

# Alien invasive pathogens and pests in the changing environment: Focus on North Europe

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## Preamble

On 12–13 September 2023, a panel of forest pathologists and entomologists gathered in Visby, Gotland Island, Baltic Sea, Sweden. The participants, a total of 20, represented all five Nordic Countries, three Baltic States, and Ukraine, and also joined by the local stakeholders representing the Swedish Forestry Agency and Gotland Administration. The purpose was to conduct a scientific workshop focusing on invasive tree pests and diseases emerging in each respective country, as well as to participate in an excursion to damaged woodland sites of Gotland. The Nordic Forest Research (SNS) financially supported the event (<https://nordicforestresearch.org/n2023-01/>).

**Keywords:** *Corinectria* / *Neonectria* cankers; *Hymenoscyphus fraxineus*; Emerald Ash Borer; *Diplodia sapinea*; Acute Oak Decline; *Polygraphus proximus*; emerging birch pests; Bronze Birch Borer; *Thekopsora areolata*; seedborn conifer fungi

## Forest pests in Iceland

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Birch is the only native forest-forming species in Iceland. At the time of human settlement, almost 1,150 years ago, birch forest and woodland covered about 25% of Iceland's land area, but today the cover is about 1.5%. Birch is the most important tree species. Currently, a total of 80 insect pests are found on woody plants in Iceland, 32 of which are found on birch. Of those, 21 are native (present before 1900), and 11 introduced (after 1900). In recent years, some important birch pests have been introduced to Iceland. There are, for instance, three species of leaf-mining insects, one virtually small moth and two wasps, that feed inside birch leaves on their larval stages. Those are *Heringocrania unimaculella* (first record 2005), *Scolioneura betuleti* (2016), and *Fenusella nana* (2022). Their distribution area has been expanding fast since their introduction, but no leaf-mining species was found on birch in Iceland before 2005. The most important disease is birch rust, caused by fungus *Melampsori-*

*dium betulinum*, damaging trees in winter and predisposing them to other diseases. Successful planting of exotic tree and shrub species in Iceland started by the end of the 19<sup>th</sup> century. In total, 150 new tree and shrub species have been tried. Nowadays, 0.5 % of Iceland are covered by exotic tree species, and 3–4 million seedlings are planted annually. The most important exotic species in forestry are: Siberian larch, Sitka spruce, lodgepole pine and black cottonwood. Their most important pests are green spruce aphid (*Elatobium abietinum*) and the brassy willow beetle (*Phratora vitellinae*). The red-headed pine sawfly (*Acantholyda erythrocephala*) has also been causing more damage in recent years.

**Keywords:** *Betula pubescens*; leaf-mining insects; *Heringocrania unimaculella*; *Scolioneura betuleti*; *Fenusella nana*; *Melampsorium botulinum*

## Recent findings on invasive tree-infesting fungi *Corinectria fuckeliana*, *Neonectria neomacrospora* and *Diplodia sapinea*

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*Corinectria fuckeliana* is a common colonizer of conifer bark wounds, branch stubs and stem cracks. In addition of being a harmless colonizer, it has been reported to cause canker disease on conifers in North and South America as well as in New Zealand. Since the beginning of 2000s, the fungus has been recognized as a serious pathogen of Norway spruce in the Northern Hemisphere, including Finland, Norway, and Sweden. In Finland, similar spruce canker symptoms have been recorded already 90 years ago, but the fungus has not caused noticeable problems until it was discovered causing a serious disease in spruce provenance trials in the early 2000s. Subsequently, since mid-2000s the damage was also increasingly observed in forest areas. Currently, the fungus is confirmed from over 50 forest sites with diseased spruces of all ages. The worst symptoms are seen on 5–25-year-old trees, typically in fast-growing stands on former agricultural land.

Another related pathogenic fungus, *Neonectria neomacrospora* was for the first time reported on fir trees in an arboretum in south-east Finland in 2018. The symptoms on infected trees were bleeding stems, branch cankers, and necrotic shoots. Pathogenicity tests demonstrated the abi-

lity of the fungus to cause top dieback in several species of fir (*Abies balsamea*, *A. fraseri*, *A. sibirica*), but symptoms were also induced on Norway spruce. More recently, in 2022, reports on dying fir trees started coming from several urban and semi-urban areas, mostly from south-west Finland. The disease affects most heavily *A. sibirica* and *A. lasiocarpa*, but the fungus was not isolated from *A. sachalinensis* or *A. homolepis*.

The pathogen *Diplodia sapinea* was found in Finland for the first-time causing tip blight in Scots pine in 2021. The disease has been monitored, and the spread of the pathogen in Finland during the last two years has been recorded. In 2022, it was for the first time observed to cause similar symptoms to juniper, most often growing in the vicinity of the infected pine trees. Predisposing factors to the disease are increased radiation and temperature, draught, and hailstorms, and *Ips acuminatus* is commonly found in the diseased pines in the east coast of Finland.

**Keywords:** *Corinectria fuckeliana*; *Neonectria neomacrospora*; *Diplodia sapinea*; canker; *Picea abies*; *Abies* spp.; *Pinus sylvestris*

## Bronze Birch Borer – to be a threat or not to be?

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The Bronze Birch Borer (BBB), *Agrilus anxius* (Coleoptera: Buprestidae), is native to North America and, thus far, endemic to the region. Its obligatory host tree is birch (*Betula* spp.). The potential threat this species poses to European birch trees has been recognized for several years as the species has been on the A1 list of the European and Mediterranean Plant Protection Organization (EPPO). Since 2019, this Union quarantine pest became one of the priority pests for the European Union. In trials conducted in North America, exotic European birch species, *Betula pendula* and *B. pubescens*, proved to be extremely susceptible to the BBB, suffering higher mortality rates and succumbing more quickly than native North American birch species. Mortality among these trees reached 100%. This may reflect the lack of common evolutionary history between European birches and BBB. Adult beetles have a short lifespan and feed on birch leaves, while larvae develop within the living phloem. Upon hatching, larvae bore through the bark into the cambium zone, creating distinctive zigzag-shaped galleries. They then penetrate the sapwood, pupate there or within the bark, and emerge

through D-shaped exit holes. The feeding process disrupts sap flow, leading to the drying and eventual death of infested branches. Like most insect pests, BBB prefer trees that are stressed, such as those experiencing drought or other environmental pressures. Potential pathways for the introduction of the BBB to the EU from North America include birch plants for planting, birch wood with or without bark (including cut branches), furniture, and other items made of untreated birch wood, as well as birch wood packaging materials and hardwood chips containing birch. Although the EU regulated much of these, some materials are not covered or may not adhere to ISPM 15 standards. Currently, the trade of wood chips from broadleaved trees to the EU is unregulated. Wood chips often contain a mixture of hardwoods, and the presence of birch chips within these loads is uncertain. The quality of chipped wood for energy purposes is generally low, making it more likely to contain insect infested tree parts. Importantly, a small portion of prepupae may survive even through chipping, as demonstrated for another priority pest, the emerald ash borer, *Agrilus*

*planipennis* (McCullough et al. 2007, *Journal of Economic Entomology* 100). Regular surveys are conducted in the EU to detect priority pests. However, the probability of detecting pests among wood chips is known to be very low, requiring extensive sampling volumes that are impossible in practice (Økland et al. 2012, *Scandinavian Journal of Forest Research* 27). Birch trees are prevalent across northern Europe, and not all grow in optimal conditions, particularly in urban, industrial, or abandoned areas. The arrival of the BBB is most likely to occur near harbours and importing companies in rural areas. The likelihood of BBB establishing itself is the greatest in regions abundant with birch trees. Currently, damage to spruce trees due to bark beetles has increased interest in cultivation of mixed stands of conifers and birch. In Eastern Europe,

birch is used as shelter crop for other tree species, when bark beetle killed spruce plantations are replanted. Consequently, the importance of birch is on the rise, magnifying the potential losses should BBB invade and establish itself. Early warning signs of infestation, such as branch dieback, yellowing, and the decline of birch crowns near potential entry points, can prompt timely eradication measures. These indicators are observable by local residents, underscoring the importance of keeping them informed about such symptoms. It is crucial to instruct them to promptly alert plant health inspectors for further investigation and intervention.

**Keywords:** *Agrilus anxius*; European *Betula* spp.; Bronze Birch Borer; EU quarantine pest; invasion pathways; wood chips

## The potential for arrival and establishment by the Four-Eyed Fir Bark Beetle (*Polygraphus proximus*) in the European Union

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The four-eyed fir bark beetle (*Polygraphus proximus*) (FFBB) is a potential invader to the EU. It naturally occurs in Far East Asia: Japan, Korea, China and Russia (Musolin et al. 2022, *Forests*), where it colonises the native species of fir (*Abies* spp.) that are tolerant to the beetle. Consequently, within the native range, the beetle is not economically important. In the mid-1970s, the FFBB invaded central Siberia (Krasnoyarsk Region) and accomplished rapid host-tree species switch to the Siberian fir (*Abies sibirica*), resulting in massive lethal attacks. Death rate of attacked Siberian fir trees is about 75%. Feeding on new host-tree species may be associated with the depletion of initial host trees coinciding with high beetle abundance. Currently, the invaded area comprises 1/3 of Siberian fir distribution. Most recent mass outbreaks have been observed in Udmurtia, eastern Volga region (approx. 2,000 km west from Krasnoyarsk). In both cases, the most likely means of spread is by raw-timber railroad transportation. Adults are small (2.5–3.5 mm) yet can fly several kms. The beetle has a very long flight period (May–June, Aug–Sept), accomplishing sister broods. The FFBB has a 2-yr life cycle in most regions, 1-yr in northern locations. Overwintering larvae and adults survive –37.3°C. The FFBB utilizes natural disturbances and drought, mostly occupies weakened trees, and may benefit from pathogens such as *Heterobasi-*

*dion* and *Armillaria*. The beetle prefers smaller trees (thin bark) that occur in groups. FFBB genetic research suggests distinctive populations and thus multiple introductions in Russia. During pheromone trapping at three sites in 2019–2021 in eastern Finland next to timber transportation routes, and close to the Russian border, no FFBB specimens were captured. The factors favouring likelihood for appearance within the EU are temperature tolerance of the beetle, efficient reproduction, and rapid adaptation to new host tree species. Factors that also increase outbreak likelihood are drought, windstorms, conifer monocultures, and abundant smallish host trees. Upon eventual invasion the FFBB might impose risk for Silver fir *Abies alba*, native to Central Europe. Risks for Scots pine and Norway spruce may be considered low, despite accidental observations; it would require another host-tree (genus) switch. Where to expect the first EU findings? Due to the Russian attack on Ukraine and consequent decline of timber import, nowhere. But basically, any timber storage site or harbour, or arboretum, at or near the EU eastern borders represents a potential threat.

**Keywords:** *Polygraphus proximus*; bark beetle invasion; *Abies sibirica*; central Siberia; *Abies alba*; timber transportation

## Occurrence and pathogenicity of *Corinectria* spp. – a canker disease of *Abies* and its association with *Polygraphus proximus*

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The preliminary study is presented on the occurrence and pathogenicity of *Corinectria* spp. – a canker disease of fir (*Abies*), and its eventual association with *Polygraphus proximus*. On branches of Siberian fir, cankers and the presence of *Corinectria*-like fungi have been regularly observed in the invasive areas of the beetle in central Siberia. From the cankers, fungal cultures resembling *Corinectria* spp. have been isolated. Molecular analyses have demonstrated that the isolates do indeed belong to the genus, but also represent two previously unknown species, preliminary assigned names of X and Y. The aims of the work were to investigate the pathogenicity of those fungi to different conifers (other than Siberian fir), and the eventual association of those fungi to the bark beetle. Results demonstrated that both *Corinectria* spp. X et Y caused dieback exclusively to Subalpine fir (*Abies lasiocarpa*, a species from western North America), but neither to Norway spruce, Scots pine, Douglas fir, nor to Siberian larch. Can *Polygraphus proximus* be a vector for new *Corinectria* species? To answer this, adult beetles of *Polygraphus proximus* were sampled in central Siberia and analysed using molecular methods

(sequencing of fungal DNA). As a result, *Corinectria* sp. X was commonly detected in the beetles. Conclusions: i) *Corinectria* sp. X et Y are pathogenic to at least two *Abies* species (*A. sibirica* and *A. lasiocarpa*); but more species of fir need to be tested, in particular European native *Abies alba* (tests ongoing); ii) *Corinectria* sp. X is a more aggressive pathogen than *Corinectria* sp. Y; iii) among conifer species tested, European *Corinectria fuckeliana* (used as control) was pathogenic exclusively to Norway spruce; iv) neither *Corinectria* sp. X nor Y caused disease symptoms on *Pinus*, *Larix* and *Pseudotsuga* (both the fungal species currently are being described as the new species for science, or *sp.nov.*); v) *Polygraphus proximus* is a potential vector for the new *Corinectria* species X.

**Keywords:** *Corinectria* spp.; canker; *Polygraphus proximus*; *Abies* spp.; fungal species novae; pathogenicity tests

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## *Thekopsora areolata* – the causal agent of Cherry Spruce Rust

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Swedish forests comprise 23.6 M ha of productive stands, 40% of which is Norway spruce, and 260,000 ha are harvested yearly. In total, 200 million Norway spruce seedlings are planted annually, 70% of which are produced from seeds collected in seed orchards. Rust fungus *Thekopsora areolata* is the causal agent of Cherry Spruce Rust damaging cones and seeds, which pose a threat towards successful forest regeneration. Today there is a lack of seeds produced in seed orchards in many planting zones, mainly due to irregular flowering, but also due to the disease, which occurs as regular epidemics and can result in seed losses by 50–100%. Recent studies have demonstrated high genotypic diversity of this rust fungus, and no clear genetic differentiation between locations has been detected, indicating random mating and long-distance dispersal. The pathogen produces several types of spores, two of which, aeciospores and urediniospores, are crucial for the airborne

spread. Moreover, those spores infect bird cherry (*Prunus padus*), the intermediate host of the pathogen. Thus, the research has also addressed infection process, aiming to answer the following questions: i) how similar/different are aeciospores and urediniospores? ii) do aeciospores and urediniospores use the same mechanism to infect *P. padus* leaves? iii) which genes are involved in the infection process? Conclusions: i) *T. areolata* is dominantly outcrossing and highly polymorphic with high gene flow; ii) its basidiospores are spread during at least four consecutive days of rain; iii) its multiple basidiospores infect pistillate cones, where vegetative spread and mating occurs; iv) aecio- and urediniospores largely use the same genetic machinery.

**Keywords:** *Thekopsora areolata*; Cherry Spruce Rust; *Picea abies*; spruce cones; *Prunus padus*; spruce seed orchards

## Seedborne fungi of commercial coniferous species

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The study concerned seed-borne fungal communities of conifers and important fungal pathogens, such as *Siroccocus conigenus*, *Sydowia polyspora*, *Phoma herbarum*, *Diplodia sapinea*, and others. In Sweden, most reforestation originates from commercial seed material with Norway spruce (*Picea abies*) and Scots pine (*Pinus sylvestris*) being the dominating tree species produced. Still, there is also larch (*Larix* sp.) seedling production. Sweden imports about 13% of the seed material, and from a phytosanitary point of view, traded seeds can be the carriers of fungal pathogens. In this respect, seed-borne fungi are important. Those can accomplish horizontal transmission (on seed surface, vegetation, soil, airborne dispersion, water, seed processing) and vertical transmission (inside the seed, via mother plant, or colonisation of seed surface). To avoid the spread of fungal pathogens through seed material, an integrated pest management (IPM) is necessary for sustainable seedling production. The IPM includes reduced usage of fungicides and prioritizes the use of preventative control methods. In the study, fungal communities on seeds were investigated to identify seed-borne fungi on commercially important conifer seeds, with a focus on fungal pathogens. The following hypotheses were investigated: i) fungal communities will differentiate between the seed surface and the seed tissue; ii) fungal communities will be host-specific inside the seeds; iii) fungal communities differentiate between geographical regions; iv) there is higher fungal diversity in the southern regions. A total of 44 seed origins were included: 21 spruce and pine seed orchards

in Sweden and 13 spruce, pine and larch seed orchards in Finland, plus 10 (5+5) seed stands of spruce in Poland and Belarus. DNA was extracted from both the seed surface and the seed tissue and subjected to molecular analyses as amplification of ITS2 region and sequencing. The results demonstrated: i) a differentiated fungal community composition between the seed surface and the seed tissue; ii) a strong fungal community host-affinity on the seed surface, while no strong host associations from the seed tissue; iii) geographical differentiation in fungal community composition; iv) fungal diversity were higher on spruce seeds but no clear regional differences found. Common fungal pathogens were *S. polyspora* (13.3%), *P. herbarum* (11.2%) and *S. conigenus* (3.8%). A low abundance of *D. sapinea* was detected. In conclusion, the fungal community composition was confirmed to differ between seed surface and seed tissue. Opposite to the hypothesis, the fungal community composition was host-specific on the seed surface, and spruce and pine seeds were confirmed to have regional dependence on the seed surface.

**Keywords:** *Cladosporium* sp.; conifer seedborne fungi; *Siroccocus conigenus*; *Sydowia polyspora*; *Fusarium* sp.; *Diplodia sapinea*; seed orchard

### Acknowledgements

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## *Diplodia sapinea* as a contributing factor in the crown dieback of Scots pine after a severe drought

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The study investigated *Diplodia sapinea* as a contributing factor in the crown dieback of Scots pine after a severe drought. The fungus is the causal agent of Diplodia tip blight (DTB), and is an opportunistic forest pathogen. Under “normal” conditions, the fungus is a latent inhabitant of asymptomatic pine trees. The symptoms emerge and develop on trees following abiotic stress. Drought-exposed Scots pine responds with stomata closure and needle biomass loss, resulting in crown dieback leading to reduced tree vigour. The initial emergence of the disease in the Nordic countries was first observed in Swedish forests in 2013, with outbreaks reported close to Arlanda, and subsequently on Gotland and in the southern parts of the country. In 2018, exceptional temperature anomalies and

drought were recorded in Scandinavia. Consequently, widespread crown dieback was observed on Scots pine on Gotland in late summer; symptoms were consistent with Diplodia tip blight. The presence of the disease was then confirmed and had a large ecological impact on pine stands on the island. The objective of the presented work was to describe the mortality and recovery of drought-induced crown dieback and its relation to DTB disease in Scots pine stands on Gotland. The hypotheses: drought effect in combination with *D. sapinea* infection limit recovery of the trees: i) severely damaged trees (> 70% of the crown) show higher mortality rates; ii) mildly damaged trees (< 30%) show a higher probability of recovery; iii) *D. sapinea* is abundant independently of tree and site health; iv) spore

dispersal of *D. sapinea* is associated with the occurrence of dieback of pine at the sample site. Visual estimation of crown dieback was conducted in 2018, 2019, and 2020 and included healthy and disease-affected sites and trees, and asymptomatic and symptomatic shoots. The results were as follows. Increasing defoliation was observed during the 1<sup>st</sup> year post-drought and was also recorded on newly grown shoots. Crown dieback observed during the 1<sup>st</sup> year doubled. The abundance of *D. sapinea* in twigs and spore traps confirmed the presence of the pathogen on affected and healthy sites and was detected during all seasons. It correlated with high precipitation during sampling, but no significant seasonal pattern was noted. Latent *D. sapinea* infections were also detected in asymptomatic twigs. Site properties likely contributed to contrasting dieback patterns between affected and healthy sites. There was a high

potential for canopy recovery even in trees with high defoliation levels. Surviving trees showed a clear recovery within the 2<sup>nd</sup> year, as manifested by newly grown shoots. Nevertheless, *D. sapinea* contributed to defoliation and potentially affected post-drought recovery. Severely affected pines experienced increased mortality, which in those trees exceeded 50%. As the recovery of the surviving trees was independent of the previous dieback level, immediate sanitary cuttings of symptomatic trees may not be necessary.

**Keywords:** *Diplodia sapinea*; *Pinus sylvestris*; drought; Gotland Island; defoliation damage; post-drought recovery

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### Field investigation: damaged birch stands, Uråsa, Småland, South Sweden

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A new phenomenon of damage in birch stands has been reported. It was observed in self-regenerated birch stands after the storm Gudrun 2005 (age 18 years) in Småland, south Sweden. The stands were thinned twice, the last time a few years ago. The problems with birch were first noticed in 2019, the year after a severe drought. No other tree species in the area are affected. The affected area comprises about 1,000 ha, and consists mainly of flat, relatively moist soil, but includes also healthy trees (stands). Both individual trees and stands are affected. In the areas visited, 'all' birches showed symptoms, even younger ones (self-regenerated in following years after 2005). The symptoms include crown thinning, dead shoots and dead

buds, losing the top, stem breakage at 2–4 m height, and tree death. Molecular analyses of symptomatic tissues (direct sequencing of DNA) revealed the presence of the following fungi: *Peniophora incarnata*, *Hormonema carpetanum*, and *Alpinaria* sp. The latter two have been found to be associated with shoot dieback of *Betula pendula* in Latvia (unpublished). A study focusing on this new damage phenomenon has been initiated, and is coordinated by the Department of Forest Mycology and Plant Pathology, Swedish University of Agricultural Sciences, Uppsala.

**Keywords:** *Betula pendula*; dieback; *Peniophora incarnata*; *Hormonema carpetanum*; *Alpinaria* sp., forest regeneration

### What can we learn by fungal community profiling and trait analysis of the invasive ash dieback pathogen *Hymenoscyphus fraxineus* and indigenous fungi?

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The review summarized the current understanding of the biological traits of the invasive ash dieback pathogen *Hymenoscyphus fraxineus* in comparison to those of the harmless ash leaf associate and saprotroph *Hymenoscyphus albidus*, indigenous to Europe. Among other features, it was noted that the pathogen has larger diameter of the fruiting bodies (up to 8.5 mm), as compared to the harmless relative (4 mm), implying its higher fecundity. Moreover, the pathogen accomplishes airborne spread by genetically diverse spores (sexual ascospores), while airborne spores dispersed by the *H. albidus* are clonal (ascospores genetically identical to the parental fruiting body). The pathogen also has the capacity to produce different type of

spores (asexual conidia), which are absent in *H. albidus*. The conidia of *H. fraxineus* are known to serve as spermatia that facilitate mating between sexually compatible strains, whereas their role in disease epidemiology, if any, remains unknown. It is pointed out that *H. fraxineus* is superior to *H. albidus* in capacity to secure sporulation niche, namely fallen leaves, on which fruiting bodies dispersing spores are produced. Another specific trait of the pathogen is the ability to induce local necroses in leaf tissues relatively early in the growing season, whereas *H. albidus* can apparently induce similar symptoms, but only in autumn-senescence weakened leaves. These contrasts may well reflect differences in the fecundity and subsequent infection

pressure by the two fungi. In their saprotrophic domain (colonised fallen leaves), both fungi tend to suppress fungal diversity within the areas they have managed to secure with pseudosclerotial layers. It is concluded that the high investment in survival structures (fruiting bodies and pseudosclerotia), high fecundity, and production of recombinant offspring by *H. fraxineus* (pathogen) may represent an adaptation to harsh environmental conditions, the native hosts of the fungus (different *Fraxinus* spp.) spanning across multiple climate zones in Asia, including regions

with cold dry winters, and either hot, warm, or cold summers. Consequently, the pathogen appears to have evolved a strategy that is typical to unstable or unpredictable environments. By contrast, *H. albidus* (saprotroph) appears to have a strategy that is typical to stable or predictable environments.

**Keywords:** *Hymenoscyphus fraxineus*; *Hymenoscyphus albidus*; ash (*Fraxinus*) dieback; invasive pathogen; indigenous fungus; biological traits

### Ash (*Fraxinus* spp.) dieback monitoring in Estonia

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The results of long-term ash dieback (ADB) monitoring in Estonia are presented. Ash dieback caused by the invasive fungus *Hymenoscyphus fraxineus* has been present in Estonian forests since the early 1990s. The disease affects trees in all age classes, regardless of growing conditions. The aim was to check what the real situation of in Estonian forests is. Monitoring of 20 stands was conducted where *F. excelsior* is a canopy forming tree species (at least 20%). On each site, stem DBH (diameter at breast height) was measured and tree health status (score 1–5, ranging from symptomless to dead) was estimated for 20 ash trees. The average height of the stand and its basal area were also estimated. Moreover, health monitoring of seedlings has also been accomplished in 20 sampling areas (10 under old forest stands + 10 on clear-cut areas) on which ash is/was a canopy forming tree species. All seedlings within 10 × 10 m area plots were counted and their health status was evaluated. The results were compared with a survey carried out in the same areas in 2016. Those areas are assigned to be re-evaluated at 3-year intervals. Finally, the monitoring of exotic *Fraxinus* species in Estonian parks

and cities has been conducted. Using historical records and map data, the exact locations of several arboreta were determined, as well as the presence in those of several exotic species of ash. DBH, tree height and tree crown diameter were measured and general health condition of each tree was evaluated. The following conclusions were drawn: i) overall health of *F. excelsior* seedlings is good, the health of old trees varies between different regions of the country; ii) the overall health of seedlings has improved significantly when compared to the previous surveys; iii) the western part of the country stands out with its large percentage of dead trees among both seedlings and old trees (consequence of higher *Fraxinus* percentage in the stands); iv) the exotic ash species growing in Estonian parks and arboreta are in good overall health, only *F. mandshurica* seems to suffer more; v) *F. pennsylvanica* has been by far the most common exotic *Fraxinus* species planted in Estonia.

**Keywords:** *Hymenoscyphus fraxineus*; ash (*Fraxinus*) dieback; invasive pathogen; woodland monitoring; natural regeneration; *Fraxinus mandshurica*

### The ash dieback situation in Denmark, and recommendations for disease management

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The pathogen fungus *Hymenoscyphus fraxineus* originates in Far East Asia, where it is a harmless endophyte / weak leaf pathogen. In its invasive areas of Europe (e.g. Denmark), impact in mature stands becomes visible 5–10 years after arrival of the fungus, and severe symptoms develop 5–10 years later. The disease has a more externally visible impact on older trees and forest stands, but also young stands are heavily affected. Young trees die faster and more frequently, but sick older trees also (slowly) die. Thus, of monitored ADB-symptomatic trees, 35% of young trees and 85% of mature ash remained alive after 10 years; of trees with severe crown symptoms in 2010,

by 2021 all young ash died, and 55% of mature ash died (Madsen et al. 2021, *Forest Ecol. Manag.*). Loss of diameter increment was estimated to be about 60% (Bruno Bilde Jørgensen, unpubl.). In heavily infected stands, even trees with healthy-looking crowns can have extensive basal bark necroses caused by honey fungus (*Armillaria*). This may affect long term survival as well as timber quality. Although susceptible trees show severe symptoms followed by slow decline, certain numbers of trees remain healthy-looking. One example could be the Danish experiment, where young ash seedlings were planted in 2004 and subsequently suffered severe impact of ADB; in 2021 healthy-

looking survivors comprised only 6%, yet those hopefully will be future seed orchard. Indeed, nursery plants show damage in the 2<sup>nd</sup> or 3<sup>rd</sup> growth season, and regeneration and young plantings suffer low survival. But on the other hand, healthy trees seem to produce more progeny and as a result tolerant *Fraxinus excelsior* stock is slowly becoming available. At the age of 10–50 years, fast development of ash dieback is being observed, but many stands are expected to be still present after 10 years and longer. Thus, it is advised to look for tolerant trees and keep them or take graft material. While planting, mix in other tree species: alder, oak, poplar, Sitka spruce, and perhaps sycamore. Two options are recommended for severely affected stands: i) clear-cut those and start over (with other tree species); ii) ... or turn your back, let nature take its course, and wait for the tolerant stock.

Management of ADB-symptomatic ash at the age of 50+ depends on site factors, e.g. soil moisture and honey fungus. In general, one should expect a slow decline of those over a period of 20+ years. As pointed out above, diameter growth decreases fast at ADB impact. In such stands, monitoring and management strategies are important. Epicormic shoots in crowns are “normal”, yet when those appear on stems, it is a warning signal. Basal lesions are initially hard to detect in older trees, but it should be kept in mind that those imply discoloration of wood, le-

ading to loss of timber value. Therefore, it is recommended to fell timber trees exhibiting symptoms described above (salvage felling). Ash dieback is a forestry problem, and it is expected that it will gradually get worse. However, most ash forests will last for years, which may provide the possibility; will it be long enough to propagate tolerant ash? Indeed, current experiences demonstrate that many old ash trees persist. Even in forests. But particularly in open landscapes and parks. Therefore, one should keep ash wherever possible. In brief, in forest stands it is recommended to cut sick ash trees when it provides the possibility to save commercial timber value (it can be dangerous to fell dead or dying ash trees – use harvesters). Otherwise, leave natural ash woodlands untouched, if possible, as the selective sanitary felling improves only visual health, not actual health. Diseased landscape and urban ash trees will survive even longer; here, prune rather than fell to ensure aesthetic value, as here the aesthetics matter. Thus, it is recommended to retain urban ash trees in parks and along roads (unless they must be cut for safety reasons). In each instance, minimize safety issues and accidents, costs of monitoring and intervention.

**Keywords:** *Hymenoscyphus fraxineus*; ash (*Fraxinus*) dieback; invasive pathogen; woodland monitoring; ash dieback management; natural regeneration

## Acute Oak Decline in Europe

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Acute Oak Decline (AOD) is a recently described decline-disease that was first recognized in the United Kingdom (2009) and has been increasingly observed in mainland Europe over a broad geographic range. Over the past five years (2019–2024) AOD has been newly reported in France, Italy, Latvia, Poland, Portugal, Slovakia, Spain, and Switzerland, thus from the utmost south-west to north-east of the EU. The disease has been found on pedunculate oak (*Quercus robur*) and sessile oak (*Quercus petraea*) from northern to southern Europe, in cork oak (*Quercus suber*) in southern Europe and non-native oaks such as Turkish oak (*Quercus cerris*) and northern red oak (*Quercus rubra*). There are four diagnostic symptoms of AOD: i) weeping patches on stems; ii) cracks in the outer bark; iii) necrotic lesions in inner bark; iv) larval galleries of the *Agrilus biguttatus* beetle, native to some parts of Europe. Two bacteria were consistently isolated from affected oak lesions, namely *Brenneria goodwinii* and *Gibbsiella quercinecans*. Several other bacterial species were also frequently isolated, including *Rahnella victoriana* and *Lonsdalea britannica*. However, there is a body of evidence to support that the central cause of stem necrosis is *B. goodwinii*, which encodes a type III secretion system and associated effectors. Recent studies found that *B. goodwinii* is an oak endophyte which does not survive long outside

of the host and may switch between commensal and pathogenic lifestyles. In contrast *G. quercinecans* is a generalist which can persist in many environments. It has previously been suggested that *B. goodwinii* and *G. quercinecans* cooperate within oak tissues. The ability of *B. goodwinii* and *G. quercinecans* to cause larger necrotic lesions when grown together has been demonstrated, and these bacteria in combination have fulfilled Koch's postulates using modern molecular techniques in combination with traditional plant pathology methods. Moreover, it was also demonstrated that co-culture with larvae of *A. biguttatus* increases bark necrosis, and expression of plant pathogenicity genes within *B. goodwinii*, including type III secretion system effectors. Oaks which suffer from bacterial necrosis are thought to be in the final stages of decline and are likely to have been affected by multiple abiotic stressors such as drought and higher temperatures. The increasing unpredictability of climatic conditions threatens to increase the incidence of AOD in European forests as stressed oaks enter a decline spiral and the trees become suitable candidates for *A. biguttatus* infestation and bacterial necrosis.

**Keywords:** Acute Oak Decline; *Agrilus biguttatus*; necrosis; *Brenneria goodwinii*; *Gibbsiella quercinecans*; bacterial tree disease



## Emerald Ash Borer in Ukraine: a review and distribution survey

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Currently, two invasive pests affecting ash trees (*Fraxinus* spp.) are present in Ukraine, ash dieback (ADB) causing fungus *Hymenoscyphus fraxineus*, and the wood-boring buprestid beetle Emerald Ash Borer (*Agrilus planipennis*; EAB). For the first time, the beetle has been detected in eastern Ukraine in 2019. Both native European ash *Fraxinus excelsior* (most common in forests) and introduced green ash *F. pennsylvanica* (in field shelterbelts) are susceptible. In 2019, no EAB was found in forest stands, and the beetle was observed only in shelterbelts. The total area of the outbreak comprised 13.3 ha in 2019 in one region, but by 2023, this area had surged significantly, expanding to 1211.7 hectares in three regions. Attacks by EAB and ADB are often accompanied by the ash bark beetles *Hylesinus crenatus* and *H. varius*, galleries of which are observed both on thin branches and under thick bark. EAB eradication attempts by removal of infested trees have been conducted during October 2019 – March 2020. Subsequent EAB monitoring included: i) trap lure option, using green and violet traps, deploying those in the canopy in sunny location, randomly in locations, where no EAB has been detected, and more densely in locations, where the EAB was found; ii) trap trees, by girdling young and old trees, making wounds on stems, and checking those for the attacks after flying period. Nevertheless, a general trend observed in all three locations during field surveys in 2019–2022 was that the EAB is expanding further west.

**The beetle was commonly found also on ash trees with visually healthy crowns, e.g. not exhibiting dieback (ADB) symptoms. Consequently, the results demonstrated that EAB attacks and kills *Fraxinus* trees indiscriminately of their health status.** Furthermore, results of EAB spread capacity have revealed that in a year there is 50% chance for the EAB to be found within 10.5 km radius from the point of initial spread, and less than 5% chance for the EAB to be found beyond ca. 22 km from the point. Generally, in Ukraine the estimations for EAB yearly spread rates are up to 16.5 km radius from the point of the advancing (EAB) front (data for 2022, Jan). More recently, the evidence was obtained for spread of the EAB by “hitchhiking” (by transport vehicles, as railway and cars), as in 2022 the EAB has been found in the parks of Kyiv, thus at over 500 km distance from its geographically continuous invasive populations in eastern Ukraine.

**Keywords:** *Agrilus planipennis*; Emerald Ash Borer; *Fraxinus* (ash) dieback; *Hymenoscyphus fraxineus*; *Hylesinus* bark beetles

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## Alien forest pathogens and pests: recent studies in Latvia, and other on-going research related to decay fungi and root rot biocontrol

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In Latvia, Emerald Ash Borer (*Agrilus planipennis*) was monitored in years 2021–2023. In 2021, six European ash (*Fraxinus excelsior*) stands close to the Latvia–Russia / Latvia–Belarus border were surveyed, and window traps were used. In 2022, one extra plot, located in Northern Latvia (close to the Estonian border), was included in the survey and pheromone traps were used. In 2023, additional twelve plots located along the main transport infrastructure (arterial roads and railways), using pheromone traps, were investigated. Emerald ash borer has not been detected at any of the nineteen investigated sites.

In 2021, the occurrence of invasive needle pathogens was investigated in 165 young Scots pine stands

and 10 arboreta and urban green space areas with pines. *Dothistroma septosporum* was detected all over the country, in 49 stands and 4 arboreta, and *Diplodia sapinea* as well, all over the country, in 30 stands and 5 arboreta. *Lecanosticta acicola* was detected only in 3 arboreta in central Latvia. In 2022, a total of 50 young Scots pine stands and 9 arboreta and urban green space areas with pines were investigated. Infections of *Dothistroma septosporum* were detected in 47 of those, and *Diplodia sapinea* in 20, and again, in all parts of the country.

Our investigations of wood-inhabiting fungi focused on the following species of deciduous trees: silver birch (*Betula pendula*), European aspen (*Populus tremula*), small-leaved linden (*Tilia cordata*), Norway maple

(*Acer platanoides*), and common oak (*Quercus robur*). Stumps and stems of birch and aspen were studied on more than 180 selective thinning sites and clear-cuts. Wood samples from five stumps per stand were collected, and subjected to pure culture isolations, morphotyping, and sequencing. *Sistotrema brinkmannii* and *Bjerkandera* spp. were dominant saprotrophs for both tree species. Also, the root rot pathogen *Heterobasidion* was isolated from some birch stumps. Furthermore, a total of 366 wood samples were taken from 60 stems of aspen, and 360 samples from 60 stems of birch. The main pathogens found were *Phellinus* spp., detected in 55% of aspen and 13% of birch stems, and *Armillaria* spp., detected in 12% of birch. Among the saprotrophs, *Bjerkandera* spp. dominated (yet occasionally present in living trees). In a similar manner, wood samples were taken from living stems on 34 woodland sites featuring maple, linden, and oak, situated mainly in the central parts of Latvia, from south to north. A total of 15 trees of each species were sampled on each site at stem height 1.3 m, and 675 samples collected. Most often isolated fungi belonged to the genera *Alternaria*, *Fusarium*, and *Epicoccum*. Among wood decay fungi, *Heterobasidion* and *Peniophora* were occasionally isolated.

The antagonistic ability of saprotrophs *Phlebiopsis gigantea* and *Hypholoma* spp. to control root rot pathogen *Heterobasidion* spp. was evaluated. The hypothesis was drawn that the efficacy of mixed suspensions of *P. gigantea* and *Hypholoma* spp. to prevent *Heterobasidion* is higher than efficacy of each individual species. In the first field experiment, wood discs (84) and log pieces (84) of *Picea abies* and *Pinus sylvestris* were treated with the Rotstop and Latvian PG382 *P. gigantea* isolates, and with *Hypholoma capnoides* and *H. fasciculare* isolates. After 5 days of exposure (at 8–17°C), no *Heterobasidion* infection was detected in *P. sylvestris* wood discs treated with the Rotstop and Latvian PG382 *P. gigantea*

isolates, while in those treated with *H. capnoides* infection rate was 28%, treated with *H. fasciculare* was 57%, and in untreated controls was 57%. By contrast, among *P. abies* wood discs treated with the Rotstop and Latvian PG382 *P. gigantea* isolates 71% and 86% were infected by the pathogen. In the discs treated with *H. capnoides* infection rate was 86%, treated with *H. fasciculare* was 100%, and in the untreated controls 85%. In the log pieces, the mean efficacy of Rotstop and Latvian PG382 *P. gigantea* isolates was estimated as 70% and 100%, respectively, that of *H. fasciculare* was 18%, and that of *H. capnoides*, no efficacy was observed.

In the 2<sup>nd</sup> experiment the effect of treatment of *P. abies* stumps with Latvian isolates of *P. gigantea* PG382 and PG182 in mixed suspension with *H. fasciculare* was investigated. Here, the efficacy varied between 70% and 100%, and no significant differences were found between the effect of mixed treatments as compared with those in which each respective *P. gigantea* isolate was applied, and as compared with the Rotstop treatment. In conclusion, the use of mixed suspensions of *P. gigantea* and *H. fasciculare* did not increase the efficacy of *Heterobasidion* control as compared with the treatments by *P. gigantea* isolates (incl. Rotstop) alone.

**Keywords:** *Agrilus planipennis*, *Dothistroma septosporum*, *Diplodia sapinea*, *Lecanosticta acicola*, wood decay, *Phlebiopsis gigantea*, *Hypholoma* spp.

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## Distribution of *Diplodia sapinea* (Fr.) Fuckel in Lithuania

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For the northern Baltic region, *Diplodia sapinea*, a well-known pine pathogen around the world, was first recorded in Estonia on Austrian pine (*Pinus nigra*) in 2007. In 2012, the native Scots pine (*Pinus sylvestris*) was found symptomatic: first in Estonia, then in Latvia, and in 2013, in North-West Russia. In Lithuania, *D. sapinea* was first reported (without molecular verification) on Scots pine in 2000. In a study conducted during 1994–1999, thirty species of *Diplodia* were identified, and *D. sapinea* was one of the most common species. A decade after the first report on *D. sapinea* in Lithuania, the pathogen was found to be associated with pine dieback, and during the following ye-

ars, the pathogen spread and caused notable losses. Yet, during the period 2000–2007 its outbreaks were not detected, but in 2008 the damage was again recorded. Subsequently, in 2009, *D. sapinea* diseased area in south-eastern Lithuania comprised 29 ha. In the following years, sporadic damage by *D. sapinea* occurred periodically. For example, those were not recorded in the period 2012–2016 and 2018–2020, yet the increase of the damage was observed in 2011, 2017 and 2021. Both mature trees and seedlings exhibited symptoms of the disease. Diseased trees were usually infested by pests. The observed fluctuations could be explained by the fact that disease symptoms become vis-

ible when trees are weakened by stress. As *D. sapinea* has an endophytic mode in its lifecycle, this makes it difficult to be detected before the symptoms do appear. Climatic factors as drought combined with high temperatures have made favourable conditions for the spread of the pathogen. In 2008, for example, the trees were first weakened by droughts and high temperatures, and as a result they were infected by *D. sapinea*, and later, following insect (mainly eight-toothed bark beetle, *Ips acuminatus*) attacks, the trees suddenly succumbed to the dieback. However, until 2009 *D. sapinea* was rare and affected only individual pine trees. But in 2009, high humidity in May and June created favourable conditions for the growth of the fungus. Additionally, dense-growing, overgrown with unwanted vegetation or exposing to other stress factors made young trees susceptible to massive infections. Another example

of climatic impact was observed in 2021, when the warm autumn weather was favourable for pine to initiate bursting of new shoots that were subsequently infected by the pathogen. Consequently, *D. sapinea* spread massively after a warm and humid autumn and mostly affected new shoots that have not yet fully matured. Snowy winter and wet and prolonged spring also create conditions for the further development of *D. sapinea*, as well as the loose snow cover that formed on the unfrozen soil. Notably, spraying of insecticide Foray 76B (against needle insect pests) did not influence the abundance of the *D. sapinea*, as it, despite the treatment, was detected in all investigated *P. sylvestris* stands: treated, untreated, and healthy-looking.

**Keywords:** *Diplodia sapinea*; *Pinus sylvestris*; dieback; drought; *Ips acuminatus*; insecticide Foray 76B

### ***Ips typographus*: the current status in Lithuania, its phenological adaptation in response to climate change**

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Recently, the massive attacks of *Ips typographus* were triggered by the warm winters, dry summers and increasing squalls, resulting in a high amount of windbreaks and windfalls in Lithuania. In 2023, abundance of the pest was two and three times higher as compared to 2022 and 2021, respectively. As a result, the state of the natural disaster was declared in five municipalities of Lithuania in August 2023. Flight activity and abundant development of the 2<sup>nd</sup> generation of *I. typographus* in 2023 obviously indicated that the damages will increase in 2024 and are predicted to be 1 million m<sup>3</sup> of infested timber of *Picea abies*.

The main part of the previous studies in Lithuania were based on bark beetle phenology and performed still in the past century. At that time three beetle generations (two parental and one sister) per year were peculiar to *I. typographus* populations. In the present study, *I. typographus* phenology was analysed using two models: i) “PHENIPS – A comprehensive phenology model of *Ips typographus* as a tool for hazard rating of bark beetle infestation”, developed at the University of Natural Resources and Life Sciences, Vienna, Austria (Baier et al. 2007, Forest Ecol. Manag.), and ii) “Calculation procedure for RITY – A phenology model of *Ips typographus*” developed at the Slovenian Forestry Institute, Ljubljana, Slovenia (Ogris 2020, MethodsX), based on the PHENIPS model. These phenology models are founded on the following indicators: lower developmental threshold, threshold of imago flight onset, sum of effective temperatures for full development of one generation and for each of the development stages and the number of generations per year. These parameters vary depending on the topoclimatic conditions of the region, including air temperatures and terrain traits.

In 2022, a pilot study, where the model “RITY-2” was tested employing the actual dataset of bark beetle monitoring in 2016 obtained using pheromone traps and meteorological data, was carried out. The study included employing nine pheromone traps on one testing ground. Since the obtained sample size was occurred to be too small, and no data was available on the timing of developmental stages from egg to imago in the same year (2016), consequently, it yielded just theoretical results. Subsequently, development of the bark beetle generations was modelled according to the 2016 daily air temperatures. All the developmental stages (egg, larvae, pupae, imago) of the bark beetle – the 1<sup>st</sup> parental brood (A), 1<sup>st</sup> sister brood (B), 2<sup>nd</sup> parental brood (C) and 2<sup>nd</sup> sister brood (D) – were modelled. According to the field monitoring data, the peak of imago flight of the 2<sup>nd</sup> sister brood was recorded from 25 July to 2 August; it corresponded to the modelled onset with an error of  $\pm 4$  days. Moreover, the results showed that the sum of effective temperatures (K) of 334.2 Ld is required for preimaginal development, and 222.8 Ld (i.e. two-thirds of the sum of effective temperatures of the preimaginal stage) is required for feeding of adults of the main generation. The sum of the effective temperatures of 557 Ld is required for the complete development. For successful wintering, the bark beetle must complete the stage of preimaginal development (egg-larva). For that, before the beginning of the cold period, 60% of the effective temperatures are required, from effective temperatures of the entire development cycle (i.e. 557 Ld). Phenology validation study continued in 2023.

Conclusions: i) RITY-2, based on the air temperature parameters (which are recalculated for the forest envi-

ronment), could be used for the predicting *I. typographus* distribution; ii) prophylactic pest control activities could be provided during the different development stages, i.e. timing of pheromone traps or trap trees installation in particular year and region; iii) theoretical model issues were confirmed by the more detailed observations (2022 and 2023), when 4 generations of *I. typographus* (2 parental and 2 sister) were found; iv) the 2<sup>nd</sup> sister brood fully completed its development cycle and will hibernate in the imago stage; v) the lower development threshold is 6°C instead of 8.3°C, therefore requiring the sum of 597 Ld effective

temperature instead of 557 Ld, showing better phenological adaptation in response to changing climate; vi) in 2023, *I. typographus* started its development 19 days earlier than in 2022, which, combined with a warm autumn, led to successful completion of the 2<sup>nd</sup> sister generation; vii) calibration of the phenology model requires the provision of data collected from more detailed field investigations.

**Keywords:** *Ips typographus*; phenological adaptation; climate change; *Picea abies*; ecological modelling; PHENIPS, RITY-2