





Gefördert durch:



Bundesministerium für Umwelt, Naturschutz und nukleare Sicherheit

aufgrund eines Beschlusses des Deutschen Bundestages



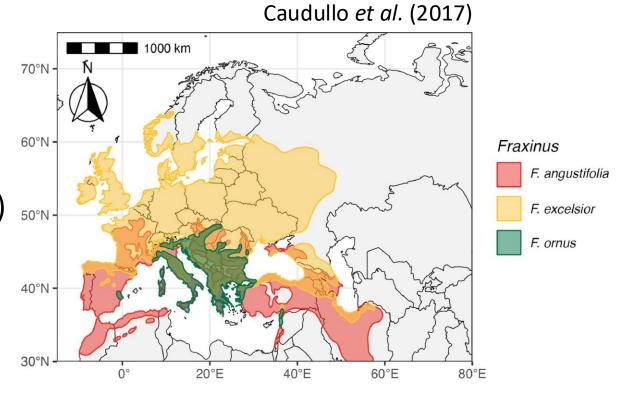




### Major ash species in Europe



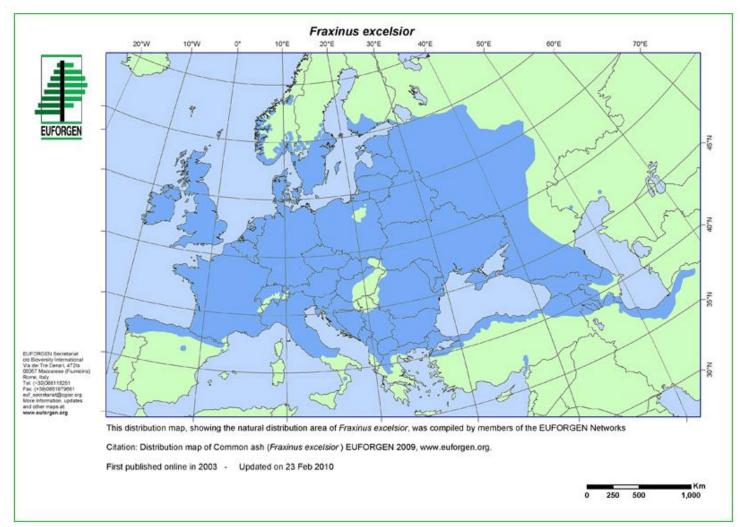
- Narrow-leaved ash (*F. angustifolia*)
- Manna/ flowering ash (F. ornus)
- Exotic green ash (F. pennsylvanica)



While the ranges of ash species overlap in Europe, F. excelsior is dominant.



### F. excelsior - an important tree in Europe





Source: Zelimir Borzan



### F. excelsior - an important tree in Europe



Source: Wikipedia

#### The European ash (Fraxinus excelsior)

The European ash is an important tree in woodland, hedgerows, riverbanks, commercial forestry and urban areas.

It was the second or third most frequent deciduous tree species in Central Europe., accounting for approx. 1% of the forests

In Germany, it is estimated that it comprised 2.4% of forest area (250,000 ha), or 74.7 million solid cubic meters (2001/2002).



### F. excelsior as a cultural legacy



Yggdrasil

In Norse mythology, ash was the tree of life, Yggdrasil, which harboured all the life in the universe. Moreover, the first human was created from ash wood by Oden.

- Often appears in old literature, artwork and poetry
- Historically used for weapons, tools, cart wheels...
- Popular ornamental tree (parks, gardens, hedges)







"Omens", which guided actions and decisions.



Hurley stick, hurling ball



### F. excelsior as an ecological species

- Ash trees are found in on marginal lands
  - Calcareous soils, floodplain forests and ravine forests
- Therefore, provides important ecosystem services
  - Especially in already compromised flood plain forests
- There are > 1000 known associated species
  - approx. 150 are obligate or highly-associated species
  - these species include fungi, invertebrates, lichens...
  - ... as well as wood mice, squirrels, birds and bats...
  - ... and the scarce fritillary (Euphydryas maturna)

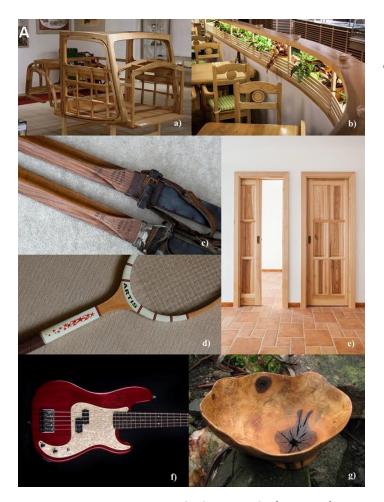


Euphydryas maturna Source: Wikipedia

Mitchell et al. (2014); Hultberg et al. (2020)



### F. excelsior as a commercial species



Source: Rozsypalek et al. (2017)

#### Tree characteristics

- Relatively fast growing
- High regeneration potential
- Drought resistant (climate change)

#### Wood as a commercial material

- Excellent wood properties (hard but elastic)
- Commonly used for tool handles, high quality furniture, sports equipment, musical instruments, veneer, flooring



### New devastating disease



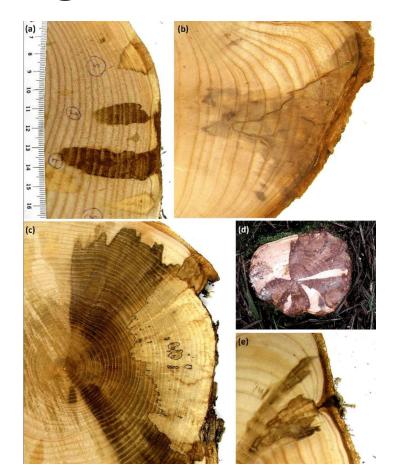


Images of crown defoliation and crown dieback in mature ash trees (Fraxinus excelsior)



### New devastating disease



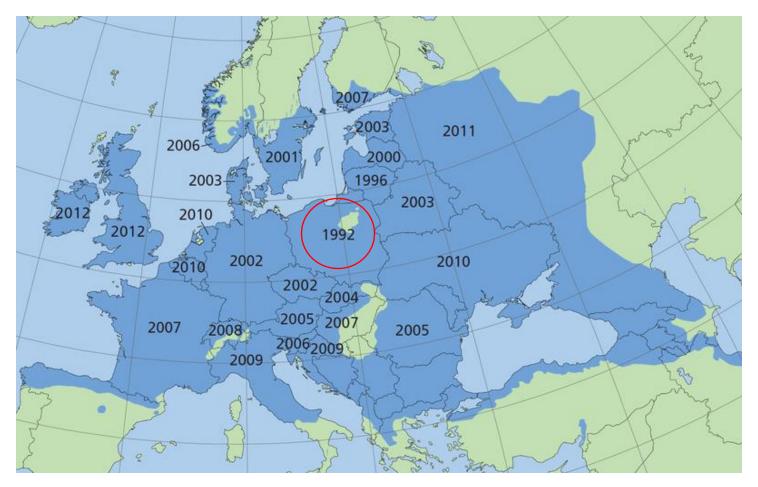




Necrotic lesion on stem of saplings (A, B); overview of differently developed collar necroses on investigated stem discs (a, b, c, d, e); and stem collar necrosis / rot (d)



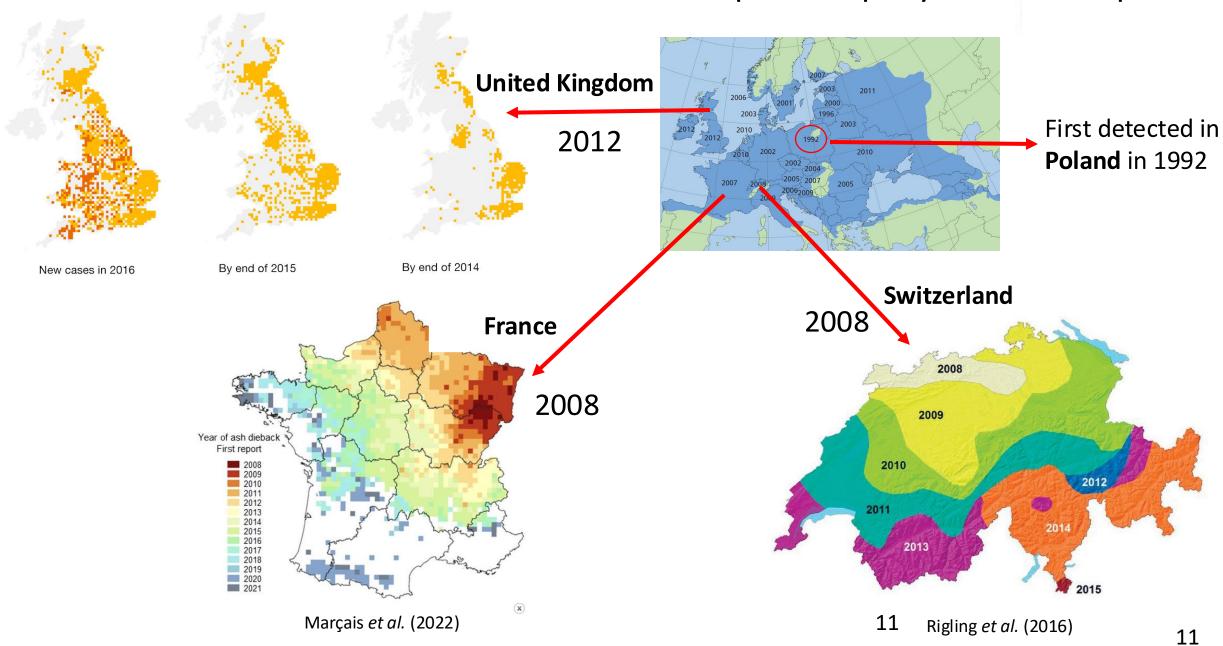
### Spread of Disease in Europe



The disease was first detected in Europe in 1992 in Poland. The year when ash dieback was observed is given for each country. In blue: The natural range of *F. excelsior* (© EUFORGEN).

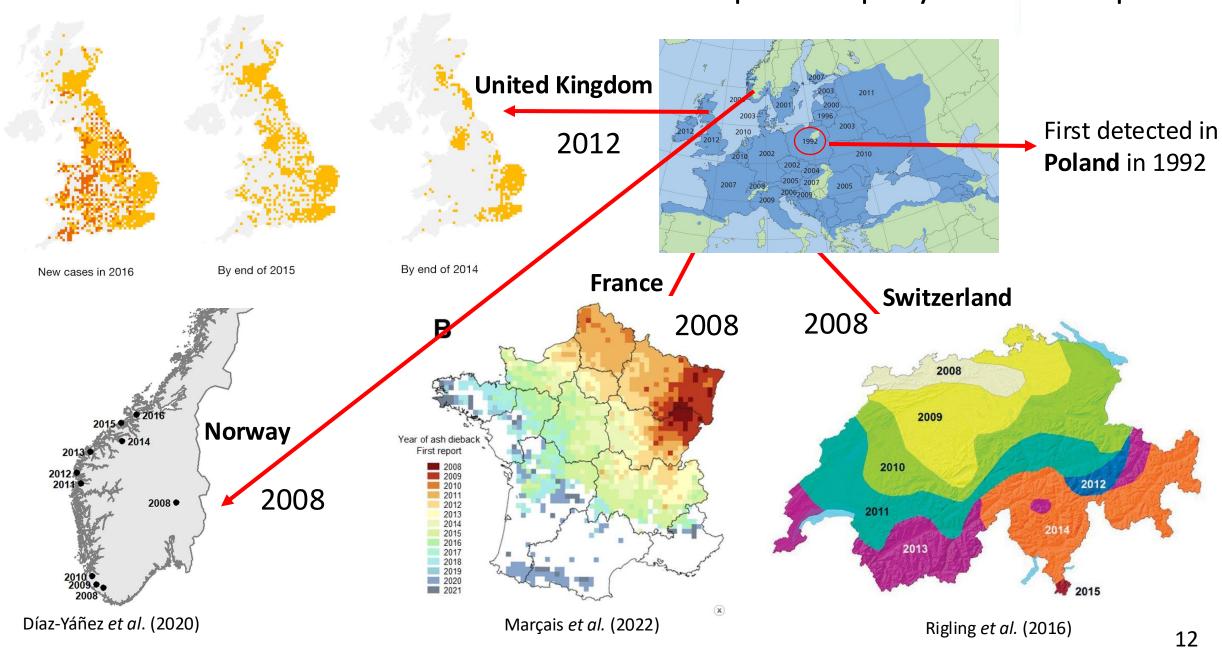
Guardian graphic. Source: Forestry Commission

#### The disease spread rapidly across Europe



Guardian graphic. Source: Forestry Commission

#### The disease spread rapidly across Europe





### What is causal agent of the disease?





• In 2006, the anamorph (asexual reproductive stage) is identified and named *Chalara fraxinea* sp. nov.

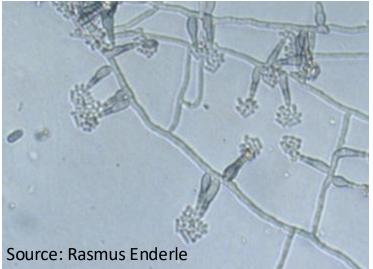
• In 2009, the teleomorph (sexual reproductive state) is tentatively identified as *Hymenoscyphus albidus*.

 However, H. albidus is widespread in Europe, had never been reported to be pathogenic in plants.



### What is causal agent of the disease?





- Then, reclassified as a cryptic species: *H. pseudoalbidus*
- H. pseudoalbidus exhibits a distinct morphology, mating system and is genetically divergent from H. albidus
  - -> H. pseudoalbidus is classified as a separate species
- In 2014, it was renamed as *Hymenoscyphus fraxineus*



### Introduction of an exotic fungus

Manchurian ash (F. mandshurica) is host to a saprotrophic fungus, called H. fraxineus



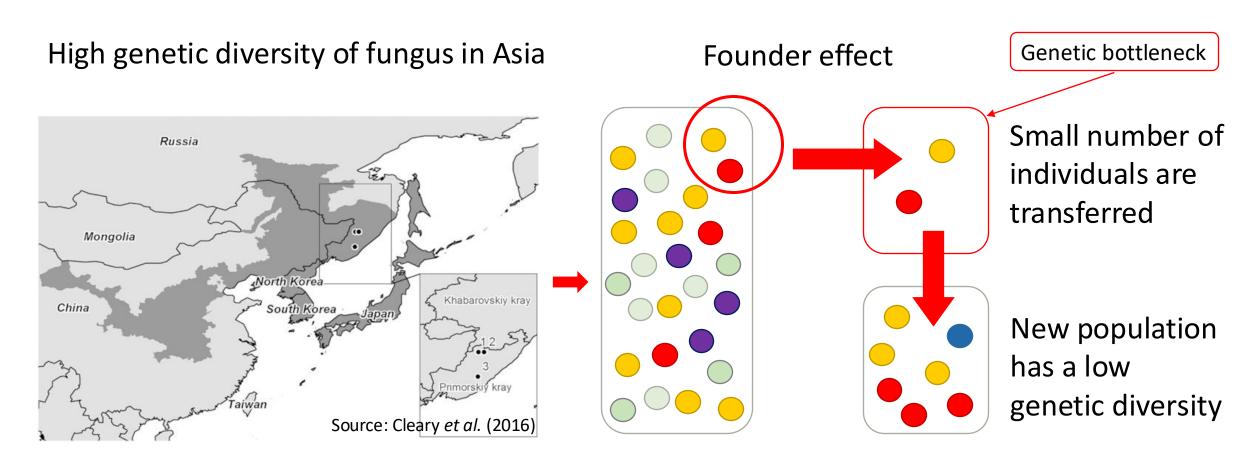
Natural distribution of *F. mandshurica* in Asia (see shaded area)



Manchurian ash (F. mandshurica)



### Introduction of an exotic fungus



The invasion was founded by two divergent individuals (McMullan et al. 2018)



### Introduction of an exotic fungus

#### **Globilisation: Plant trade**

#### Saprotrophic fungus



H. fraxineus



Host jump to naïve host

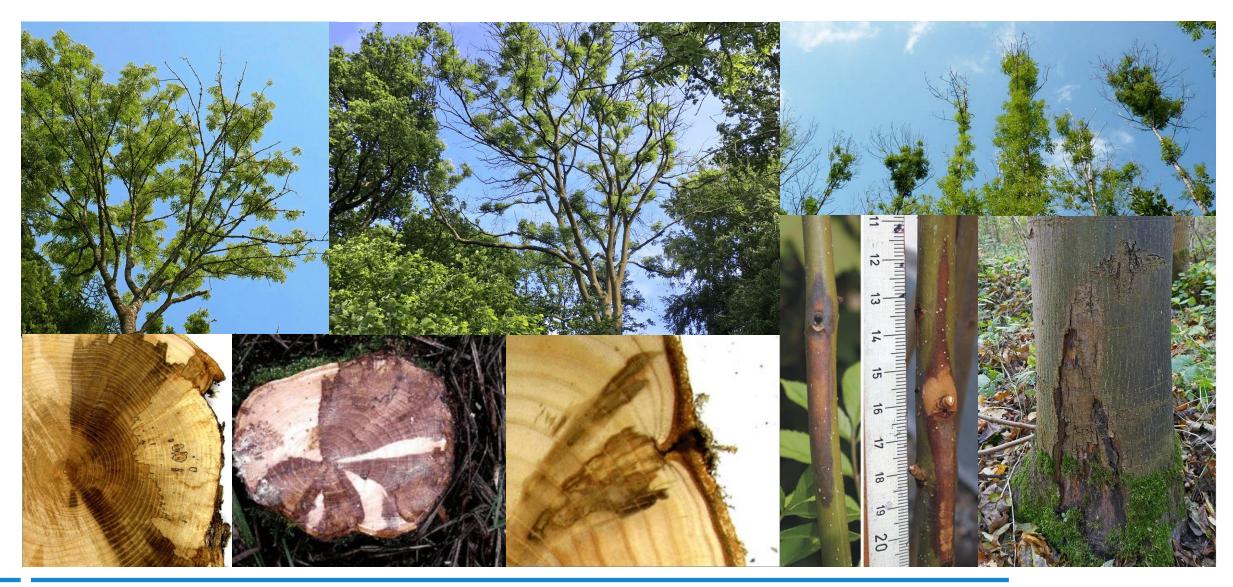


European ash (F. excelsior)

It is possibly introduced in **nursery stock of Manchurian ash** from Asia into Europe. Successfully colonises the European ash (*F. excelsior*), **a novel and naïve host in Europe**.



## Symptoms



Sources: Kirisits et al. (2012); Enderle et al. (2017); Langer et al. (2022); Andrin Gross; Lewis White

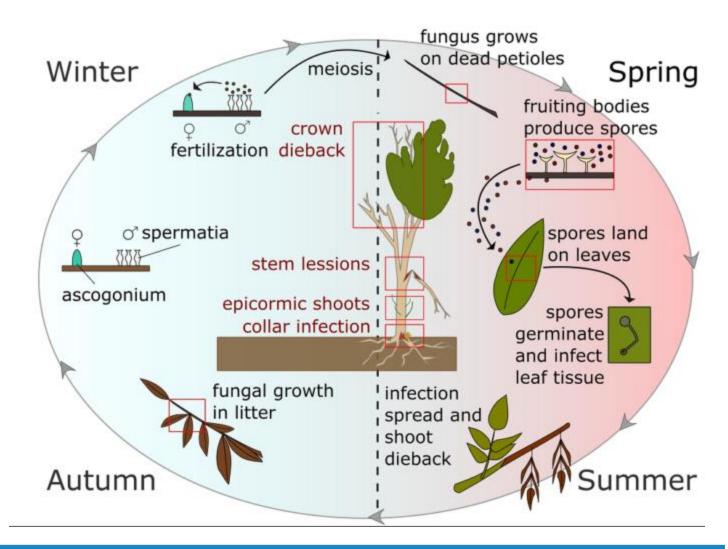


# lenticel leaflet stem rachis petiole

fruiting bodies on a fallen petiole

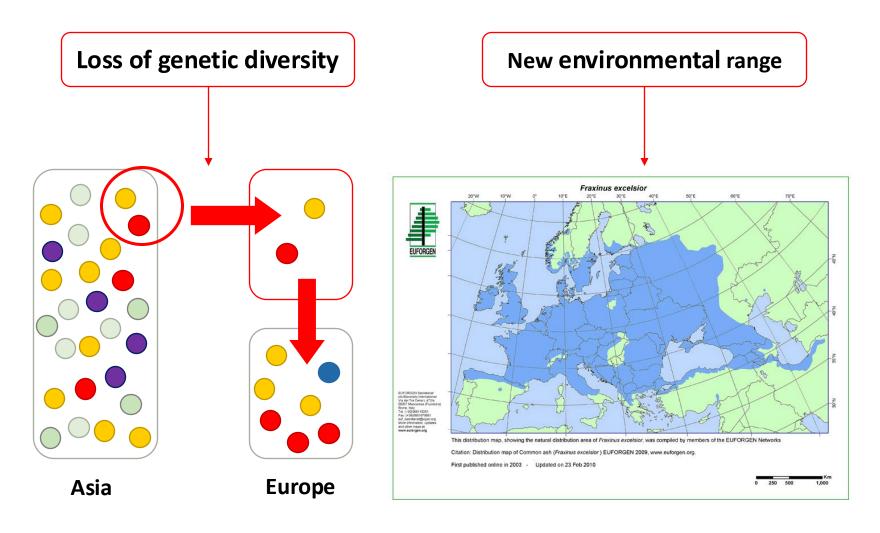


### Life cycle of *H. fraxineus*

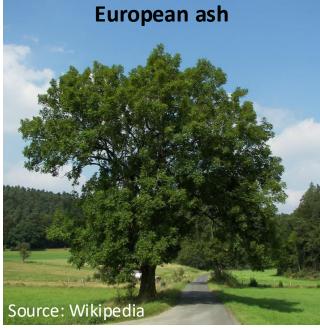




### Summary



Novel host species

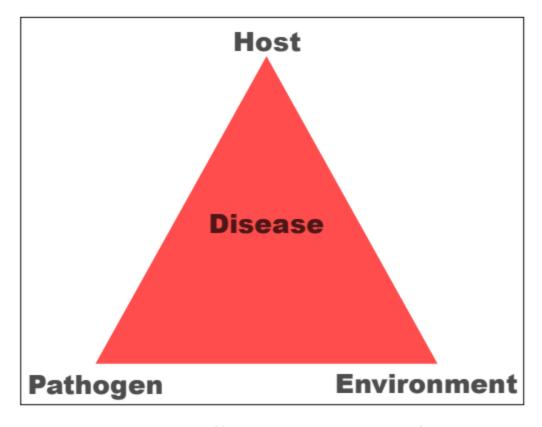




### The Disease Triangle

#### An epidemic requires:

- (i) a virulent pathogen with a high reproduction rate
  - -> H. fraxineus
- (ii) an accumulation of susceptible host
  - -> European ash
- (iii) suitable environmental conditions for the pathogen
  - -> Central Europe



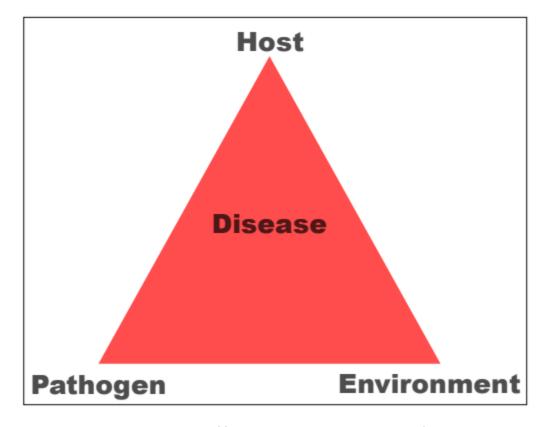
Source: https://www.ncforestservice.gov/



### The Disease Triangle

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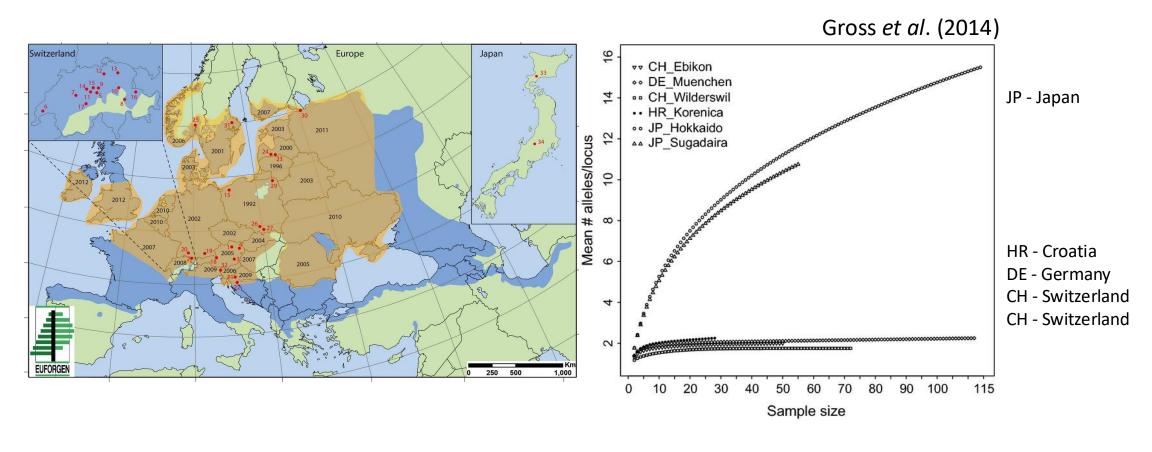
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Source: https://www.ncforestservice.gov/



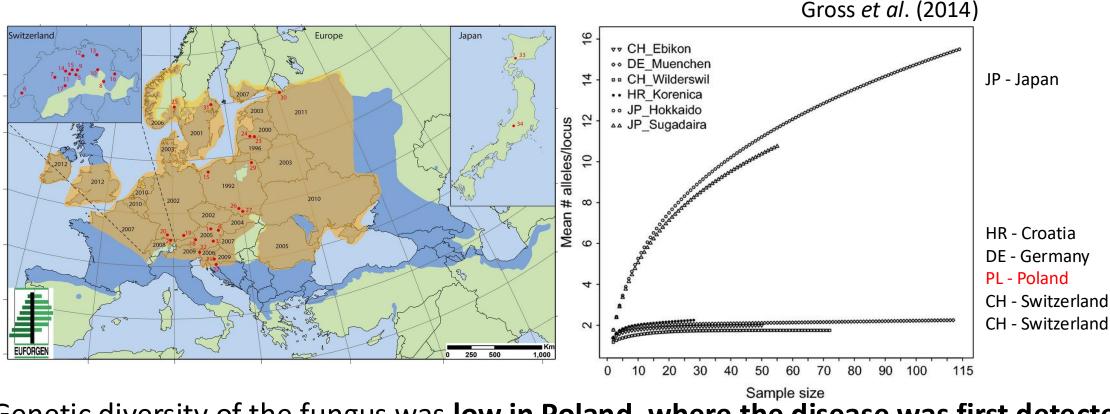
## Genetic diversity of *H. fraxineus*



Genetic diversity is higher in Japan than in Europe, as expected (Gross et al. 2014).



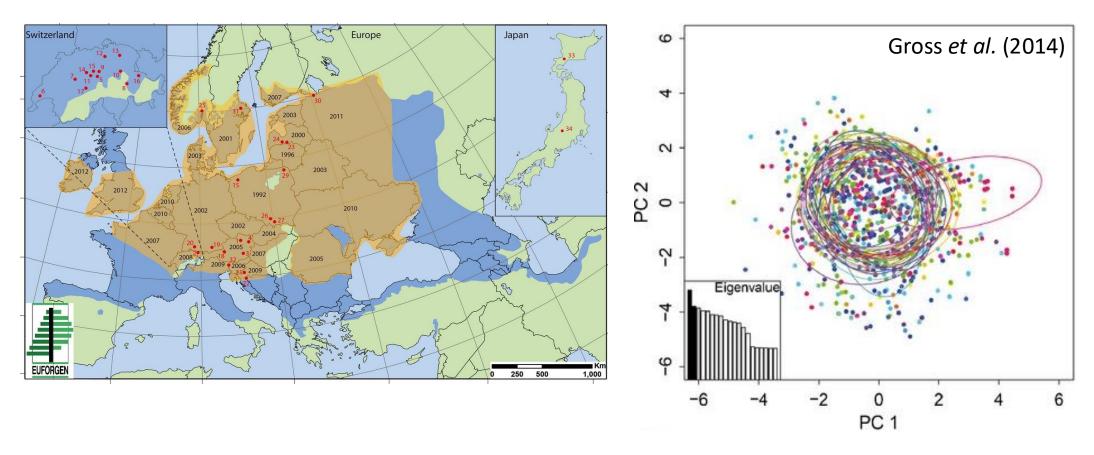
### Genetic diversity of *H. fraxineus*



- Genetic diversity of the fungus was low in Poland, where the disease was first detected
- From there, the genetic diversity increased slightly in a westwards direction
- But, it was also lower at the epidemic's front in Switzerland (CH).



## Genetic diversity of *H. fraxineus*

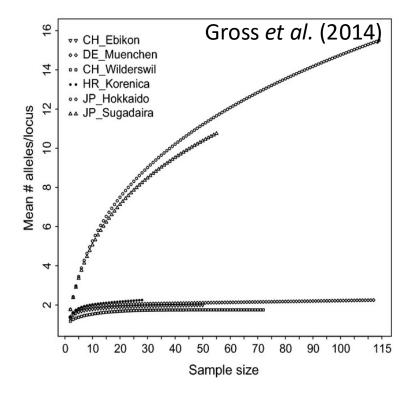


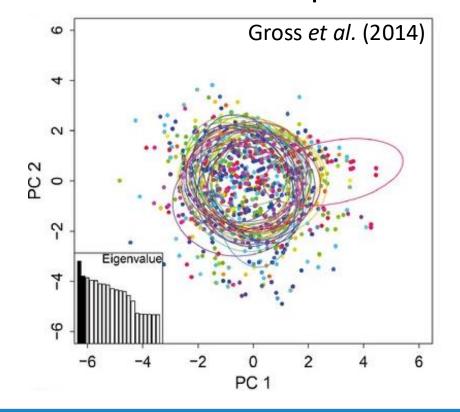
No evident population structure was found in Europe (Gross et al. 2014).



### Virulence of *H. fraxineus*

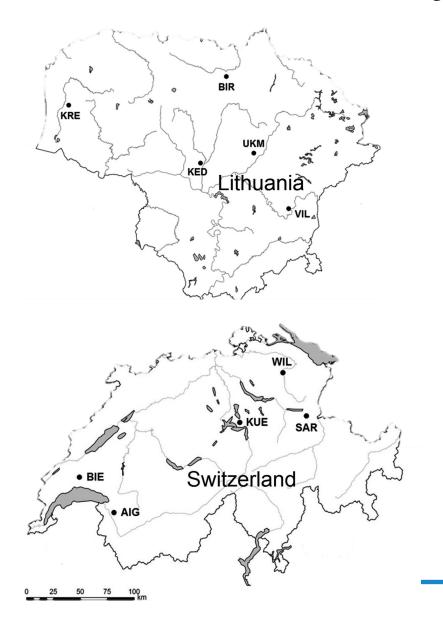
Despite low genetic variation and high frequency of genetic admixture, large differences in virulence are observed in infection experiments...

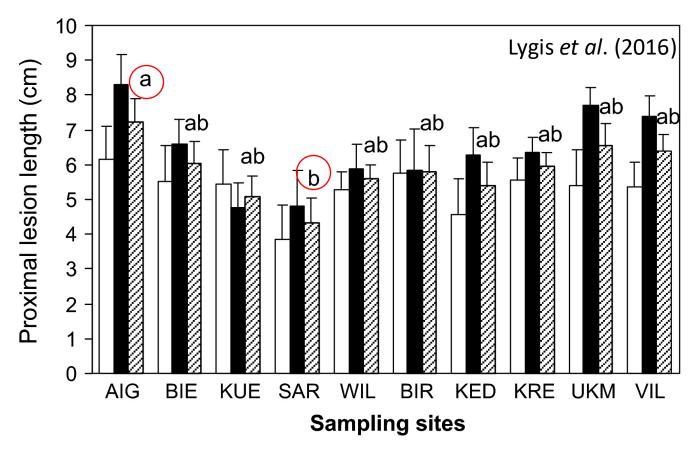




### –jKi

### Virulence of *H. fraxineus* in Europe

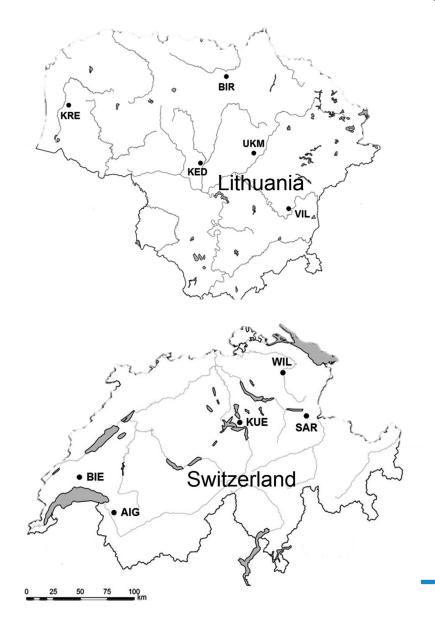


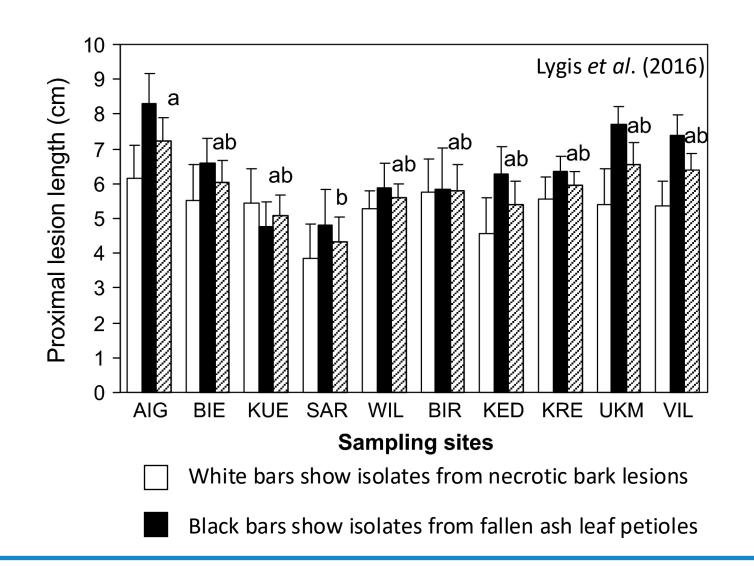


Hatched bars show the mean lesion length for all the isolates combined.

### -jKi

### Virulence of *H. fraxineus* in Europe





### Virulence of *H. fraxineus*



viridio

Many plant pathogens use toxins to attack host cells

- Viridiol has phytotoxic effects on ash seedlings
  - > However, high production is also observed in non-pathogenic *H. albidus*
- Polyphenoloxidases are involved in lignin degradation during infection
  - > However, the same is true for the non-pathogenic *H. albidus*

On susceptible exotic hosts the necrotrophic behaviour is seemingly accentuated.

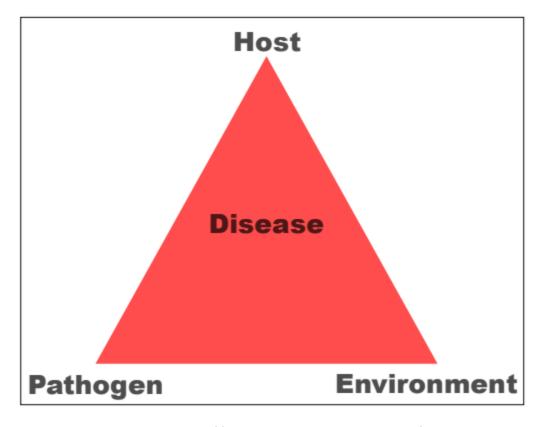
> The mechanism driving this shift in behaviour is still unknown.



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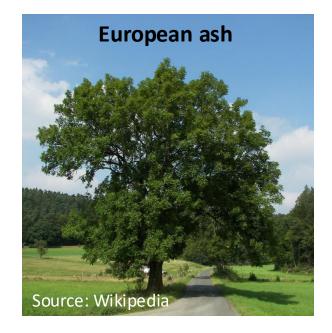
Source: https://www.ncforestservice.gov/

### Susceptibility of the host, F. excelsion

The European ash was the second or third most frequent deciduous tree species in Central Europe, accounting for approx. 1% of forests.

There is high susceptibility in European ash genotypes

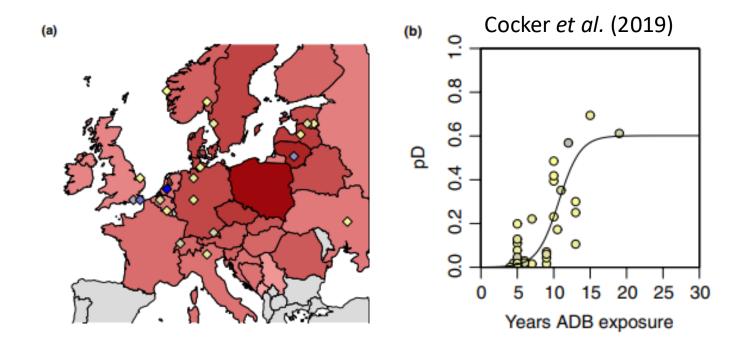
approx. 95% of trees are considered susceptible, showing significant symptoms and mortality





### Susceptibility of the host, F. excelsion

No site had 100% mortality, even after 20 years of disease presence.



- (a) Geographical distribution of woodlands (larger sampling marked blue, fewer trees in yellow).
- (b) Logistical model for mortality of woodland ash trees (pD, proportion of dead trees) over time.



# Susceptibility of Novel Host Tree Characteristics

Individual Tree Characteristics	Symptoms are more severe	
Host species	F. excelsior, compared to F. angustifolia	
Gender	male trees	
Age	younger trees	
Diameter at breast height (DBH)	trees with smaller DBH	Europe
Crown surface projection	trees with smaller crowns	Boy Alband

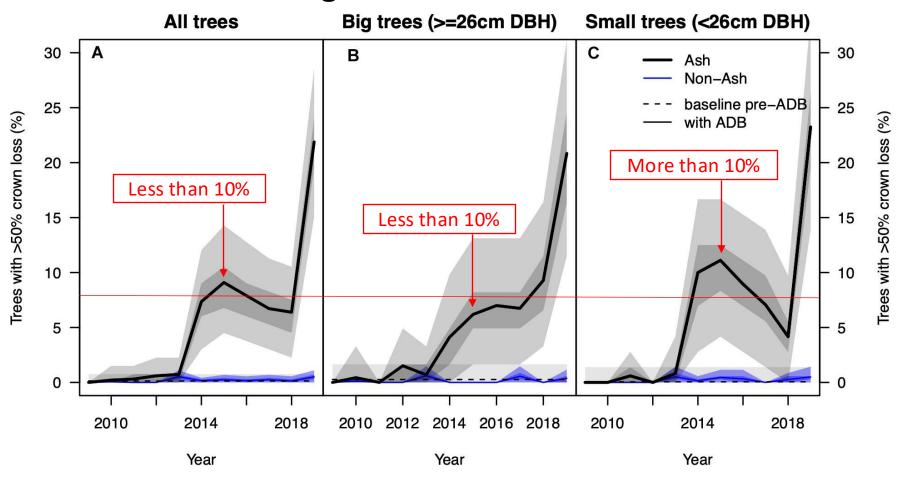
Certain tree characteristics are associated with more severe symptoms in European ash trees





## Case study in Switzerland

### Crown loss is higher in trees with a small DBH

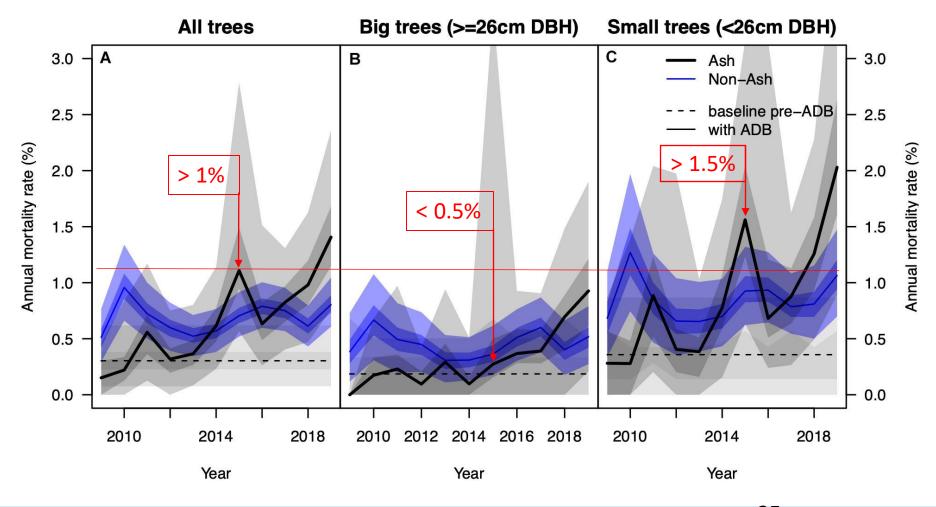


Klesse *et al.* (2021) 34



### Case study in Switzerland

#### Mortality is also higher in trees with a small DBH



Klesse *et al.* (2021) 35



# Susceptibility of Novel Host Tree Characteristics

Individual Tree Characteristics	Symptoms are more severe
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Crown surface projection	trees with smaller crowns  European ash

Symptoms are more severe
producing leaves later (flushing)
shedding leaves later





# Susceptibility of Novel Host Site and Environmental Conditions

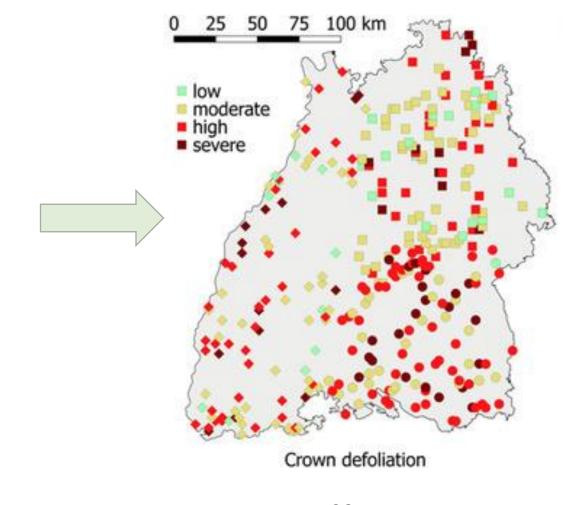
Site and Environmental Conditions	Symptoms are more severe
Stand species composition	pure ash stands, plantations
Stand composition (shaded vs. open)	shaded forested sites
Altitude	lower altitude
Temperature	low to moderate temperature
Moisture, humidity	more humid sites
Soil type	unspecified

Enderle *et al.* (2019)



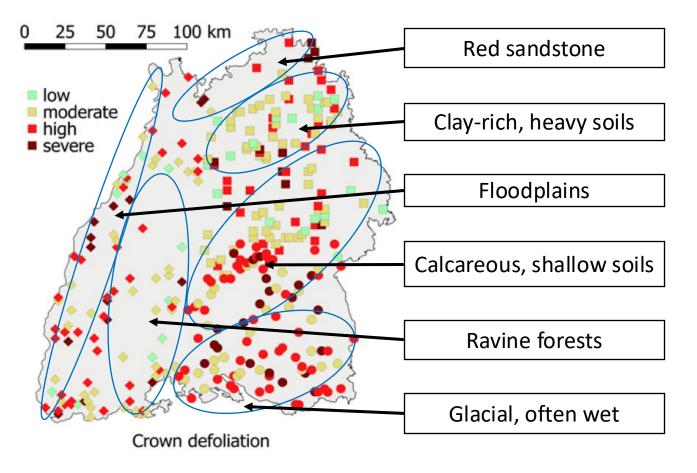
### Crown Defoliation







### Crown Defoliation

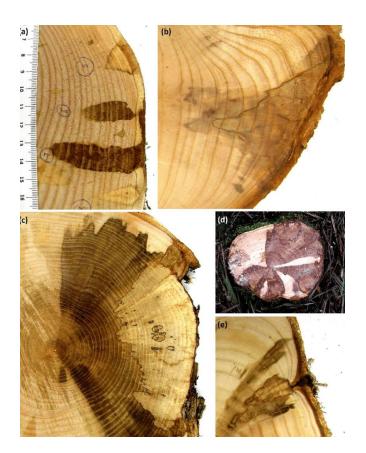


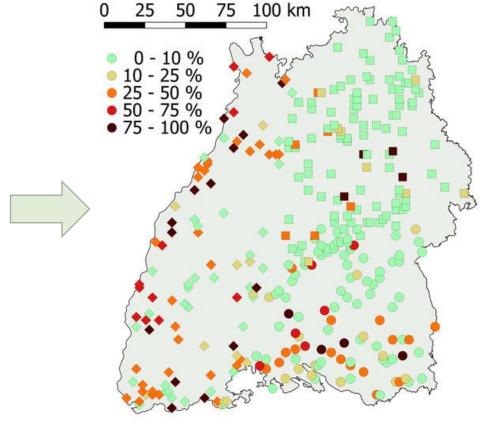
- Trees were **healthiest** in north-east
- Healthiest trees were on drier sites in north-east on heavy, clay-rich soils
- Severe defoliation was found on the floodplains or water-influenced sites
- The tested factors did not have a significant influence in this study

Enderle *et al.* (2018) 39

### Root Collar Necroses



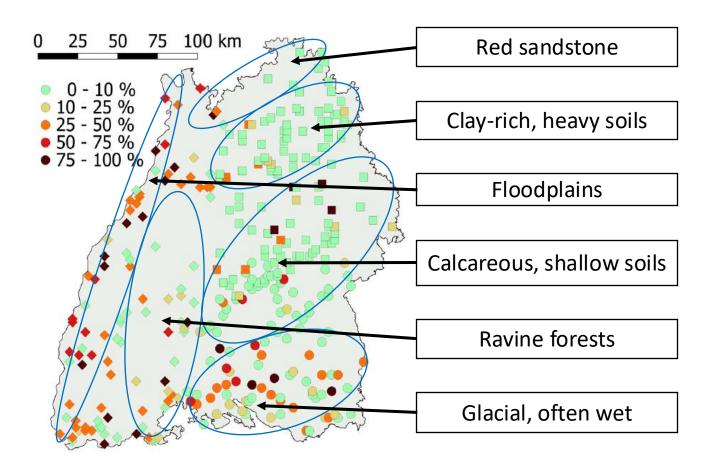




#### -jKi

41

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Enderle *et al.* (2018)



### Summary

Influencing factors

Symptoms are more severe...

Host species

Gender

Age

Diameter at breast height (DBH)

Crown surface projection

Phenology in spring

Leaf senescence/leaf shedding in autumn

Stand species composition

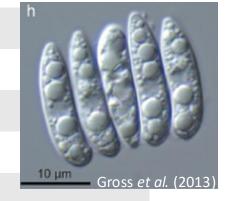
Stand composition (shaded vs. open)

Altitude

Temperature

Moisture, humidity

Soil type

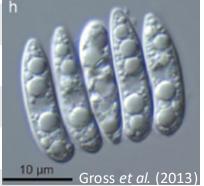






## Summary

Host species	F. excelsior, compared to F. angustifolia
Gender	male trees
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Moisture, humidity	more humid sites
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### Susceptibility to Secondary Colonisers

#### Armillaria spp., the honey fungus

Armillaria spp., is known to colonise necrosis caused by H. fraxineus

These secondary colonisers pose additional health and safety risks

Fruiting bodies of Armillaria spp. on mature ash tree





### Susceptibility to Secondary Colonisers

#### Hylesinus spp.

- ash trees infected by ADB are predisposed to Hylesinus spp.
  - > Hylesinus crenatus
  - > Hylesinus fraxini

Hylesinus cranatus



Hylesinus fraxini





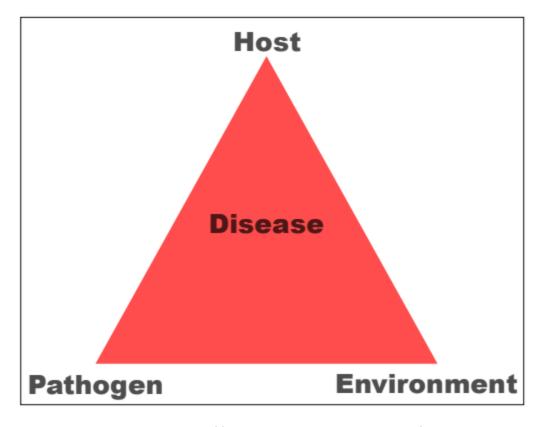




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Source: https://www.ncforestservice.gov/

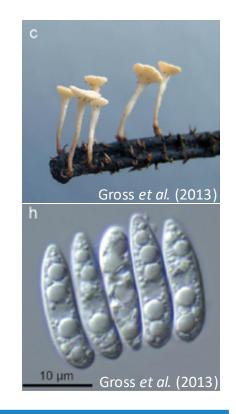
#### -jKi

### **Environmental Factors**

- 20 °C is optimum for mycelial growth in culture (ceases at 30 °C)
  - > In ash tissues the fungus appears to be more heat sensitive.

- Ascospores are produced abundantly during spring and summer
- Ascospore melanization after ejection and attachment to a host surface suggest ability to withstand adverse conditions *in-planta*.
- Ascospores are drought sensitive, spores can desiccate within 24 h

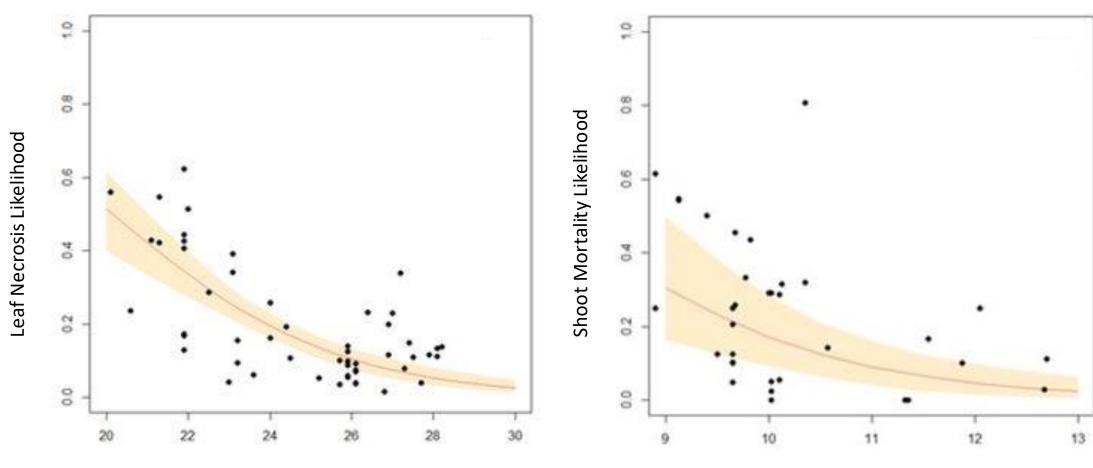




THE PATHOGEN THE HOST TREE THE ENVIRONMENT



### Climate change. Good News?



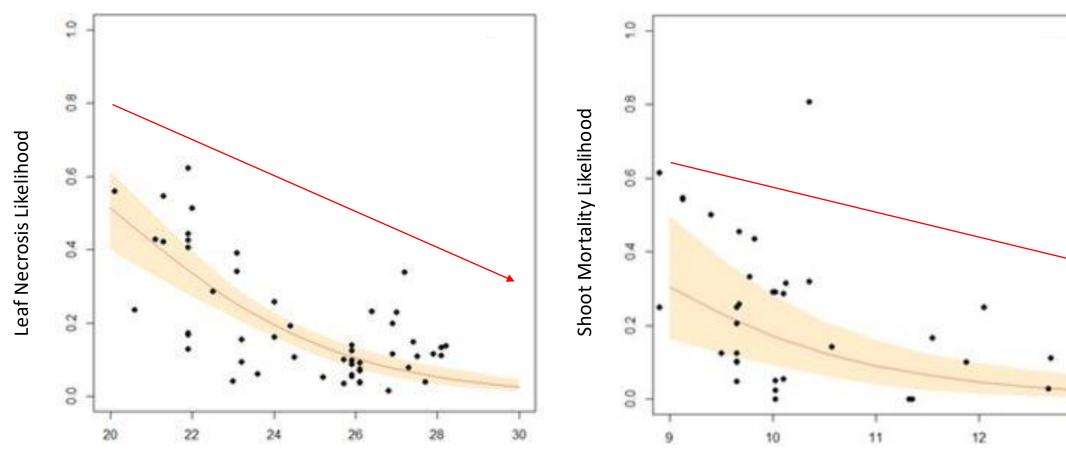
Average daily maximal temperature in July- August (°C)

Average temperature in September-December (°C)

Marçais *et al.* (2022) 48



### Climate change. Good News?



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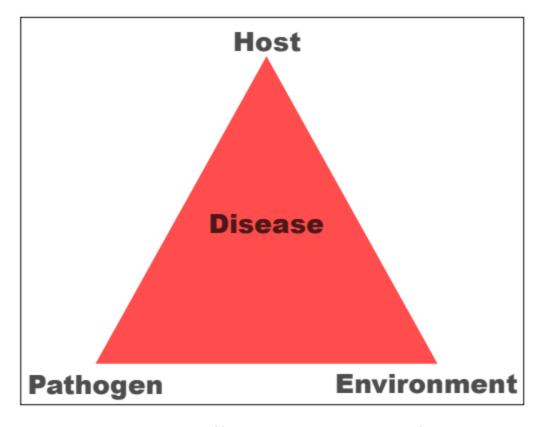
Marçais *et al.* (2022) 49



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Source: https://www.ncforestservice.gov/



### Disease management

We must control spread of the fungus to novel areas, specifically through trade





### Forest management

The relevant strategy depends on objective, site, stand age and damage

#### General recommendations:

- Maintain healthy or slightly damaged trees
- In forests of habitat conservation value, allow natural succession
- Minimise risks for operational staff, forest visitors and infrastructure
- Use other tree species to regenerate or replant for commercial uses
  - Acer, Betula, Fagus, Populus, Prunus, Quercus, Sorbus, Taxus, Tilia, or Ulmus spp.
  - a.k.a. Maple, Birch, Beech, Poplar, Cherry, Oak, Rowan, Yew, Lime/Linden, or Elm



### Forest management

FOREST MANAGEMENT

Why is it recommended to maintain also slightly damaged trees?

- The low resistance means ash populations will decline, which will leave only a few resistant trees to parent new generations of trees
- Healthy-looking trees are needed as future seed trees, thereby supporting the natural selection in favour of decreased susceptibility
- Despite long distance pollen flow, management will be necessary to prevent inbreeding in increasingly fragmented populations



### Resistance of European ash

TREE RESISTANCE

>approx. 5% of trees are considered resistant, showing no or only insignificant symptoms

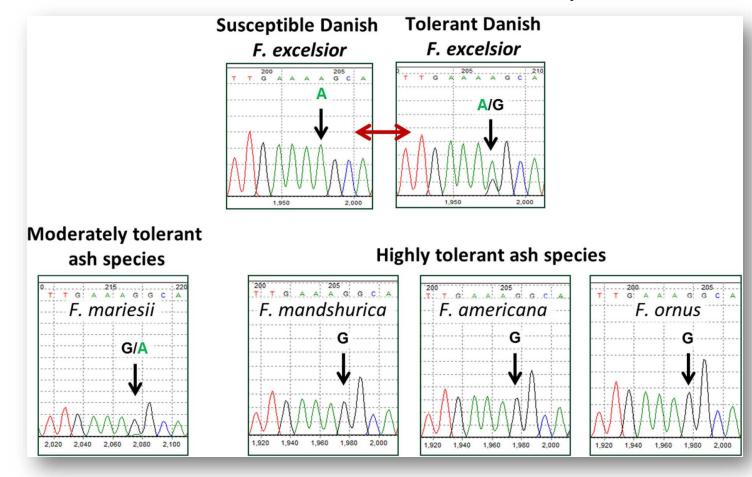
> However, this **tolerance is heritable**, which gives reason for hope for the recovery of *F. excelsior* 





### Resistance of European ash

Identification of resistance markers in Danish ash trees by associative transcriptomics



Sequence chromatograms showing the cSNP position in a number of Fraxinus species

Harper *et al.* (2016) 55

DREST MANAGEMENT TREE RESISTANCE NOVEL THREATS NOVEL APPROACHE

### Breeding for Resistance



Identifying and promoting genetic and heritable resistance in Europe  $\triangleright e.g.$  Austrian programme "Esche in Not" (ash in distress) from 2015-2024





Source: Rasmus Enderle 56

### Breeding for Resistance



Identifying and promoting genetic and heritable resistance in Europe  $\triangleright e.g.$  Austrian programme "Esche in Not" (ash in distress) from 2015-2024

However, breeding takes decades and resistance screening is labour intensive The establishment of seed orchards began in spring 2024 The first seed harvests are expected in approximately ten years

Source: Rasmus Enderle 57

The publication of the ash genome and subsequent genomic studies are helping



About us Why protect? Science Education

Work With Us

NEWS

30.07.2024

A new reference genome from a healthy ash tree

Our researchers have been busy creating a new genome resource

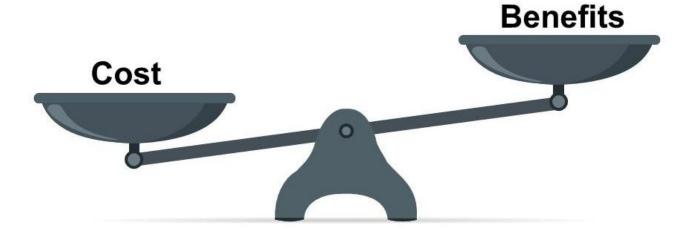
It is now possible to predict tree health with over 90% accuracy!



### Costs of Breeding for Resistance?

#### Possible trade-offs when selecting for host resistance:

- Growth?
- Competitiveness?
- Fecundity?



Source: https://www.slideteam.net/

OREST MANAGEMENT TREE RESISTANCE

### Costs of Breeding for Resistance?

#### Possible trade-offs when selecting for host resistance:

- Growth?
- Competitiveness?
- Fecundity?
- Susceptibility to novel threats?







### Emerald Ash Borer - Bad news!







Mankato Reception: Sat, Nov 12th at 3pm. 1700 block of Broad Street Blvd (south side of blvd) Rochester Reception: Sat, Nov 19th at 3pm. Soldiers Field Memorial Park (east side of running track)

Native to Asia, the Emerald Ash Borer was first found near Detroit in 2002. Since its arrival, it has killed hundreds of millions of ash trees throughout the Midwest and East. The larvae feed on the inner bark of the tree. This inhibits the tree from carrying water and nutrients to its living foliage. The adult beetles exit the trees in the spring and fly to new specimens to lay their eggs and repeat the cycle. In 2008 it arrived in Minnesota. Minnesota has some 900 million ash trees, more than any other state. As of 2016, the beetle has only been documented in the Twin Cities and Rochester. Humans transporting firewood is the most common means for the spread. Please do your part to promote awareness about this problem.

www.dnr.state.mn.us/invasives/terrestrialanimals/eab/index.html www.mda.state.mn.us/emeraldashborer





Josh K. Winkler is a fiscal year 2016 recipient of an Artist initiative grant from the Minnesota State Arts Board. This activity is made possible by the voters of Minnesota through a grant from the Minnesota State Arts Board, thanks to a legislative appropriation from the arts and culture heritage fund.





 The emerald ash borer is native from East Asia

 It is an insect that seriously threatens all Northern American *Fraxinus* species

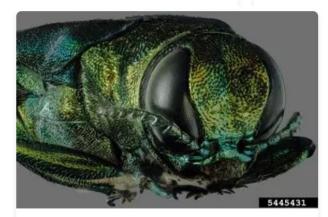


### Emerald Ash Borer - Bad news!



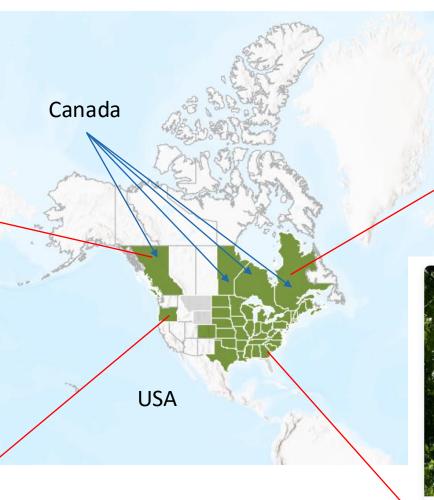
About EAB

The emerald ash borer and why it's a threat to ash trees worldwide.



#### Emerald Ash Borer Impacts on American Indian Communities

Emerald ash borer (EAB) infestation is a major concern for American Indian people. Many American Indian cultures and traditions rely on ash trees for the wood needed for making baskets, lacrosse sticks, pipe stems, flutes, and medicinal remedies. The ash tree is a central figure in some traditional and religious stories told by several American Indian tribes.







If You Find EAB

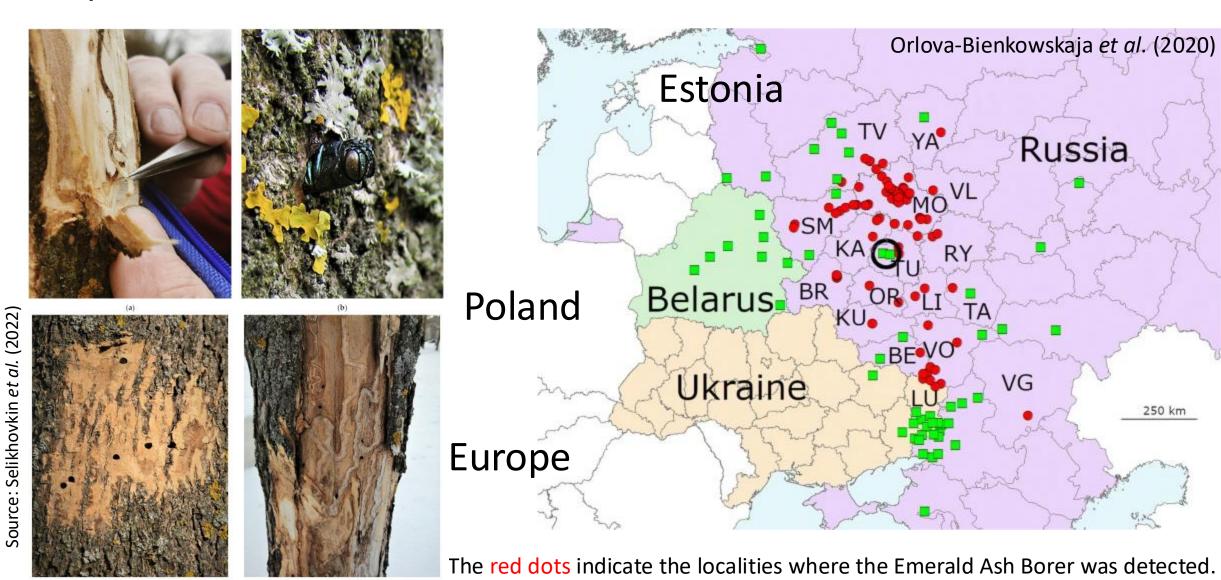
Where it is a particular state, contact resources in each state, regulations in each state, etc.)



Black Ash: A Foundational Wetland and Cultural Keystone Species

Learn about black ash ecology and the cultural signifigance of the species.

### Spread of the Emerald Ash Borer



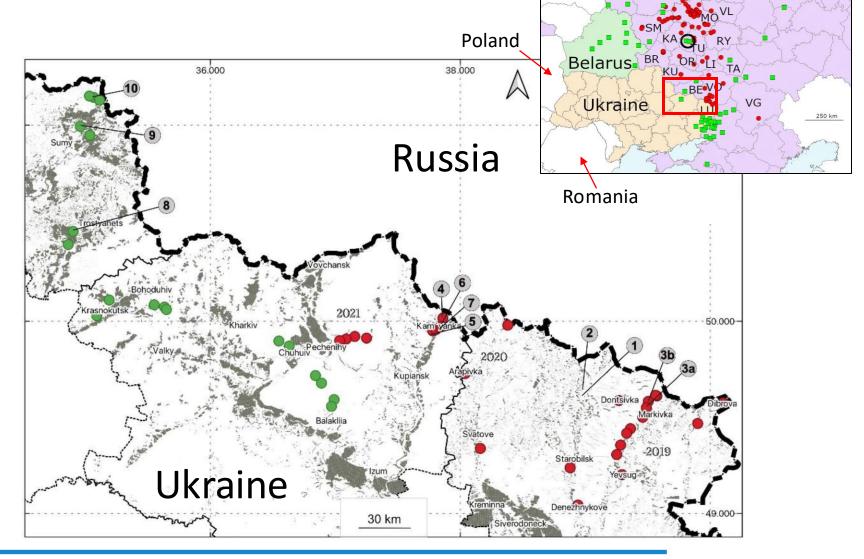
Estonia

Russia

Spread of the Emerald Ash Borer

Results from monitoring in 2021

The red dots indicate the localities where Emerald Ash Borer was detected in Ukraine.





# New Phytologist

Full paper 🙃 Open Access 💿 🕞 🕏



A glimmer of hope – ash genotypes with increased resistance to ash dieback pathogen show cross-resistance to emerald ash borer

Martin M. Gossner, Anouchka Perret-Gentil, Elisabeth Britt, Valentin Queloz, Gaétan Glauser, Tim Ladd, Amanda D. Roe, Michelle Cleary, Mateusz Liziniewicz, Lene R. Nielsen ... See all authors V

First published: 21 June 2023 https://doi.org/10.1111/nph.19068 | Citations: 1

 The ash genotypes with a higher ADB resistance also possess an increased resistance against EAB.

 These results are from Scandinavian and Swiss genotypes, with early and late instar larvae.

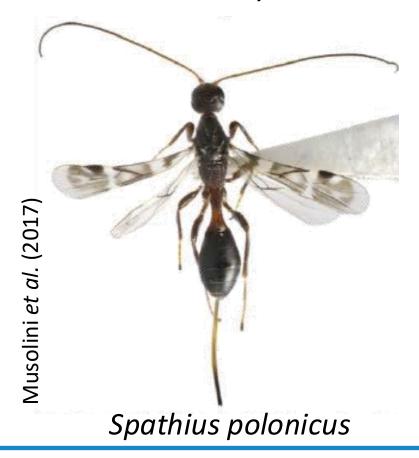


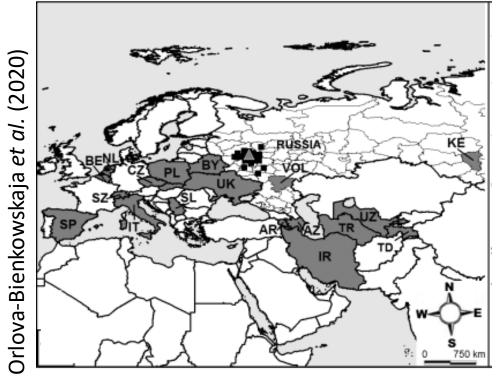


### Biocontrol for Emerald Ash Borer?

There is a known parasitoid (*Spathius polonicus*) of Emerald Ash Borer

> Has successfully induced 50% mortality of Emerald Ash Borer larvae in Moscow





Range of parasitoid (shaded areas)

#### -jKi

### Responses in research











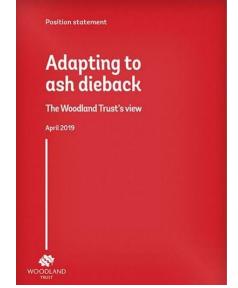
THE







PROJECT





### Responses in research





Jörg Grüner

(FVA-BW)

coordination

Network

Economic

analyses





Ben Bubner

(Thünen-Institut)

characterisation

Plus tree

selection

Genetic

Vegetative

propagation

application

Resistance marker:

Resistance marker:

development

architecture

Secondary

biomarkers

metabolism and

Genetic



Ralf Kätzel

(LFE)

Biomarkers

Resistance

indicators

Remote sensing:

interpretation key







Gitta Langer (NW-FVA)

Root collar necrosis: locational factors

Root collar necrosis: histology

Infection trials

Pathogen-host

interactions

Secondary

metabolites

Fungi in the Remote sensing: rhizosphere inventory design

Remote sensing: modelling

Hyperspectral imaging

Terrestrial monitoring

Histology

Ralf Nagel (NW-FVA)

Stand dynamics

Fundamentals in ecology

Susanne Jochner-Oette (KU Eichstätt)

Remote sensing: spectral signature

Virus diversity in ash

Molecular monitoring

Virus defence genes

- Demonstration project Intra-national cooperation
- Cross-disciplinary cooperation

#### Funding agencies:

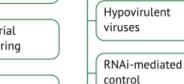
aufgrund eines Beschlusses des Deutschen Bundestages



für Umwelt, Naturschutz







Microbiota

#### -jKi

### Responses in research





Jörg Grüner (FVA-BW)

Network coordination

Economic analyses

- Demonstration project
- International cooperation
- Cross-disciplinary cooperation

#### Funding agencies:

Gefördert durch:



Bundesministerium für Umwelt, Naturschutz und nukleare Sicherheit







Ben Bubner (Thünen-Institut)

Plus tree selection

Genetic characterisation

Vegetative propagation

Resistance marker: application

Resistance marker: development

Genetic architecture

Secondary metabolism and biomarkers



Ralf Kätzel (LFE)

Biomarkers

Resistance indicators

Remote sensing: interpretation key

Remote sensing: inventory design

Remote sensing: modelling

Hyperspectral imaging

Terrestrial monitoring

Histology



Gitta Langer (NW-FVA)

Root collar necrosis: locational factors

Root collar necrosis: histology

Infection trials

Fungi in the rhizosphere

Pathogen-host interactions

Secondary metabolites

Hypovirulent viruses

RNAi-mediated control

Microbiota



Ralf Nagel

(NW-FVA)

Stand dynamics

Fundamentals in

ecology

Susanne Jochner-Oette

Susanne Jochner-Oette (KU Eichstätt)

Remote sensing: spectral signature

Virus diversity in ash

Molecular monitoring

Virus defence genes



Bundesforschungsinstitut für Kulturpflanzen Federal Research Centre for Cultivated Plants





### Infection trials with *H. fraxineus*











## Infection trials with *H. fraxineus*

Symptom development in infected saplings e.g. wilting









### Viruses

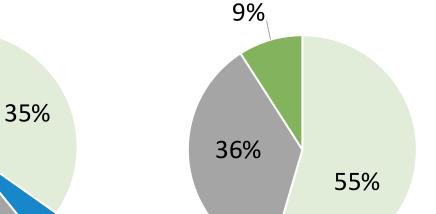




#### HfMV1 positive

9%\_

52%



0%

HfMV1 negative

■ not virulent ■ mid virulence

4%

■ low virulence ■ high virulence

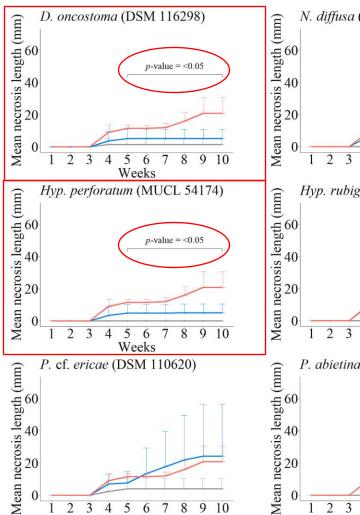
- Mycoviruses: HfMV1 and HaVV1
- HfMV1 showed no signs of causing hypovirulence (Lutz et al. 2024)
- HaVV1 still needs to be tested

### Endophytes

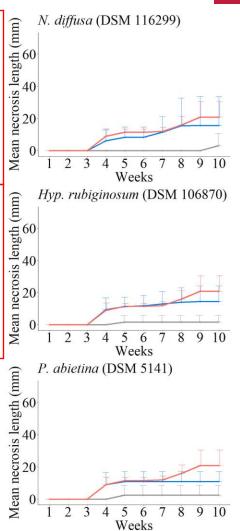








Weeks



- Six fungal endophytes were selected
- Two significantly reduced necrosis length under greenhouse conditions
- More tests needed, including field trials and assessment of synergies

### Priming

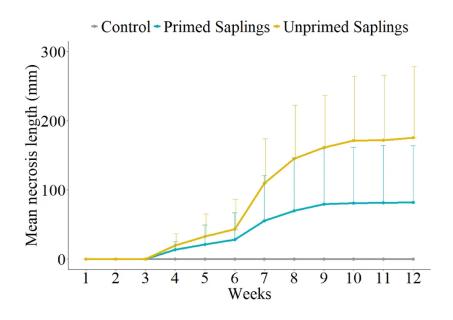






The primed state can be described as:

"an augmented activated defence response towards a pathogen" Ridley et al. (2025)



- Priming is an established method in agriculture
- On average, necrosis expansion was reduced by 53% in primed saplings
- More studies necessary, including to assess the long-term effectiveness



### Responses in research







FraxForFuture 2020-2024

FraxForFuture 2 2025-2028

#### -jKi

### Conclusion

- European ash is threatened by *H. fraxineus* 
  - > socio-culturally and economically impacts
- There are many factors that can increase susceptibility
- Forest management and tree breeding can reduce losses and damages
- However, we must prevent future introductions of the fungus
- We must also be aware of climate change impacts and novel threats e.g. EAB



Lewis White



#### –jKi

### References for further reading

- Ridley et al. (2025) Fungal endophytes with anti-fungal metabolites reduce symptoms of ash dieback in *Fraxinus* excelsior in a greenhouse experiment. DOI: <a href="https://10.1016/j.funbio.2025.101646">https://10.1016/j.funbio.2025.101646</a>
- Ridley et al. (2025) Priming of ash saplings with a low virulent Hymenoscyphus fraxineus strain as a possible disease control approach for reducing symptoms of ash dieback. DOI: <a href="https://10.1007/s41348-024-01056-z">https://10.1007/s41348-024-01056-z</a>
- Lutz et al. (2024) Identification of viruses for biocontrol of the ash dieback disease. DOI: <a href="https://10.1007/s41348-023-00804-x">https://10.1007/s41348-023-00804-x</a>
- Langer et al. (2022) FraxForFuture—research on European ash dieback in Germany. DOI: https://doi.org/10.1007/s41348-022-00670-z
- Marcais et al. (2022) Chapter 13 Ash dieback. Volume 2: Forest Tree Health. DOI: <a href="https://doi.org/10.1016/B978-0-323-85042-1.00022-7">https://doi.org/10.1016/B978-0-323-85042-1.00022-7</a>
- Vasaitis & Enderle (ed.) (2017) Dieback of European Ash (Fraxinus spp.) Consequences and Guidelines for Sustainable Management
- Enderle & Vasaitis (ed.) (2017) Advances in Ash Dieback Research, And Some Other Invasive Diseases Of Trees
- Gross *et al.* (2013) *Hymenoscyphus pseudoalbidus*, the causal agent of European ash dieback. DOI: <a href="https://doi.org/10.1111/mpp.12073">https://doi.org/10.1111/mpp.12073</a>