Geographically and genetically distinct populations of scots pine (*Pinus sylvestris*) do not differ in resistance to damage the large pine weevil (*Hylobius abietis*), a common garden translocation study

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Abstract

Damage to coniferous plantation crops from the large pine weevil Hylobius abietis causes economic losses of $\in 140$ million in Europe per annum. Current mitigation strategies are labour intensive and only partially effective. Natural resistance of host plants to insect pests has been used in many crop species to reduce damage as part of an integrated pest management strategy. Here, we conducted a common garden experiment in an area known to have pine weevils, with seedlings collected from 21 naturally occurring populations of Pinus sylvestris across Scotland

3 Introduction

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The large pine weevil (Hylobius abietis L. Coleoptera: Curculionidae) is a common pest of newly planted conifer plantations in Europe, generally causing damage to saplings up to five years old (?). Adult weevils 15 emerge from tree stumps and feed on the bark and buds of coniferious saplings. Circular lesions on the bark 16 and buds of saplings as a result of feeding may cause a reduction in growth rate, stem deformation and an 17 increased susceptibility to infection by airborne diseases of trees (?). While pine weevils may inhabit adult 18 coniferous trees in both natural and planted coniferous forests, recently clearfelled and restocked coniferous forest plantation sites provide enriched habitat for breeding pine weevils. Adults lay eggs within the numerous 20 stumps of clearfelled trees. with newly emerged juvenile weevils feeding on young saplings planted for restocking, 21 causing damage and mortality. A single adult weevil can damage several plants over the course of a season, with 22 50% sapling mortality observed across infected plantation sites in the UK and Ireland (?). On commercial 23 conifer plantations, H. abietis causes annual economic losses of €140 million per annum in Europe, of which $\in 2.75$ million occurs in the UK (?). The potential for climate change to enhance the damage caused by H. abietis, by reducing life cycle length () and encouraging migration into previously weevil free areas (), has prompted discussion of the effectiveness of current management practices and possible alternatives (). 27

Management of *H. abietis* currently relies on a variety of chemical, biological and physical measures, with integrated pest management schemes tending to yield greater success (). Physical deterrents include piling felling debris over exposed stumps to reduce egg laying, or stump removal. The application of entomopathogenic nematodes after clearfelling has been shown to reduce the number of adult weevils in clearfelled site (?). The most common method of control is the addition of chemicals at the time of restocking, with *H. abietis* being the only insect pest against which routine chemical controls are applied in the UK and Ireland (?).

Scots pine (*Pinus sylvestris* L.) is one of the UK's three endemic coniferous tree species. It constitutes

³⁵ percentage of the UK's commercial plantation forestry by biomass, while natural populations are restricted to enclaves in the north of Scotland. Caledonian remanents. Pine weevils attack scots pine plantations

Rather than the application of costly pesticides or biological control agents, breeding weevil resistant *P. sylvestris* varieties, or identifying existing genetically distinct population of *P. sylvestris* for commercial plantation forestry may provide a low cost method of reducing economic losses from weevil damage.

other crops where natural resistance has worked

Here, we conducted a common garden experiment to assess the resistance of *P. sylvestris* saplings grown from seed collected in populations across the natural longitudinal range of Caledonian Pine to damage from the large pine weevil *H. abietis*.

44 Materials & Methods

45 Study species

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- Scots pine (*Pinus sylvestris*) is the most widely distributed pine species in the world. It's range spans Eurasia from the arctic circle in Scandinavia to the dry northern mediterranean in Spain and Turkey and to the eastern edge of Siberia (?).
- Scotland represents the western limit of its distribution, where it is the dominant canopy tree species of the Caledonian pine forest. Scots pine grows well under low grazing, shade and competition Matyas 2004.
- Scots pine is wind pollinated, with monoecious flowering beginning between the ages of 15 and 30.
- Previous studies have identified genetically distinct populations of Scots pine in Scotland, with populations becoming more genetically isolated over distance as you move further west, against the prevailing wind direction ().

55 Study sites

Sites were situated within the range of the historical range extent of the Caledonian pine forest. Sites were chosen by accessibility and to ensure geographical isolation. Previous studies have shown cryptic genetic variation between these sites (?) which supports the assertion that despite strong cross-pollination effects between the populations, some degree of genetic isolation occurs. Variation in isolatedness between sites follows a predictable longitudinal gradient, with sites on the western extreme of the Caledonian pine range being more isolated due to the prevailing West-¿East wind direction.

62 Experimental design

- 63 Seedlings were grown for YEARS in glass houses in CONDITIONS before being planted in the common garden.
- The common garden was located at COORDS in a SIZE patch of recently clear-felled sitka spruce plantation, surrounded by existing adult sitka spruce (*Picea sitchensis*) plantation on all sides. All sitka spruce surrounding the common garden was planted at the same time WHEN. Planting was divided into four blocks of equal size

running perpendicular to the average slope of the site. Each block contained 167 saplings, with saplings placed in a grid pattern within each block, with a gap between each sapling of METRES and between saplings and the plot edge FIGURE REF. Saplings were randomly assigned to grid points.

Pine weevil infestation occurred naturally across the site, with adult weevils likely travelling from the adult sitks spruce plantation adjacent to the site.

Pine weevil lesions were counted on each sapling stem between DATES. This is roughly between the two seasonal peaks of weevil feeding that are commonly observed in the UK. Isolated lesions tended to be roughly circular with a diameter of ~3 mm. Where a larger continuous legion, as when a stem was girdled, the larger legion was photographed with a scale and the area estimated by tracing the lesion with Imagej version 1.50g7 (?). Weevil damage is therefore expressed as the mm² area of stem lesions per sapling.

77 Statistical analysis

- To isolate the effects of sapling genetic origin resistance to damage by pine weevils, we conducted generalised least squares linear models using the 'nlme' package (?) in R version 3.4.2 (?). Due to many saplings receiving no weevil damage, zero-inflated data was transformed by adding 1 to all measurements and log transforming.
- 81 Results
- 82 Discussion
- 83 Conclusion