

EPPO Datasheet: *Gremmeniella abietina*

Last updated: 2023-06-09

IDENTITY

Preferred name: *Gremmeniella abietina*

Authority: (Lagerberg) Morelet

Taxonomic position: Fungi: Ascomycota: Pezizomycotina:
Leotiomycetes: Helotiales: Helotiaceae

Other scientific names: *Ascocalyx abietina* (Lagerberg) Schläpfer-Bernhard, *Brunchorstia destruens* Eriksson, *Brunchorstia pinea* var. *cembrae* Morelet, *Brunchorstia pinea* var. *pini* Morelet, *Brunchorstia pinea* (Karsten) von Höhnel, *Brunchorstia pini* Allescher, *Crumenula abietina* Lagerberg, *Crumenula pinea* (Karsten) Ferdinandsen & C.A.Jørgensen, *Excipulina pinea* Karsten, *Godronia abietina* (Ellis & Everhart) Seaver, *Lagerbergia abietina* (Lagerberg) J.Reid, *Scleroderris abietina* (Lagerberg) Gremmen, *Scleroderris lagerbergii* Gremmen, *Septoria pinea* Karsten

Common names: brunchorstia disease of pine, canker of conifers, dieback of pine, scleroderris canker of conifers (US), shoot blight of pine

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EU Categorization: PZ Quarantine pest (Annex III)

EPPO Code: GREMAB



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Notes on taxonomy and nomenclature

Within the genus *Gremmeniella*, three species have been recognized: *G. abietina* (Lagerberg) Morelet, *G. laricina* (Ettinger) O. Petrini, L.E. Petrini, G. Laflamme, & G.B. Ouellette, and *G. juniperina* L. Holm & K. Holm (Petrini *et al.*, 1989). The latter two species appear to be host-specific on the *Larix* and *Juniperus* genera, respectively (Petrini *et al.*, 1989). *G. abietina* is a virulent haploid ascomycete and complex species, since several taxonomic entities (varieties, races, and biotypes) have been recognized within this species, including two varieties, *G. abietina* var. *abietina* that mainly infects pines, and *G. abietina* var. *balsamea* that infects firs and spruces (Botella & Hantula, 2018). Moreover, three races of *G. abietina* from different continents were described, i.e. Asian, North American, and European, and it has even been suggested to consider them separate species (Hamelin & Rail, 1997; Uotila *et al.*, 2000; Botella & Hantula, 2018). The up-to-date taxonomy of *G. abietina* includes three biotypes that have been identified in the European race based on morphological traits (spore length, septa numbers), symptoms, and molecular markers (Hellgren & Högberg, 1995; Hamelin *et al.*, 1996; Hantula & Muller, 1997). The Alpine biotype has been described in the Alpine region. It can grow in harsh environments of snow and extreme temperatures and produce abundant pycnidia and apothecia in the field (Hellgren & Högberg 1995). The biotype A (European, large tree type, LTT) is the most pathogenic biotype (Botella *et al.*, 2015). This biotype is widespread from the Italian Apennines to Northern Sweden, and it was introduced in North America in the 1970s (Botella *et al.*, 2015; Botella & Hantula, 2018). The biotype B (Scandinavian, small tree type, STT) was found first in Northern European countries, but it also seems to be present in high mountainous areas of the Lake District of Türkiye (Botella *et al.*, 2015).

HOSTS

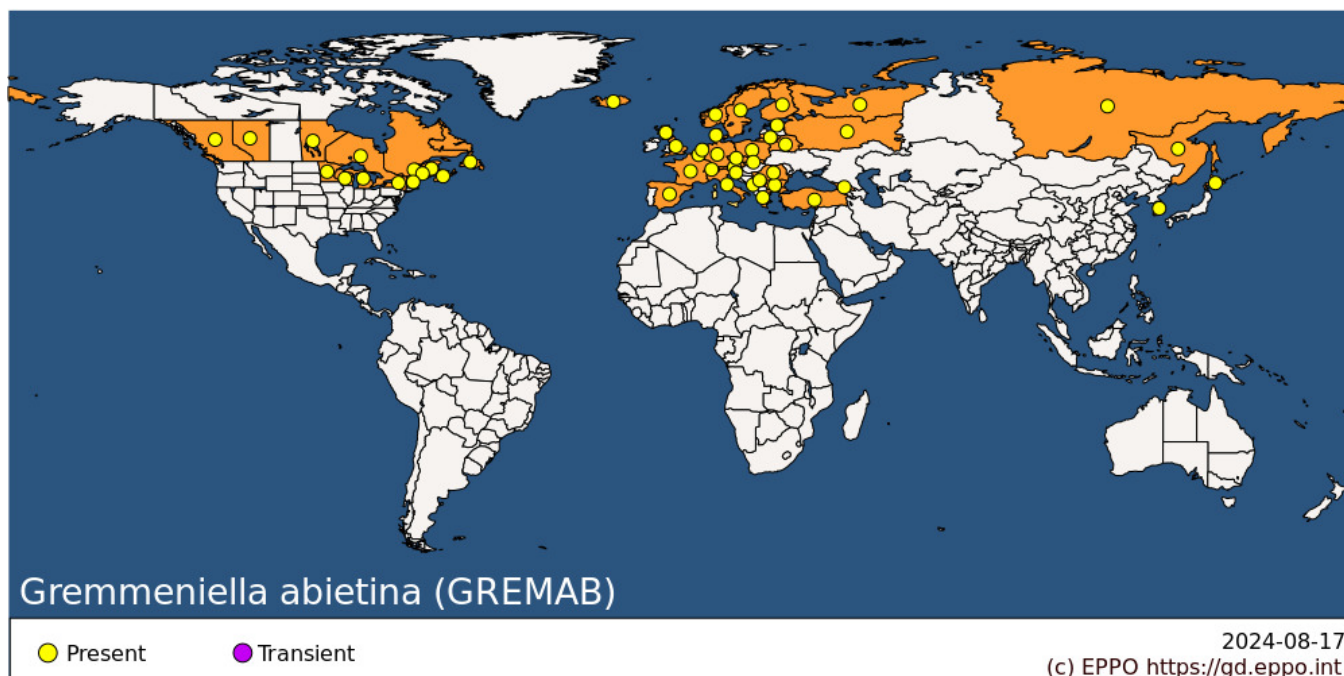
The host range of *G. abietina* is mostly confined to *Pinus* species and other conifers such as *Abies* spp., *Picea* spp., *Larix* spp., and *Pseudotsuga* spp., of which two major European host species (*Pinus sylvestris* and *Picea abies*) occur widely in the EPPO region. *Abies sachalinensis* and *P. contorta* are also considered important hosts (Jeger *et al.*, 2017). The North American race occurs in the USA and Canada mainly on *Pinus resinosa*, *Pinus banksiana*, and *Pinus contorta*, where it causes damage on low parts of trees that remain under snow cover during the winter (Petrini *et al.*, 1989). The European race generally infects the entire crown of hosts, and it is found on several different hosts

belonging mainly to *Pinus* spp. in Europe and North America. The Asian race has been found only in Japan on *A. sachalinensis* (Jeger *et al.*, 2017). The two- and three-needled pines are most susceptible; five-needled pines and spruces are much less susceptible or tolerant while other conifers seem to be resistant (Sinclair & Lyon, 2005). For additional information, see Sinclair & Lyon (2005).

Host list: *Abies balsamea*, *Abies nordmanniana* subsp. *equitrojani*, *Abies sachalinensis*, *Larix lyallii*, *Picea abies*, *Picea glauca*, *Picea jezoensis*, *Picea mariana*, *Picea omorika*, *Picea rubens*, *Picea sitchensis*, *Pinus banksiana*, *Pinus cembra*, *Pinus contorta*, *Pinus densiflora*, *Pinus halepensis*, *Pinus koraiensis*, *Pinus monticola*, *Pinus mugo*, *Pinus nigra* subsp. *laricio*, *Pinus nigra* subsp. *pallasiana*, *Pinus nigra*, *Pinus pinaster*, *Pinus pinea*, *Pinus ponderosa*, *Pinus radiata*, *Pinus resinosa*, *Pinus rigida*, *Pinus sabiniana*, *Pinus strobus*, *Pinus sylvestris*, *Pinus wallichiana*, *Pseudotsuga menziesii*

GEOGRAPHICAL DISTRIBUTION

G. abietina is a destructive pathogen of conifers in North, Central, and South Europe, north-eastern North America, and East Asia (Botella & Hantula, 2018). *G. abietina* causes a disease known as Scleroderris canker in North America and Brunchorstia dieback in Europe (Yokota *et al.*, 1974; Skilling *et al.*, 1984; Petrini *et al.* 1989; Hudler & Neal, 1990; Sinclair & Lyon, 2005, Thompsen, 2009; Botella & Hantula, 2018).



EPPO Region: Austria, Belarus, Belgium, Bulgaria, Czech Republic, Denmark, Estonia, Finland, France (mainland), Georgia, Germany, Greece (mainland), Italy (mainland), Lithuania, Montenegro, Netherlands, Norway, Poland, Romania, Russia (Central Russia, Eastern Siberia, Far East, Northern Russia), Serbia, Slovakia, Slovenia, Spain (mainland), Sweden, Switzerland, Türkiye, United Kingdom (England, Scotland)

Asia: Japan (Hokkaido), Korea, Republic

North America: Canada (Alberta, British Columbia, Manitoba, New Brunswick, Newfoundland, Nova Scotia, Ontario, Québec), United States of America (Maine, Michigan, Minnesota, New Hampshire, New York, Wisconsin)

BIOLOGY

The ascomycete fungus *G. abietina* is a pathogen which causes cankers on stems and trunks, dieback and death of the trees. The germinating conidia or ascospores enter the apical buds and developing shoots, especially during cool, wet springs (Venier *et al.*, 1998; Witzell & Karlman, 2000; Thompsen, 2009). The initial attack of *G. abietina* typically starts in the lower part of the crown and spreads to branches in the upper part of the crown, and then towards the stem, causing perennial cankers (Hellgren & Barklund, 1992; Sinclair & Lyon, 2005). Wounded needles, buds and shoots are particularly susceptible to infection. Conidia and ascospores are both infectious, however

ascospores are relatively less important in disease spread (Sinclair & Lyon, 2005). In Europe, conidia and ascospores are released during periods of wet weather, conidia are dispersed mainly in water, ascospores in air. Pycnidia starts to release conidia in late spring, whereas apothecia begin to release ascospores in summer (Hellgren & Barklund, 1992; Sinclair & Lyon, 2005). Infection occurs primarily in spring and summer, but it may be as late as October (Sinclair & Lyon, 2005). Needle bases turn orange to brown while the tip may be still green, and ultimately fall off (Hellgren & Barklund, 1992; Sinclair & Lyon, 2005). Colonization proceeds both inter- and intracellularly facilitated by hyphal secretion of different enzymes; shoots start dying, in the spring following infection, from the tips (Gremmen, 1972; Sinclair & Lyon, 2005). Small, black pycnidia develop during the first and second years of symptomatic infection and appear at the base of dead needles or on dead shoot tips throughout the year; but more commonly in the spring and early autumn (Gremmen, 1972; Sinclair & Lyon, 2005). Pycnidia release conidia, completing the disease cycle (Sinclair & Lyon, 2005). The fungus overwinters as mycelium or as immature fruiting bodies in the conifer host. *G. abietina* is seasonal in its development in the host plant and sexual reproduction and completion of the whole pathogen cycle takes two years; apothecia occur more commonly in the same place where pycnidia were located one year after the shoots die (Gremmen, 1972; Hellgren and Barklund, 1992; Sinclair & Lyon, 2005). In addition, the ambient temperature at this time influences both the number of ascospores that are discharged per apothecium and how soon dispersal commences. Maximum spore discharge takes place at approximately 17°C. If free water is available in the form of rain for 4–8 h at this temperature, major spore release will take place (Skilling, 1972). Snowy winters also frequently promote damage by *G. abietina* (Witzell and Karlman, 2000). This results in significant loss of foliage, further weakening of the trees due to secondary attack by other fungi and insects, and finally death. *G. abietina* can grow at temperatures of -6°C. This facilitates tree colonization due to the suppression of the immune system of trees during the winter, and disease appears frequently in sites which are very wet for a long time (such as frost pockets and cold air drainages) and where snow lingers in spring (Sinclair & Lyon, 2005).

DETECTION AND IDENTIFICATION

Symptoms

Generally, *G. abietina* initially infects and kills buds and the current-year shoots. The pathogen can then remain latent for several years or continue to grow along the branch towards the main stem (Skilling *et al.*, 1986; Bernhold, 2008). The North American and European races cause similar symptoms on susceptible hosts with abundant canker formation and production of both pycnidia and apothecia on smaller trees, particularly on those covered with snow in the winter (Uotila, 1985; Barklund, 1990; Hamelin *et al.*, 1996; Bernhold, 2008). Initial infection by *G. abietina* occurs in developing shoots in the spring and resinous lesions form primarily during the host's period of dormancy in cortical tissue of the first- or second-year twigs, then they extend into needle bases and buds (Gremmen, 1972; Sinclair & Lyon, 2005). However, the first symptoms may not appear until the following winter when resin exudation can be observed on the buds (Gremmen, 1972). Typically, needle bases turn orange-brown by early spring and the discoloration gradually extending to the needle tips and symptomatic needle clusters can be pulled from twigs readily and needles can drop during late spring and summer (Sinclair & Lyon, 2005). The necrosis formed under the short shoots weakens the stability of the needles (they easily fall out) and cuts off all the transport of water and nutrients into the needles (Roll-Hansen, 1964; Skilling, 1972). The characteristic yellow colouration of the xylem tissues and distorted shoots can also be seen (Read, 1967; Skilling, 1972). Consequently, the previous year's needles are often bent down and red to brown, starting from the base and moving towards the tip (Roll-Hansen, 1964; EPPO, 2009). Buds on twigs with discoloured needles frequently do not open and resin droplets appear on the bark of the current year's shoots (Sinclair & Lyon, 2005; EPPO, 2009). After about one year, the dead shoots lose most or all of their red-brown needles and appear dry (Bernhold, 2008). One year later bunches of light-green needles may develop from adventitious buds, present at the base of the dead shoots and sometimes giving the appearance of small witches' brooms (Roll-Hansen, 1964; Skilling, 1972; EPPO, 2009). *G. abietina* mycelia can survive up to eight years in *P. sylvestris* in symptomless shoots (Hellgren & Barklund, 1992). *G. abietina* can remain in the shoots during unfavourable conditions and rapidly recolonize a stand when conditions become favourable for the pathogen (Bernhold, 2008).

G. abietina can cause larger necroses in the inner tissue of stems and branches resulting in cankers on stems and branches (Skilling, 1972). The cambial region in necrotic lesions caused by the North American race is often become discoloured or turns yellow-green due to a pigment produced by the pathogen (Sinclair & Lyon, 2005). Moreover,

trunk cankers caused by the North American race are elongate, resinous, and usually have a greenish discoloration while the European race causes death of apical buds and adjacent needled on *P. banksiana* and *P. contorta* (tip blight) and stem cankers are quite rare (Sinclair & Lyon, 2005). Stem cankers caused by the biotype LTT of *G. abietina* are rare in Sweden, even though small cankers on branches do occur (Hellgren & Högberg, 1996; Bernhold, 2008). On the other hand, cankers on the main stem caused by the biotype STT of *G. abietina* are frequently found in Northern Sweden, especially on *P. contorta*, and these are formed after infection through the base of diseased branches and through bark damage caused by snow, frost, and ice formation (Bernhold, 2008).

Morphology

Pycnidia of *G. abietina* develop on needles and stems after appearance of symptoms on needles (Sinclair & Lyon, 2005). In this species, they are generally more common than apothecia (EPPO, 2009). Pycnidia form singly or in clusters in the bark of dead twigs or needle bases; they are irregular in shape, multilocular with one or more fertile cavity, dark-brown to black on the surface and white within, stromatic, up to 1 mm wide (Roll-Hansen, 1964; Skilling, 1972; Sinclair & Lyon, 2005; EPPO, 2009). Pycnidia produce conidia in a mucilaginous matrix. Conidiophores completely line the inside of the pycnidial cavity, they are hyaline, cylindrical. Conidia are individually colourless, usually four celled, sometimes five-seven celled, hyaline, cylindrical, somewhat curved, $25\text{--}40 \times 3.0\text{--}3.5 \mu\text{m}$ and have pointed ends (Sutton, 1980; Sinclair & Lyon, 2005). Microconidia are $5 \times 3 \mu\text{m}$; they have been also found in pycnidia of *G. abietina* on many *Pinus* species in Europe and North America (Roll-Hansen, 1964; Skilling, 1972; Sutton, 1980) and their function is still unclear (Bergdahl, 1984; Sinclair & Lyon, 2005).

Brown cuplike apothecia appear in the spring on needles and twigs which died 1–2 years ago (Gremmen, 1972; Bergdahl, 1984; Sinclair & Lyon, 2005). Apothecia are gregarious, erumpent, superficial, about 1 mm in diameter, slightly taller than wide, measure $400\text{--}1200 \times 120\text{--}250 \mu\text{m}$ with short stalks and the fertile inner surface is cream-coloured (Sutton, 1980; Sinclair & Lyon, 2005). Asci subclavate, short-stipitate, inoperculate, eight-spored, $100\text{--}120 \times 6.5\text{--}10.5 \mu\text{m}$ and ascus wall bitunicate. Ascospores are colourless, biseriate, 3-septate, typically 4-guttulate with rounded ends, hyaline, sometimes slightly curved, $15\text{--}22 \times 3\text{--}5 \mu\text{m}$ (Punithalingam & Gibson, 1973; Sutton, 1980; Sinclair & Lyon, 2005).

Detection and inspection methods

G. abietina can be identified based on the species' morphological structures following the EPPO diagnostic protocol PM 7/92 (1): *Gremmeniella abietina*. If mature fruiting structures are present, *G. abietina* can easily be identified *in vivo*. In the absence of visible fruiting structures, the symptomatic samples should be incubated in a moist chamber or used for culturing to induce sporulation and then identification is possible using the classical morphological examination; the presence of pycnidia and the conidia as well as the ascomata and ascospores can distinguish *G. abietina*. Culture media and isolation techniques from needles and shoots are also described in the EPPO diagnostic protocol (EPPO, 2009, 2019).

Pine seedlings in nurseries should be inspected for orange to brown discoloration at the bases of needles in early May. By July, needles and branch tips of infested plants become brown. Needles may fall down from branch tips when the slightest pressure is applied. In young pine trees, green discoloration appears beneath the bark of dead branches. Stem cankers are rare but small branch cankers are commonly found. Throughout the year, but in particular in the spring and the early autumn, black pycnidia or light-brown apothecia are likely to be visible at the base of dead needles or on dead branch tips.

In forests, the earliest visible symptoms of the disease are indicators of the presence of infection in the buds or shoots. The most obvious early symptom is the orange to brown discoloration of the needle bases in the spring prior to shoot elongation.

The three races (Asian, European, and North American) within *G. abietina* have been distinguished by immunological and other methods (Dorworth & Krywienczyk, 1975). Since the European and North American races do not differ morphologically from each other, several molecular approaches have been used to allow their identification (Hogberg, 1995; Hamelin *et al.*, 1996, 2000; Hantula & Muller, 1997; Zeng *et al.*, 2005). Then, three biotypes have been distinguished within the European race based on disease symptoms and conidia morphology (Hamelin *et al.*, 1993; Zeng *et al.*, 2005; Botella *et al.*, 2015; Lamarche *et al.*, 2015). Different varieties, races, and

biotypes of *G. abietina* can be also identified and separated using different DNA methods (Hamelin *et al.*, 1996, 1998, 2000; Hantula & Muller, 1997; Zeng *et al.*, 2005; Botella *et al.*, 2015; Lamarche *et al.*, 2015) and some other methods (Appendix 1 of the EPPO diagnostic protocol: EPPO, 2009, 2019), but such differentiation is not required for quarantine purposes; given the uncertainty about the taxonomic position of the varieties within *G. abietina*, none of these tests are recommended (EPPO, 2009).

The presence of *G. abietina* on shoots, needles and roots can be revealed using *G. abietina* specific primers for conventional PCR (developed by Dr. Elna Stenström, Swedish University of Agricultural Sciences, Sweden) (Adomas & Asiogbu, 2006).

PATHWAYS FOR MOVEMENT

G. abietina can survive in an endophytic stage for an uncertain period of time during unfavourable conditions for the pathogen in symptomless shoots (Petrini *et al.*, 1989; Hellgren & Barklund, 1992). Therefore, it might be moved over long distances in infected asymptomatic plants (Jeger *et al.*, 2017). Under favourable wet conditions, conidia released from the infected tissues are dispersed by a water splash mechanism (Thomsen, 2009). Long-distance dispersal of the fungus is considered to occur largely through wind-borne ascospores (Sinclair & Lyon, 2005). The widespread distribution of the pathogen may lead to infection from conidia or ascospores entering the nursery site of production from surrounding areas (Jeger *et al.*, 2017). Transport of infected nursery stock or movement of infected Christmas trees may provide alternative means of long-distance dispersal (Patton *et al.*, 1984; Santamaría *et al.*, 2007; Jeger *et al.*, 2017).

PEST SIGNIFICANCE

Economic impact

Scleroderris canker caused by *G. abietina* is a major disease of pines in snowy regions of Europe and eastern North America (Sinclair & Lyon, 2005; Bernhold, 2008). It kills seedlings, saplings, and stunts and deforms survivors, and reduces timber quality due to the trunk cankers (Sinclair & Lyon, 2005). The European race of *G. abietina* is more aggressive than the North American race, in which can be seen on most of their common hosts, and the European race kills shoots and causes cankers and dieback on trees of all sizes (Sinclair & Lyon, 2005). Large scale outbreaks may occur in years with long period of cool and moist weather in the spring. Tree mortality depends on the size of the host at the time of infection: nursery seedlings and saplings are much more susceptible and die soon after infection, usually during the first year, whereas larger trees gradually die over several years. Large stem cankers reduce height growth (Bernhold, 2008) and may eventually kill the tree by strangulation (Roll-Hansen, 1964). In Fennoscandia, *G. abietina* is the most severe shoot pathogen of native *P. sylvestris* (Roll-Hansen, 1964; Wulff *et al.*, 2006) and introduced *P. contorta* and *P. cembra*; it also damages *P. abies* (Bernhold, 2008). So far, the most severe outbreak of *G. abietina* in Sweden took place in 2001–2003 and almost 500 000 ha of middle-aged pine-dominated forests were attacked. To control the disease, many *P. sylvestris* stands were thinned or clear-cut, resulting in large losses for the forest owners due to the costs of the phytosanitary management and reduction in income (Wulff *et al.*, 2006). *G. abietina* also destroyed many plantations of *P. resinosa* in Newfoundland, Southern Quebec, Canada, and adjacent regions of the USA (Sinclair & Lyon, 2005)

G. abietina is most damaging to the stands of conifers growing close to the limit of their species' ranges and its attacks are favoured by shaded conditions, by dense, badly aerated plantations in which humidity is high, and by weather damage, such as temperature oscillations during shoot elongation.

Control

Complete eradication of the pest is difficult due to the latent nature of the disease (Santamaría *et al.*, 2007). However, suitable control measures could reduce its spread and virulence. In the field, silvicultural measures have been recommended for the control of *G. abietina* such as maintaining a suitable stand density, planting resistant species (Sinclair & Lyon, 2005), or removal of diseased trees from the stand (Aalto-Kallonen and Kurkela, 1985). Moreover, during reforestation, planting of tolerant and resistance hosts is recommended (Santamaría *et al.*, 2007). Also, as pathogen's attacks are facilitated by shaded conditions, by dense, badly aerated plantations, appropriate

spacing between plants and thinning may reduce the risk of infection, and pruning of the lower branches has proved to be a very effective method of reducing the incidence of *G. abietina* on early stage of outbreak when only a small number of trees are infested (Laflamme, 1999). The damage to new plantation on sites conducive to infection can be minimized by planting resistant tree species (Witzell & Karlman, 2000). Delaying pine planting until after two growing seasons following the harvesting of diseased pine trees is recommended (Laflamme and Rioux, 2015) and slash burning or complete removal of the infected slash is needed to minimize the infection risk (Bernhold *et al.*, 2006). However, once *G. abietina* is established in a plantation, it is almost impossible to control it.

Chemical control in the field is possible but expensive. It also might have deleterious impact on biodiversity (including non-target fungi) and can increase pathogen resistance to the fungicide. In general, careful selection of disease-free planting material and suitable planting sites (e.g. the sites not characterized by cool and wet springs and/or by risk of frost damage) at some distance from infested plantations are important control measures. However, in nurseries the disease may be controlled by using fungicides. Several fungicides have traditionally been recommended against this pathogen, such as, chlorothalonil, propiconazole and azoxystrobin applied from May to mid-August were reported as the most effective fungicide against both North American and European races of *G. abietina*, if spraying was carried out in the spring when temperatures were above 0°C (Santamaría *et al.*, 2007; Romeralo *et al.*, 2015). Although use of fungicides against fungal pathogens might be a useful control strategy in the short term, there is currently an increasing interest in finding effective biological control methods, and recent EU legislation (Council Directive, 2009) recommended sustainable forest management and protection of forests and their biodiversity giving priority to non-chemical methods. Identifying alternatives to chemicals (e.g., use the biological control agents or fungal endophyte filtrates) is currently a priority and has been widely studied (Laflamme 1999; Santamaría *et al.*, 2007). Some of these alternative methods have shown a strong antagonistic activity against *G. abietina* (Romeralo *et al.*, 2015).

Phytosanitary risk

G. abietina has not been classified as a quarantine pest by EPPO, but the species is a quarantine pest in three EPPO countries (Morocco, Tunisia and Israel) (EPPO, 2023). In Europe, *G. abietina* has generally been regarded as widespread and it has likely reached the limits of its natural distribution. According to the Implementing Regulation (EU) 2020/2210, *G. abietina* is a regulated pest of *Abies*, *Larix*, *Picea*, *Pinus*, and *Pseudotsuga*, intended for planting, other than seeds, for Protected Zones in Annex III (List of protected zones and the respective protected zone quarantine pests and their respective codes) for Ireland (EU, 2020).

PHYTOSANITARY MEASURES

Planting material of host tree species of *G. abietina* should be chemically treated with the fungicide chlorothalonil prior to movement. Before export to countries free from the pathogen, Christmas trees should be inspected for cankers during the summer before trading. Immersion of diseased seedlings in warm water (55°C) and immersion or spraying with dilute sodium hypochlorite eradicated the pathogen with no apparent loss in needle colour or retention (Hudler and Neal, 1990). Regulatory action by the USA and Canada now prohibits the movement of Christmas trees and nursery stock from areas where the European strain is present.

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