

Phytopathogenic Fungus *Fusarium circinatum* and Potential for Its Transmission in Russia by Insects

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Abstract—The emergence of the ascomycete *Fusarium circinatum* Nirenberg & O'Donnell (teleomorph *Gibberella circinata*), the causal agent of pitch canker of pine in Europe, is an alarming signal to Russia. This review briefly summarizes the analyses of biology of *F. circinatum* and its vectors of dissemination. Open wounds on the bark of the trunk and branches can be the gates for the droplet and airborne infection caused by the *Fusarium* phytopathogens. The infection is followed by the formation of cankers caused by the related phytopathogens, the occurrence of severe resin bleeding, and the death of the crown. In Russia, this pathogen can emerge and spread in the Black Sea coastal area (most likely toward the southeast of Krasnodar) in pine nurseries and plantations. However, it is not clear yet whether the regional climate conditions will be optimal for *F. circinatum* and how competitive it will be in new environments. The insect species associated with the pine trees are considered as an important factor for dissemination of *F. circinatum*, since they can be both the direct carriers of the fungal propagules (i.e., act as the vectors for transmitting the causal agents) and the cause of the physical damage to the pine shoots, branches, and trunk, becoming a gates for infection. After completing the life cycle on the tree infected with *F. circinatum*, the mature insect migrates to another possibly healthy tree, carrying the sticky fungal spores adhering to the surface of its body and leaving them attached to the new host tree. The study describes the insect species associated with the pines (*Pinus*). In addition, it is concerned with analyzing their role in dissemination of *F. circinatum*. It has been proved that, in Russia, there are many insects potentially capable of rapidly spreading the pitch canker of pines if *F. circinatum* invades the country.

Keywords: invasion, insects, pathogen, pine, pitch canker of pines, *Fusarium circinatum*, *Gibberella circinata*

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INTRODUCTION

Invasive phytopathogenic fungi tend to become a problem for both individual countries and entire continents. The incidence of the Dutch elm disease caused by the ascomycetes *Ophiostoma ulmi* (Buisman) Nannf. and *O. novo-ulmi* Brasier is quite well known. The disease was transferred from Europe to Russia. For instance, nearly all the elm trees in St. Petersburg, which were some of the most beautiful decorative elements in the city, died and were cut down over the past twenty years. The bark beetles (Coleoptera: Curculionidae: Scolytinae) *Scolytus multistriatus* (Marshall, 1802), *Scolytus scolytus* (Fabri-

cius, 1775), and *Scolytus pygmaeus* (Fabricius, 1787) were the vectors for transmission of the Dutch elm disease fungus (Brockhoff et al., 2016). They are the organisms associated with the sick trees, which are capable of transmitting the pathogens from one plant to another, spreading the disease (Selikhovkin and Musolin, 2013; Selikhovkin et al., 2014). The mass death of ash trees caused by the ascomycete fungus *Hymenoscyphus fraxineus* (T. Kowalski) Baral et al. (Musolin et al., 2016, 2017), which was previously known as *Chalara flaxinea* T. Kowalski and *Hymenoscyphus pseudalbidus* Queloz et al., has become another problem for Europe. In addition, this fungus is also recorded in Russia (actually in the European part of

the country and its Far East), but it has not caused any mass death among the ash stands yet (Musolin et al., 2017). Maybe the tree microbiota creates conditions unfavorable for this fungus to express its aggression as occurs in Central and Western Europe. In any event, it has not been able to do so yet. The ash trees are under the greater threat from the emerald ash borer *Agrilus planipennis* Fairmaire, 1888 (Coleoptera: Buprestidae), invading European part of Russia recently (Musolin et al., 2017; Selikhovkin et al., 2017). Furthermore, the European countries have currently faced the emergence of some other pathogen, the agent of the pitch canker of pines; it is *Gibberella circinata* Nirenberg & O'Donnell (teleomorph), which corresponds to the anamorph *Fusarium circinatum* Nirenberg & O'Donnell (class Sordariomycetes, order Hypocreales, family Nectriaceae). The pitch canker of pines caused by the rust fungi *Cronartium flaccidum* (Alb. & Schwein.) G. Winter and *Peridermium pini* (Willd.) Lév is a well-known disease within the territory of Russia. Over the past twenty years, one more high-virulence pathogen *F. circinatum* joined these two pathogens in Europe. The main route of transmitting *F. circinatum* is moving infected plant material and wood. The pathways for infection caused by this fungus are various kinds of damage to trees, including physical damage caused by insects. In addition, the pine pest insects carrying the *F. circinatum* propagules are considered one of the key factors for spread of this pathogen (Risk assessment..., 2010). Therefore, the objective of the review was to consider the potential for *F. circinatum* transmission in Russia by insects.

PHYTOPATHOGEN AND VECTORS FOR ITS TRANSMISSION

Phytopathogen. *Fusarium circinatum* on Virginia pine *Pinus virginiana* Mill. in the eastern part of the United States (North Carolina) was first described in the 1940s. Afterwards, it was recorded in Haiti. Forty years after the first description, it was found in tree stands planted in the coastal area in California, where it started infecting different species of pine and neighboring trees of Douglas fir *Pseudotsuga menziesii* (Mirb.) Franco. This pathogen was later found on 50 species of pine in South Africa, Japan, Korea, Mexico, Chili, Uruguay, Columbia, Brazil, and also in Europe: France, Spain, Italy, and Portugal (Bezoz et al., 2017; Musolin et al., 2018). It is currently included in the A2 list of quarantine organisms provided by the European and Mediterranean Plant Protection Organization (EPPO); this pathogen persists in the European and Mediterranean regions, but it is limited in distribution and, therefore, should be subject to control.

The widest presence of *F. circinatum* in Europe is observed in the Spanish coastal area of the Bay of Biscay. In addition, the occurrence of this pathogen is recorded in Portugal, Italy, and different regions of

France, including the upland area nearby the Vosges Mountains in eastern France (48° N, 7° E) (Risk assessment..., 2010). The CLIMEX area modeling software used by the authors of the review of the European Food Safety Authority to assess the risks for the *F. circinatum* distribution can prove that the Black Sea coastal area in Romania might become the marginal zone in Europe for distribution of this invasive pathogen (Risk assessment..., 2010). However, the assessment system has certain serious imperfections, according to the authors of the review, since it was based only on the recorded cases of the phytopathogen entry rates at the registration locations, while the local patterns of the weather variation were not recorded. In addition, a number of scientific works show that the *F. circinatum* distribution generally does not depend on the climate. Thus, the most important factors determining the invasion levels are susceptibility of the host plants (Hodge and Dvorak, 2000) and presence of various kinds of damage to trees (Dwinell et al., 1985; Gordon et al., 2001). Therefore, it can be assumed that the microclimatic conditions in the Black Sea coastal area may appear favorable to this pathogen. For instance, the climate is humid subtropical in the Black Sea area stretching toward the southeast of Krasnodar, through Sochi to Sukhumi, and further into the territory of Georgia. The temperatures in this region never fall below zero degrees Celsius (Kottek et al., 2006; Climate-data, 2018). The principal species of pine distributed throughout this region are Crimean pine *Pinus nigra* subsp. *pallasiana* (Lamb.) Holmboe (subspecies of black pine *Pinus nigra* J.F. Arnold) and Scots pine *Pinus sylvestris* Linnaeus. Some observations of infestations on *P. nigra* and *P. sylvestris* trees in Europe have already been recorded. The aforementioned study allows us to predict the potential distribution of *F. circinatum* in the Black Sea coastal area and the adjacent regions.

In the natural environments, only the asexual spores (micro- and macroconidia) of *F. circinatum* have been found, while the ascospores have been obtained only in vitro. Separation of micro- and macroconidia from conidiophores takes place in presence of condensed water droplets. They can further spread via airborne droplets, for instance, with water running down the tree trunks and branches or in the soil. In addition, the propagules can be found in the air samples in the areas of pathogen distribution. The temperatures for germination of *Fusarium* conidia may be of a wide range (from 5 to 25°C). However, only a temperature above 10°C is favorable to the best mycelial growth. The trees of any age from young saplings to mature and declining trees are susceptible to *Fusarium* (Figs. 1–4). The pathogen starts developing when there is some water in the tree tissues where it occurs, or there is very high humidity of the air. The infection gates can be open wounds caused by different reasons. They may be the mechanical damage, ice wedges, damage caused by insects, etc. The pathogen may



Fig. 1. Intensive resin flow as one of the symptoms of pitch canker of pines caused by fungus *Fusarium circinatum* on insignis pine *Pinus radiata*. Cantabria region, Spain, 2008. Photo by Dr. J.J. Diez; reprinted with permission.

reach the wound via water droplets or particles in the air. In addition, the *Fusarium* spores may be transmitted by the insects. The fungal growth is more intense at an elevated humidity level. Therefore, the rate of the infection spread depends on the climate and weather conditions, primarily on the temperature and humidity variations (EFSA, 2010).

In the case of infection of trees via airborne spores, branches of the last two years die first. In this path of infection, the canker does not spread to the tree crown and trunk. However, a secondary infection usually occurs, for instance, caused by the activity of insects.

In this case, all parts of the tress may be affected. The infection process is followed by the crown dieback, the canker formation on the tree branches and trunk, the occurrence of severe resin bleeding, and the trunk deformation (Figs. 1–4). The developing cankers may encircle the entire trunk, causing the death of the tree (EFSA, 2010).

Phytopathogen spreading. The main route of the global spreading of *F. circinatum* is the movement of infected plant material and wood. Infected imported seeds, seedlings, and wood are the principal sources of infection. The pathogen can survive at moderate tem-



Fig. 2. Pathological development of trunk of insignis pine *Pinus radiata* and intensive resin flow caused by fungus *Fusarium circinatum*. Cantabria region, Spain, 2008. Photo by Dr. P. Martínez-Álvarez; reprinted with permission.

peratures for a year or longer in the infected wood, while in soil, it can remain for a long time. Therefore, soil can be regarded as one more possible source of infection spreading into new areas, since *F. circinatum* can infect pine roots and seedlings (Risk assessment..., 2010).

The forest nurseries producing insignis pine *Pinus radiata* D. Don appeared to be favorable environments for the spreading of *F. circinatum*. This pathogen reduces the germination ability of pine seeds and causes lodging and withering of the seedlings (Risk assessment..., 2010).

Interestingly enough, such specific response of the trees to the damage resulting in resin bleeding may be a factor stimulating the fungal development. The number of resin ducts at the site of damage increases with the injuries, including the cell insult caused by the growing mycelium. The epithelial cells surrounding the resin ducts contain starch, which is an essential component in consumption by fungi (Risk assessment..., 2010).

The insect pests associated with pine trees are among the key factors for *F. circinatum* spreading, since they can be both the direct carriers of the fungal propagules (i.e., act as the vectors for transmitting the



Fig. 3. Sawn insignis pine *Pinus radiata* heavily infected with fungus *Fusarium circinatum*. Cantabria region, Spain, 2015. Photo by Dr. S. Markowskaja.



Fig. 4. Sawn insignis pine *Pinus radiata* with trunk deformation caused by fungus *Fusarium circinatum*. Cantabria region, Spain, 2015. Photo by Dr. S. Markowskaja.

causal agents of *Fusarium*) and the cause of the physical damage to the pine shoots, branches, and trunk, becoming gates for infection. The spores are able to penetrate the plant tissues, its cambium, and the surface layers of a tree through open wounds (Risk assessment..., 2010). However, insects are the most efficient carriers of the pathogen. After completing the life

cycle on the tree infected by *F. circinatum*, the mature insect migrates to another possibly healthy tree, carrying the sticky fungal spores adhering to the surface of its body and leaving them attached to the new host tree. Therefore, one insect can carry the fungal spores to several healthy trees. Some insect species capable of causing a mass infection outbreak or increasing the

density of their populations in the pine stands have the most harmful effects.

The trees weakened by *F. circinatum* become easy targets for bark beetles. This is exactly why bark beetles are considered the cause of spreading the spores of this pathogen (Bezoz et al., 2015, 2017; Brockerhoff et al., 2016). The leaders of the potential *F. circinatum* spreading in Europe are undoubtedly bark beetles called common pine shoot beetles *Tomicus piniperda* (Linnaeus, 1758), *Tomicus minor* (Hartig, 1834), and *Tomicus destruens* (Wollaston, 1985). The latter species (*T. destruens*) prefers the mild Mediterranean climate, occurs in the areas in the west of Europe, Spain, France, Portugal, and Italy, while the larger pine shoot beetle (*T. piniperda*) and the smaller pine shoot beetle (*T. minor*) are distributed throughout the range of pines. In Russia, they can be found widely, including Krasnodar krai, in the Far East, and on the Kola Peninsula.

The common pine shoot beetles usually inhabit weakened trees, including trees infected with *Fusarium*. A significant fraction of beetles of the parent generation and beetles of the new generation carry the sticky fungal conidia adhering to their body (Storer et al., 2004; Risk assessment..., 2010; Bezoz et al., 2017). The beetles migrate to healthy trees and have additional feeding inside the young shoots thus infecting them with the *Fusarium* spores. The shoots tunneled by the bark beetles usually start to break off, opening the gates for infection. It should be noted that the common pine shoot beetles successfully attack quite healthy pine trees in the case of the high density of their populations. In the presence of a pathogen, the beetles can infect the tree even unsuccessfully trying to inhabit it, when the tree can survive the initial attacks of the pests with the resin swamping them.

There may be one more potential vector for *Fusarium* transmission, which is the pine sawyer beetle *Monochamus galloprovincialis* (Oliver, 1975) (Coleoptera: Cerambycidae) in the case of invasion of this fungus into Russia. The adults of this sawyer, similar to the common pine shoot beetles, have additional feeding on the pine shoots, having a good opportunity to infect the healthy trees with the pitch canker of pine. In addition, the pine sawyer beetle makes notches on the trunk with its large and strong mandibles to lay eggs. The pathogen spores can also get into them (when secting the bark and during oviposition). In any case, such a phenomenon associated with the distribution of the causal agent of the pitch canker in a spruce forest during the oviposition process in the pine sawyer beetle of genus *Monochamus* was observed in coniferous stands along the Karelian Isthmus (Varentsova et al., 2017). The pine sawyer beetle does not produce outbreaks of mass reproduction. However, it is a species typical of a pine forest and can be a potential carrier of *F. circinatum*.

One more group of the pine pests may be the efficient pathogen spreading vector represented by pine weevils known as snout beetles (Coleoptera: Curculionidae). They represented by *Pissodes* (*Pissodes castaneus* (De Geer, 1775), *P. (Pissodes) pini* (Linnaeus, 1758), *P. (Pissodes) piniphilus* (Herbst, 1797), *Pissodes (Pissodes) validirostris* (C.R. Sahlberg, 1834), *Hylastes ater* (Paykull, 1800), and *Hylobius (Callirus) abietis* (Linnaeus, 1758). The beetles of the first three species (banded pine weevils *P. castaneus*, *P. pini*, and *P. piniphilus*) can have some additional feeding on the thin bark of pine trunks and branches, gnawing small bites. Because of the high density of the weevil population, there is not enough time for a tree to plug well the damaged area with sticky resin. In addition, the beetle itself, feeding on tree, may infect the wound with the fungal spores attached to its surface. The weevil larvae develop under the bark of the trees weakened by a fire or some other factors (including the development of pitch canker). A high density of the weevil population can be observed in the stands of young pine trees aged 15–20 years and over.

Two other species of weevils (*H. ater* and *H. abietis*) are the most dangerous pests in the natural regeneration of forest cultures, young plants, and seedlings of pine. Certain species of weevils (particularly *Hylastes attenuatus* Erichson, 1836) are known to be capable of carrying the spores of pathogenic fungi *Ophiostoma* spp., *Leptographium* spp., and *Fusarium* spp. (Peverieri et al., 2006; Romón et al., 2007; Bezoz et al., 2017). Larvae of *H. ater* and *H. abietis* develop in the roots or in the area of the root neck of the stumps remaining after cutting or windfall. The beetles additionally feed on the regenerating pine, gnawing rather large bites on the trunks (sometimes, up to 8–10 mm length). In Russia, the large pine weevil *H. abietis* sometimes destroy the natural regeneration in cleared areas (Popovichev and Selikhovkin, 1985), being the most serious pest of pine regeneration in Siberia and in the north of the European part of the country. Adults of *H. ater* and *H. abietis* may live up to 3–5 years, spreading fungal spores all the time. The listed characteristics of biology of the weevils indicate that both of these species may become the key carriers of *Fusarium* in the pine nurseries and forests in Russia.

In addition, the banded pinecone weevil *P. validirostris* and one more pest belonging to quite another insect group, the invasive pine seed bug *Leptoglossus occidentalis* Heidemann, 1910 (Heteroptera: Coreidae), may be significant vectors of the pathogen. The banded pinecone weevil beetles live approximately 3 years and feed on the fresh cones and roots, causing the open wounds and, therefore, infecting them. Adults and nymphs of bug *L. occidentalis* feed only on the cones, spreading the infection from tree to tree. It has not been proved yet whether the pine seed bug is capable of carrying *Fusarium*. However, it is known as a carrier of another pathogenic fungus, *Diplodia pinea* Desm. (Mjos et al., 2010). Recently, *L. occidentalis* was accidentally introduced from North America to

Europe and Japan, where it successfully feeds on the pine trees of more than 40 species (Luchi et al., 2011; Tamburini et al., 2012). In Russia, this species was first found in 2009 (Gapon, 2012); since then, it has been expanding its range in the country (Gninenko et al., 2014).

A large group of bark beetles of genus *Ips* (Coleoptera: Curculionidae: Scolytinae), which includes the most serious pine trunk pests represented by the stenographer bark beetle *Ips sexdentatus* (Börner, 1776) and the engraver bark beetle *Ips acuminatus* (Gyllenhal, 1827), as the vectors for transmitting the *Fusarium* and the other fungi will be less significant. The reason is that the bark beetle adults have additional feeding under the bark of exactly the same trees of their initial larval development or other significantly weakened trees. The engraver bark beetle may attack some branches of viable trees, but it prefers to inhabit irreversibly weakened pines. It is already known that some bark beetles, for instance, *Ips mexicanus* (Hopkins, 1905) and *Ips paraconfusus* Lanier, 1970, carry *F. circinatum* (Bezós et al., 2017). The stenographer bark beetle and the engraver bark beetle apparently have the potential to carry this pathogen to freshly inhabited trees. In any case, the impact of beetles evidently leads to death of such tree specimens even without *Fusarium* or another pathogen, thereby limiting the negative effects of the bark beetles as the carriers of infection.

The risk for infection appears to be somewhat greater from the great spruce bark beetle *Dendroctonus micans* (Kugelann, 1794) (Coleoptera: Curculionidae: Scolytinae) than the other bark beetles, since it can inhabit various coniferous tree species of different age under the varying conditions, including mature and declining pine trees, forest cultures, swamp pines, etc. The growth of the great spruce bark beetle occurs under the bark in the lower part of the trunk or in the area of the root collar for 1–2 years. In addition, the egg and larval galleries on mature trees usually occupy only a part of the trunk circumference. Therefore, the trees inhabited by the pests may remain alive, allowing the infection to develop and becoming the source of infection.

In addition, the twig beetles *Pityophthorus* spp. (Coleoptera: Curculionidae: Scolytinae) may be a significant vector for transmitting the infection. More than 25 species of this genus are known in Europe (Fauna Europaea..., 2017). The twig beetles recognized as secondary pests may inhabit the bark of both the trunk and the branches of viable trees. This allows them to carry the fungal spores successfully from dead trees to relatively healthy trees. Furthermore, some species, for instance, the pine twig beetle *P. glabratus* Eichhoff, 1878 and the Ussuri pine twig beetle *Pityophthorus pini* Kurentsov, 1941, primarily develop on pine trees. In California (United States), the twig beetles are considered a key vector of *Fusarium* (Risk assessment..., 2010). The risk of transmission of the

Fusarium propagules has been experimentally proved for *Pityophthorus pubescens* (Marshall, 1802) (Bezós et al., 2017), *Pityophthorus setosus* Blackman, 1927, *Pityophthorus carmeli* Swaine, 1918, and *Pityophthorus nitidulus* (Mannerheim, 1843) (Storer et al., 2004). In addition, the bark beetle *Polygraphus poligraphus* (Linnaeus, 1758) (Coleoptera: Curculionidae: Scolytinae), capable of developing on the trunks and branches of spruce and pine trees, may be assigned to the same group.

CONCLUSIONS

The conducted review shows there are rather many dendrophagous insects in Russia which can provoke the rapid spread of the pitch canker disease caused by the fungus *F. circinatum*. However, it is not clear yet whether this pathogen will emerge in Russia, when it may happen, and how competitive it may be compared to the native species of pathogenic fungi occupying similar ecological niches in the environments of separate regions in Russia. One may assume that the most favorable region for its distribution will be pine nurseries and young plantations located in the Black Sea coastal areas and the adjacent regions.

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