderbal		
		Kulgam		
		Pulwama		
		Sophian		
	Iran	Kermanshah West Azerbaijan	Present	Phillips et al. (2012), Abdollahzadeh (2015), Nourian et al. (2021)
	Türkiye	Isparta Province	Present	Eken (2022), CABI (2022)

Appendix C – EU annual imports of fresh produce of hosts from countries where *Diplodia bulgarica* is present, 2016–2020 (in 100 kg)

Source: Eurostat accessed on 21 March 2023

		2016	2017	2018	2019	2020
Fresh apples	India	0.01	:	:	:	0.45
	Türkiye	240.22	1,610.74	17,594.86	2,311.21	19,023.31
	Iran	:	:	2,945.28	0.38	676.65
	Serbia	259,419.33	227,815.83	104,854.11	340,445.66	214,121.46
	Sum	**259,659.56**	**229,426.57**	**125,394.25**	**342,757.25**	**233,821.87**

		2016	2017	2018	2019	2020
Fresh pears	Türkiye	13,874.34	32,003.71	67,690.28	63,998.83	113,683.44
	Iran	:	:	32.40	:	7.50
	Serbia	6,652.62	14,300.35	13,774.01	12,971.90	19,523.95
	Sum	**20,526.96**	**46,304.06**	**81,496.69**	**76,970.73**	**133,214.89**