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Incidence and impact of root infection by *Heterobasidion* spp., and the justification for preventative silvicultural measures on Scots pine trees: A case study in southern Sweden

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ABSTRACT

The distribution of *Heterobasidion* spp. infection in the root system of Scots pine (*Pinus sylvestris*) and the relationship between disease severity and growth was investigated in a mid-rotation in southern Sweden stand nine years after thinning. Twenty-four trees were mechanically uprooted to measure whole root systems and determine the percentage of infected root volume. Annual volume increment was retrospectively calculated using discs cut along the stem. No trees showed aboveground symptoms of infection, however the disease incidence belowground was 87.5% and the percentage of infected root volume ranged between 0% and 32%. The percentage of infected root volume was negatively correlated with the difference in volume increment between the last two adjacent five-year periods, indicating reduced growth in more infected trees, but not with other tree-specific growth characteristics such as diameter at breast height, tree volume or root volume. Annual volume increment of individual trees decreased with increasing percentage of infected root volume. The high incidence of *Heterobasidion* spp. and reduced volume growth in seemingly healthy Scots pine warrants preventative stump treatment during thinnings to minimize the establishment of *Heterobasidion*, especially in first rotation forests.

1. Introduction

Species of *Heterobasidion* are important fungal pathogens of northern temperate and boreal forests causing root and butt rot in conifers, damage to merchantable wood and severe economic losses (Bendz-Hellgren et al., 1998). *Heterobasidion* spp. infect freshly cut stumps and wounds with air-borne basidiospores (Rishbeth, 1951a; Isomäki and Kallio, 1974), and fungal mycelia subsequently spreads to neighboring trees through root contacts and grafts (Rishbeth, 1951b). High incidence of *Heterobasidion* spp. usually occurs on dry, sandy soils with high pH and low organic matter content, especially in stands occupying previously arable land (Rishbeth, 1951b; Alexander et al., 1975; Stenlid and Redfern, 1998; Redfern et al., 2010).

Two species of *Heterobasidion* occur in Sweden: *Heterobasidion annosum* sensu stricto (Fr.) Bref. and *Heterobasidion parviporum* Niemelä & Korhonen. Scots pine (*Pinus sylvestris* L.) is highly susceptible to infection by *H. annosum* s.s. (Laine, 1976; Gibbs et al., 2002). Seedlings and young trees may also be attacked by *H. parviporum* (Korhonen, 1978). Infection in Scots pine trees may lead to reduced volume growth and tree mortality (Burdekin, 1972; Gibbs et al., 2002). However, it is difficult to identify infected Scots pine trees in practice since the fungus is more frequently confined to the roots and decay in root tissue is seldom visible at the stump surface (Bendz-Hellgren et al., 1998). In addition, diseased trees may survive for decades without showing obvious crown symptoms (Greig, 1998). Previous studies suggest that crown symptoms and the presence of basidiocarps alone greatly underestimate the actual disease incidence (Bradford et al., 1978b; Kurkela, 2002; Rönnberg et al., 2006). Despite earlier reports of the damage on Scots pine, the effect from *Heterobasidion* spp. is usually overlooked by Swedish forest owners (Rönnberg et al., 2006). Stump treatment against primary infection by *Heterobasidion* spp. with the biological control agent *Phlebiopsis gigantea* (Fr.) Jül (Rishbeth, 1963; Korhonen et al., 1993), a common practice in Swedish forestry on Norway spruce, is hence generally not conducted on Scots pine (Thor, 2001). Knowledge on the losses caused by *Heterobasidion* spp. on this host species is thus needed to justify preventative treatments, and to optimize forest management through modeling efforts.

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Losses caused by *Heterobasidion* spp. have been estimated for different coniferous species using various methods. Burdekin (1972) found that timber production in Scots pine was reduced up to 40% through tree mortality and loss in volume growth caused by *Heterobasidion* spp., as compared to standard yield tables. By examining the incidence of *Heterobasidion* spp. from increment cores at stump height, Bendz-Hellgren and Stenlid (1997) estimated that diseased Norway spruce trees in southern Sweden lost up to 10% of volume growth in 20 years. In eastern United States of America (USA), Bradford et al. (1978a) examined the root systems of loblolly pine (*Pinus taeda* L.) and suggested that volume growth in trees infected by *Heterobasidion* spp. was reduced by up to 19% during a 5-year period. In general, more intensive sampling techniques, i.e. below-ground excavations, reveal more infection (Alexander and Skelly, 1974; Bradford et al., 1978b). The intensive sampling of root systems in Douglas-fir (*Pseudotsuga menziesii* var. *glauca* (Beissn.) Franco) infected by *Armillaria ostoyae* (Romagn.) Herink, another important root pathogen of conifers in the northern hemisphere, has provided accurate estimates of the impact of non-lethal root infections on tree growth (Cruickshank et al., 2011).

The aims of the current study were to (i) determine the relationship between aboveground symptoms in Scots pine and belowground *Heterobasidion* spp. infection; (ii) evaluate the relationship between tree growth characteristics and root infection severity; and (iii) determine the influence of *Heterobasidion* spp. infection on volume growth.

2. Materials and methods

2.1. Study site and plot establishment

The study was conducted in a 36-year-old pure Scots pine plantation in southern Sweden (55°49′53″N, 14°06′31″E) in August, 2011. The stand was former pasture land with sandy alkaline soils (sand content = 88%, pH = 6.2). The site index, i.e. expected height of dominant Scots pine at 100 years, was 30 m. The stand was thinned without stump treatment in the summer of 2002 and in the spring of 2009. Basidiocarps of *Heterobasidion* spp. were frequently observed on stumps on the site in 2011. In the stand two 8-m radius plots were randomly located around a stump from the previous thinning to ensure that plot trees would be located to a nearby possible source of inoculum. In one plot the center stump had *Heterobasidion* spp. basidiocarps while in the other plot no basidiocarp was present. The two plots were separated by approximately 100 m. In each plot, diameter at breast height (DBH) was measured. Trees were numbered and the north direction was marked with paint on the stem. The vertically projected crown widths in the north–south and east–west directions were also measured.

2.2. Extraction of trees and field measurements

Whole trees and root systems were extracted and sampled according to Cruickshank et al. (2011) with some modification. In each plot, trees and roots were carefully pulled from the soil using a single-grip harvester (EcoLog 580B, EcoLog AB, Sweden), and laid on the ground. Trees were cut at the base of the stem and broken roots were manually excavated. The soil was completely removed from the roots with a scrubbing brush. Fine roots (e.g. diameter <0.5 cm) were removed during the cleaning. Stem discs were cut at 0 m, 0.5 m, 1 m, 1.3 m, 2 m and then at every 2 m interval until the stem diameter was less than 5 cm. The distance between the top stem disc and the apex of the tree was measured. Total tree height was the sum of all measured intervals. The north direction was marked on the cross-section of each stem disc. In addition, five branches were randomly collected from all trees from each of the upper, middle and lower crown. Needle retention was measured by counting the number of annual cohorts of branches that retained needles. The stump and root system of each tree and all stem discs were brought to the laboratory for further analyses.

2.3. Stem discs analysis

In the lab, stem discs were dried at room temperature (approximately 20 °C) for 30–60 days and then sanded with a belt sander. The disc surfaces were polished until all rings were clearly identified. Discs were scanned and ring widths were measured along two perpendicular directions using WinDendro software (2005, Régent Instruments Inc., Canada). The annual DBH, height and volume increments of each tree were retrospectively calculated using WinSTEM software (2005, Régent Instruments Inc., Canada). The radius of a stem disc was the quadratic mean of the two perpendicular radii, i.e. $((r_1^2 + r_2^2)/2)^{0.5}$. Diameter was then calculated by multiplying the mean radius by 2. Height increment was calculated using a linear interpolation method assuming equal annual height increment for the period delimited for the year ring difference on two consecutive stem discs (WinSTEM, 2004). The annual height increment between the top disc and the apex was calculated using the same method. Total stem volume was calculated as the sum of a frustum of a cone between each of the two consecutive discs and the cone from the top disc to the apex of the tree, i.e. $\Sigma(\pi h_{a-b}\cdot(r_a^2 + r_a r_b + r_b^2)/3)$, whereby h_{a-b} is the height of the frustum between each two consecutive discs a and b, while r_a and r_b represent the radius of discs a and b, respectively.

2.4. Root measurements and sampling for Heterobasidion spp.

For each extracted root system, root length was measured from the root collar to the distal tip. If a root was broken, it was noted accordingly and the length was measured to the breaking point. The diameters of all roots were measured using a caliper with an accuracy of 0.1 cm at each 50-cm interval, starting from approximately 0–5 cm distal to the root collar to a minimum diameter of 1 cm, i.e. root sections smaller than 1 cm were not measured nor sampled. The length and diameter was measured for all primary, secondary and tertiary roots. Primary roots were cut from the stumps at the root collar with an electric saw. A 5-cm disc was cut at every 25-cm length interval on all primary, secondary and tertiary roots using a bow saw. The bark and saw blade were sprayed with 70% (v/v) ethanol before each cut to avoid potential contamination by *Heterobasidion* spp. infection. Discs were immediately put in plastic bags and incubated for seven days at room temperature (approximately 20 °C). Both sides of each disc were examined for the presence of *Heterobasidion* spp. conidiophores using a dissecting microscope. Since discoloration and stain can also be caused by mechanical damage, they were not used as criteria to determine root infection.

2.5. Calculations and Statistics

2.5.1. Incidence, severity and distribution of Heterobasidion spp.

Infection severity was determined as the percentage of infected root volume and the percentage of infected primary roots. Infected root volume was calculated using discs sampled from 25-cm root segments on primary, secondary and tertiary roots. If the sampled root disc was infected by *Heterobasidion* spp., the entire 25-cm root length was also regarded as infected. Total root volume was the sum of the volume of all measured root segments, and was calculated using the formula for a frustum of a cone, i.e. $\pi h(a^2 + ab + b^2)/12$, whereby a and b are the two measured horizontal diameters and h

is the root segment length. The percentage of infected root volume for each tree was calculated as the volume of infected root segments divided by the total root volume. The percentage of infected primary roots was calculated as the number of infected primary roots divided by the total number of primary roots. A primary root was regarded as infected even if only one root segment on either primary, secondary or tertiary roots was found to be infected.

2.5.2. Relationship between infection severity and growth characteristics of trees

The difference in absolute volume increment between the two previous 5-year periods (2007–2011 and 2002–2006) was calculated and adjusted to each tree to reduce the effect of tree size on volume increment by dividing the difference by the total volume in 2002.

Crown area was calculated as an ellipse, i.e. $\pi ab/4$, whereby a and b represent the north–south and east–west crown widths, respectively. Needle retention of a tree was the mean number of needle cohorts on each sampled branch. To account for different competition status of the plots, relative DBH, i.e. the ratio between tree size and the average size of all trees in each plot (Pukkala, 1989), was computed. The difference in growth characteristics of the trees between plots was evaluated by t-test in Minitab (ver 16, Minitab Inc., State College, PA, USA). Tree growth characteristics included DBH, height, tree volume, crown area, needle retention, total root volume, the percentage of infected primary roots and the percentage of infected root volume. The relationship between each tree growth characteristic and infection severity, i.e. the percentage of infected root volume and percentage of infected primary roots, was tested using Pearson's correlation in Minitab. The difference in needle retention among the lower, middle and upper parts of the crown was tested using a simple paired t-test in Minitab. All statistical analyses were performed with a significance level of 5%.

2.5.3. Volume growth reduction

A mixed-effect regression model procedure MIXED in SAS (ver 9.3, SAS Institute Inc., Cary, NC, USA) was fit to model the annual volume increment of a tree between 2002 and 2011, using the tree growth characteristics and root infection conditions as independent variables. The independent variables initially considered for inclusion in the model were relative DBH, tree DBH, height, tree volume, crown area, needle retention, root volume, percentage of infected root volume and percentage of infected primary roots. The inclusion of model variables was based on the value of Akaike Information Criterion (AIC), i.e. the model with smaller AIC was better. Time was considered as a continuous variable and described the change in growth pattern over years, which followed a quadratic curve with a peak at year 2009, likely due to the thinning. As a result, 2009 was chosen as the center of the time variable. The model was fit to an initial dataset that included annual volume increments between the years 2002 and 2011. The measurement from the earliest year in the dataset was extracted until the percentage of infected root volume (Damage) was included in the model, indicating the point at which damage in the root system started to negatively impact volume growth. The year in which volume growth reduction commenced was also tested using piecewise regression in Statistica (ver8.0, StatSoft Inc., Tulsa, OK, USA). Autocorrelation in the trees' growth record was dealt with in SAS 9.3, by explicitly declare type = ar(1) in statement REPEATED.

3. Results

3.1. Incidence, severity and distribution of Heterobasidion spp.

Twelve trees were sampled in each of the two plots. Seventy-five percent of trees in Plot 1 (centered on a stump without basidiocarps) and 100% of trees in Plot 2 (centered on a stump with basidiocarps) were infected by *Heterobasidion* spp. despite showing no evidence of aboveground symptoms prior to being extracted (Table 1).

The total number of primary, secondary and tertiary roots were 198, 728 and 465, respectively. Of those, 33% of the primary, 17% of the secondary and 12% of the tertiary roots (average 18%) were broken due to the mechanical excavation. The average length of primary roots was 0.92 m, and the longest was 2.83 m. No taproots were observed. The total number of root sections measured was 3384 (Table 2). Eighty percent of lateral root sections with diameters between 5–9 cm and 93% with diameters between 10–16 cm occurred at 0–49 cm distal to the root collar (Table 2). The percentage of root sections with diameters between 1–4 cm (48%) and smaller than 1 cm (44%) occurred most frequently at 25–74 cm and 75–124 cm, respectively (Table 2).

A total of 1899 root sections with a diameter larger than 1 cm were examined for the presence of *Heterobasidion* spp. Of those, 7% (n = 135) were infected. Discoloration and resin staining were sometimes observed on infected root sections, but staining was also occasionally observed on non-infected root sections. The incidence of *Heterobasidion* spp. infection decreased with increasing distance from the root collar (Fig. 1). Of those 135 infected root sections, 41% (n = 55) were located at 0–24 cm distal to root collar, 32% (n = 43) at 25–49 cm, 21% (n = 29) at 50–74 cm, 4% (n = 6) at 75–99 cm and 1% (n = 2) at 100 cm or further. The largest percentage of infected roots (52%, n = 70) had a diameter between 1–4 cm, 38% (n = 52) had a diameter between 5–9 cm and 10% (n = 13) were within the range of diameters between 10–16 cm. The average percentage of infected root volume was 9.7% (min: 0% and max: 32.2%) for all trees; 9.2% (min: 0% and max: 21.1%) and 11.8% (min: 3.7% and max: 32.2%) in Plots 1 and 2, respectively. The average percentage of infected primary roots was 40.0%; 37.3% (min: 0% and max: 71%) in Plot 1 and 43.4% (min: 14% and max: 75%) in Plot 2. There was no difference in the percentage of infected root volume (p = 0.431) or in the percentage of infected primary roots (p = 0.533) between the two plots. Further, there was no correlation between the percentage of infected root volume and the percentage of infected primary roots for diseased trees (p = 0.548).

3.2. Relationship between infection severity and growth characteristics of trees

There were no significant differences in the average DBH, height, tree volume, root volume, crown area or needle retention

Table 1
Disease incidence and severity and tree growth characteristics of sampled trees in the two plots. Plot 1 (centered on a stump without basidiocarps) and Plot 2 (centered on a stump with basidiocarps). Figures are means. Numbers in brackets are the minimum and maximum value.

Plot	No. trees sampled	Belowground disease incidence (%)	Infected root volume (%)	Infected primary roots (%)	DBH (cm)	Height (m)	Tree volume (dm^3)	Root volume (dm^3)	Crown area (m^2)	Needle retention (years)
1	12	75	9.2 (0–21.1)	37.3 (0–71)	18.4	15.4	216	29.4	14.5	3.3
2	12	100	11.8 (3.7–32.2)	43.4 (14–75)	17.2	16.1	231	39.0	14.0	3.2

Table 2
Number of measured lateral root sections in each root diameter and distance class for all sampled trees. The number of infected root sections is given in brackets.

Diameter (cm)	Distance from root collar (cm)									
	0–24	25–49	50–74	75–99	100–124	125–149	150–199	200–300	Sum	Inf. freq. (%)
Between 10 and 16	55 (11)	15 (2)	4 (0)	1 (0)	0 (0)	0 (0)	0 (0)	0 (0)	75	17.3
Between 5 and 9	114 (27)	111 (17)	35 (7)	15 (1)	4 (0)	2 (0)	1 (0)	0 (0)	282	18.4
Between 1 and 4	355 (17)	502 (24)	527 (22)	364 (5)	192 (2)	107 (0)	63 (0)	17 (0)	2127	3.3
Smaller than 1	7 (–)	86 (–)	140 (–)	210 (–)	187 (–)	130 (–)	106 (–)	34 (–)	900	–
Total	531	714	706	590	383	239	170	51	3384	4.0

"Sum" is the number of measured root sections and may be bigger than the number of sampled root sections since roots that appeared dead after cutting the sections were not sampled for *Heterobasidion* analysis.

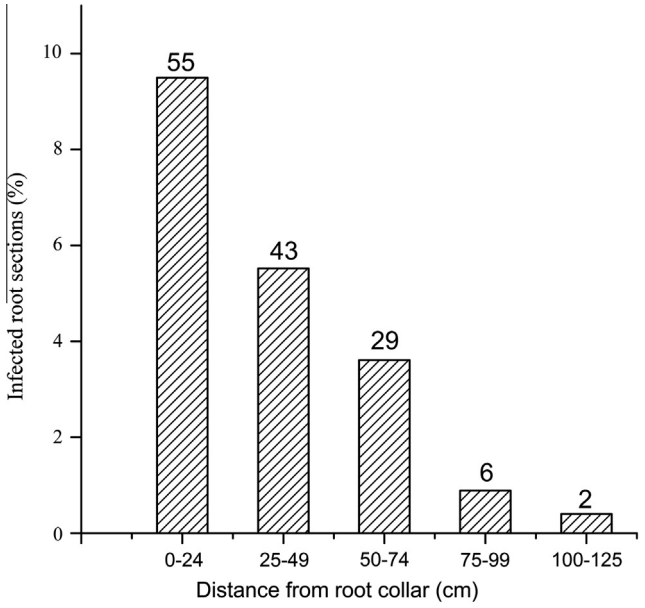

Fig. 1. Percentage of root sections infected by *Heterobasidion* spp. at increasing distances from the root collar. The numbers above bars are the number of infected root sections.

Table 3
Type III tests of fixed effects of Eq. (1), predicting the annual volume increment of Scots pine trees in the two plots.

Effect	Estimate	Num. DF	Den. DF[a]	*F*-value	*P* value
RelDBH	34.0478	1	20	63.93	<0.0001
V2002	−0.05251	1	20	7.57	0.0123
Damage	−14.2721	1	20	6.46	0.0195
Centeryear	−0.8052	1	68.7	14.28	0.0003
Centeryear square	−0.4155	1	48.4	30.26	<0.0001
Intercept	−12.7546				

"RelDBH" is the relative DBH in 2011, "V2002" is the tree volume in 2002, "Damage" is the percentage of infected root volume, and "Centeryear" is the center of the time variable (2009 in this model).

[a] ddfm = Satterthwaite.

between the two plots (Table 1). The percentage of infected root volume was negatively correlated with the difference in absolute volume increment between two 5-year periods ($r = -0.434$, $p = 0.034$), and with the adjusted difference between the two 5-year periods ($r = -0.479$, $p = 0.018$), meaning less volume increment in more infected trees. There was no correlation between the percentage of infected root volume and the volume increment in the year of sampling (2011), DBH, tree volume, root volume, crown area or needle retention for trees in the two plots. The percentage of infected primary roots was positively correlated with DBH and tree volume, but not with the absolute and adjusted volume increment difference between the most recent and the two previous 5-year periods, volume increment in 2011, root volume, crown area or needle retention. DBH was positively correlated with root volume and crown area ($r = 0.976$ and $r = 0.635$, respectively, $p < 0.001$).

Needle retention was in average 3.3 years (Table 1). On all 360 sampled branches, 100%, 99.2% and 92.2% of them retained needles from 2011, 2010 and 2009 respectively. Only 34.2% and 0.3% of the branches had needles from 2008 and 2007 respectively. There was no difference in needle retention among the lower, middle and upper parts of the crown. Trees with needle retention higher than the median had a lower percentage of infected root volume ($p = 0.018$).

3.3. Volume growth reduction

The mixed-effect regression model was fit to a dataset that included the measurement of volume increment between 2005 and 2011. The variables included in the final model were tree volume in 2002 (V2002), relative DBH in 2011 (RelDbh) and percentage of infected root volume (Damage). The formula is given as:

$$Y_{ijk} = \beta_0 + \beta_1 \mathrm{V2002}_{ij} + \beta_2 \mathrm{RelDbh}_{ij} + \beta_3 \mathrm{Damage}_{ij} + \beta_4 \mathrm{year}_{ijk} + \beta_5 \mathrm{year}^2_{ijk} + a_{0j} + a_{1j}\,\mathrm{plot}_j + b_{0i} + b_{1i}\,\mathrm{tree}_i + \varepsilon_{ijk} \quad (1)$$

where y_{ijk} is the annual volume increment (dm^3) of tree i in plot j at year k. β_0 is the intercept. β_1 is the fixed effect of tree volume in 2002, β_2 is the fixed effect of relative DBH in 2011 and β_3 is the fixed effect of the percentage of infected root volume for tree i in plot j measured in 2011. β_4 and β_5 are the linear and quadratic parameters of the year centered around 2009 for tree i in plot j at year k, respectively. a_{0j} is the random intercept for plot j, and a_{1j} is the random effect of plot j. b_{0i} is the random intercept for tree i, and b_{1i} is the random effect of tree i. ε_{ijk} is the residual error.

Annual volume increment decreased with increasing infection in the roots (Table 3). For example, with 10% root volume infected, the volume increment reduced in 2011 was 1.427 dm^3 compared to if it had no infection. Under the assumption that the amount of infection increased linearly from 0% (in 2005) to the average percentage of root volume infected in 2011, the model estimated that the annual volume increment of an intermediate tree (relative DBH = 1) with 10% root infection in 2011 would be reduced by 12.6% (Fig. 2).

4. Discussion

Although growth reduction has been commonly observed in trees infected by *Heterobasidion* spp. (Froelich et al., 1977; Bradford et al., 1978b; Bendz-Hellgren and Stenlid, 1997; Cherubini et al., 2002; Oliva et al., 2010), this is the first study to relate volume growth reduction in Scots pine trees to infection severity in the roots. The results showed that with even a small percentage (7%) of root sections infected, significant growth loss can be detected.

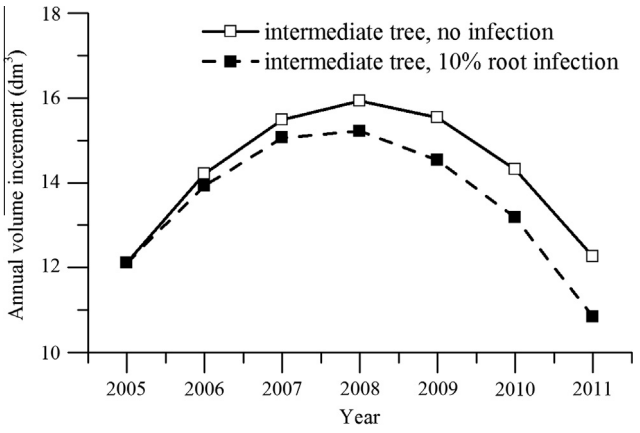

Fig. 2. Estimated annual volume increment for an intermediate tree (relative DBH = 1) with no infection (open square) compared to one tree with 10% of root volume infected in 2011 (closed square). The amount of root volume infected was assumed to increase linearly from 0% in 2005 (3 years after the first thinning which was considered the first entry point of *Heterobasidion* spp. in the stand) to 10% in 2011.

This loss will likely increase over time as the fungus colonizes more root tissue. Hence it seems prudent to consider preventative methods against *Heterobasidion* infection to increase productivity.

Decline in volume increment in infected trees may be attributed to a loss of root function, induction of defense mechanisms leading to decreased allocation of assimilates necessary for tree growth, or both (Garbelotto et al., 1997; Stamp, 2003; Oliva et al., 2012). Fungal growth in roots cause occlusion of tracheids (Joseph et al., 1998) and impair water and nutrient uptake leading to shedding of needles and subsequent reduction in carbon assimilation (Kozlowski and Pallardy, 1997). In this study, trees with low needle retention had a higher percentage of root volume infected. Larger roots with secondary growth are more important for transportation of water and nutrients compared to finer roots (Kozlowski and Pallardy, 1997). Although the amount of fine roots removed was not estimated, in a boreal forest it would normally correspond to approximately 30% of the total root biomass (Kajimoto et al., 1999; Yuan and Chen, 2010). Thus, dysfunction caused by *Heterobasidion* spp. of the larger root mass is likely to have a greater impact on tree growth. In this study, the larger roots were more frequently infected by *Heterobasidion* spp. than smaller roots.

Aboveground symptoms or signs of *Heterobasidion* infection are not adequate indicators for determining the actual incidence. All sampled trees appeared healthy (i.e. no visual crown symptoms or basidiocarps on live trees), yet 87.5% had root infections belowground. Similar results have also been observed in other field surveys of mid-rotation Scots pine stands (Kurkela, 2002; Zhang, 2012). Surveys that focus exclusively on aboveground tree conditions are therefore unreliable for estimating actual disease presence. Growth characteristics, such as crown area, DBH and tree volume were also poor indicators of disease severity in the roots. Although longer needle retention was associated with lower root infection severity, it may not always be useful to assess disease incidence since needle retention can be influenced by factors other than disease, e.g. climate (Reich et al., 1996). Furthermore, assessing disease severity using the frequency of infection in primary roots (Rönnberg et al., 2006) is not adequate, since our results showed no correlation between the percentage of infected root volume and the percentage of primary roots infected. The percentage of infected root volume was considered to be a better estimate of disease severity in trees than the other factors given above.

Yield tables for Scots pine plantations, which are based upon the productivity of seemingly healthy trees, may actually underestimate the potential site productivity (Burdekin, 1972). Better estimations of disease severity and growth impact can result in better management of the stand, e.g. timing of thinnings of stump treatment.

The timing of infection is one factor affecting disease development in stands and the resulting impact on stand productivity, i.e. early infection during the rotation may lead to a higher loss. Stumps created during the 2002 thinning were considered the first entry point for *Heterobasidion* spp. into the stand since the site was a former pasture, and presumably without substantial inoculum. It was not possible, however, to determine the time when infection transferred from a stump root to a living tree through root contact or graft, nor when the dysfunction of infected roots emerged. Rishbeth (1951a) found that the spread rate of *H. annosum* s.s. in the roots of Scots pine was almost 80 cm per year. It can be assumed that under favorable conditions, transfer of infection can take place within one or two years after stump colonization. The year at which *Heterobasidion* started impacting tree growth in our model may be questioned due to variable rates of infection and colonization which may be influenced by tree vigor and host defense mechanisms. However, a general decline in volume increment was detected as early as three years after the first thinning (in 2005).

High disease incidence as observed in this study may be attributable to the sandy, alkaline, well-drained soils, typical of pasture land, which are devoid of antagonistic microorganism (Rishbeth, 1951b) and associated with water stress (Alexander et al., 1975). Such site conditions are not uncommon for Scots pine stands in Sweden (Hallsby, 2013). Thus it is plausible to assume that Scots pine growing on similar type conditions may also suffer losses in site productivity.

The incidence and severity of disease will likely increase over time due in part to the longevity of *Heterobasidion* spp. in Scots pine stumps, e.g. up to 60 years (Greig and Pratt, 1976), but also with subsequent thinnings which create new entry points for the fungus. Although there is a trend to reduce the number of thinnings in the management of Scots pine stands, infections that manage to establish in a stand early in the rotation may pose a significant threat reducing growth for decades (Rönnberg et al., 2013). Furthermore, inoculum in stumps may be carried over to the next rotation affecting productivity of crop trees such as Scots pine, *Larix* spp. and Norway spruce. Consequently, the probability and outcome of introducing *Heterobasidion* spp. into the stand should be taken into careful consideration.

The positive correlation between tree DBH and the percentage of infected primary roots implies that larger trees with larger root systems have a higher probability of infection by *Heterobasidion* spp. This probability likely increases with stand age and tree size (Morrison et al., 2001) and with the amount and quality of inoculum (Morrison et al., 2000). However, once a tree is infected the fungus may take longer to fully colonize the root system of larger trees. Such trees, which are more likely to be left after thinning, may therefore suffer increased infection severity over time. This long-term reduced productivity is seldom accounted for in production prognosis, and may have a significant impact on forecasting potential harvest volumes. Small trees will also suffer increased infection over time but the relative loss of volume increment may be less.

The effect of competition was to some extent taken into consideration in the analysis by using relative DBH. Although dominant trees had higher volume increment compared to intermediate or suppressed trees, when root systems were infected to the same degree volume increment in the dominant trees was further reduced. After thinning, trees normally had enhanced growth regardless of infection severity, but could consequently have been higher without infection (Fig. 2). The benefits of future thinnings on growth of Scots pine may therefore be negated by the effect of chronic root infections by *Heterobasidion* spp.

Knowledge on the location of root infections from this study may be useful for modification of root disease models, e.g. RotStand (Pukkala et al., 2005), for assessing the impact of *Heterobasidion* spp. on Scots pine. In RotStand, growth reduction occurs only when infection spreads to the root collar (Pukkala et al., 2005), whereas the results from this study suggested that infected roots located at various distances from the root collar impact tree growth. The size of the root where infection transfer most likely occurs still remains unknown.

Volume growth reduction in Scots pine caused by *Heterobasidion* spp. increased with increasing percentage of infected root volume. The future of the trees in this stand is unknown but it is plausible to assume that some of the highly diseased trees, e.g. those having greater than 30% of the root volume infected, may succumb to mortality since disease centers were already evident in other areas of the stand. The lightly infected trees may survive for decades and remain asymptomatic but with reduced productivity. The cumulative effect of this loss in productivity can be significant over a rotation, though the limit remains to be defined.

The results of this study have limited applicability because of its design. The calculation of tree growth reduction is based on individual trees. However, assuming a 36-year-old Scots pine plantation with trees of similar size and infection levels, i.e. 87.5% of the trees infected and on average 10% of root volume infected, the annual growth reduction under similar site conditions could amount to 0.87 $m^3\ ha^{-1}$, corresponding to approximately 9.9% of the average annual volume increment per hectare (Skogsstyrelsen, 1984).

Treatment with *P. gigantea* has been proven effective against primary infection by *Heterobasidion* spp. on Scots pine stumps (Korhonen et al., 1993; Korhonen, 2001) and may help improve site productivity. The cost for treatment with *P. gigantea* (Thor, 1996) in Scots pine stands in southern Sweden with a site index of 30 during a typical rotation (Skogsstyrelsen, 1984), i.e. three thinnings at the stand age of 30, 40 and 55 years and a final felling at 80 years, is estimated to be 3092 SEK ha^{-1} (ca. €359) and is equivalent to 8.7 m^3 of pulpwood (355 SEK m^{-3} sub, solid under bark) (Skogsstyrelsen, 2012). When calculating using a discount rate of 3% back to the first thinning, the cost is 1568 SEK ha^{-1} (ca. €182), and the corresponding timber volume 4.4 m^3. Given the estimated annual volume growth reduction for this site and that volume loss will likely increase over time as the fungus colonizes a larger percentage of root tissue, stump treatment can be economically justified. On sites where pulpwood production is the main management target and with high incidence of *Heterobasidion* spp. infection, a shortened rotation may help avoid severe economic losses.

5. Practical implications

A high incidence of *Heterobasidion* spp. may occur in mid-rotation Scots pine stands already nine years after the first thinning. In practice, the majority of these infected trees go unnoticed. The implications of this on timber yield at rotation may be considerable since growth was already reduced and is expected to be further reduced over time. In this study, the anticipated losses caused by *Heterobasidion* spp. on Scots pine were based on only growth reduction, though tree mortality may be inevitable in the long term (Burdekin, 1972; Rönnberg et al., 2006) adding to the overall disease impact. For already infected stands, a shortened rotation may be considered. Control measures such as stump treatment with *P. gigantea* during thinnings, and conducting thinnings during the winter, may be particularly important in stands with low infection levels growing on previously arable land or pastures where the potential for disease spread is high.

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References

Alexander, S.A., Skelly, J.M., 1974. A comparison of isolation methods for determining the incidence of Fomes annosus in living loblolly pine. Eur. J. Forest Pathol. 4, 33–38.
Alexander, S.A., Skelly, J.M., Morris, C.L., 1975. Edaphic factors associated with the incidence and severity of disease caused by fomes annosus in loblolly pine plantations in virginia. Phytopathology 65, 585–591.
Bendz-Hellgren, M., Stenlid, J., 1997. Decreased volume growth of *Picea abies* in response to *Heterobasidion annosum* infection. Can. J. Forest Res. 27, 1519–1524.
Bendz-Hellgren, M., Lipponen, K., Solheim, H., Thomsen, I.M., 1998. The nordic countries. In: Woodward, S., Stenlid, J., Karjalainen, R., Hüttermann, A. (Eds.), *Heterobasidion annosum*: Biology, Ecology, Impact and Control. CAB International, Cambridge, UK, pp. 333–345.
Bradford, B., Alexander, S.A., Skelly, J.M., 1978a. Determination of growth loss of *Pinus taeda* L. caused by *Heterobasidion* annosus (Fr.) Bref. Eur. J. Forest Pathol. 8, 129–134.
Bradford, B., Skelly, J.M., Alexander, S.A., 1978b. Incidence and severity of annosus root rot in Loblolly pine plantation in Virginia. Eur. J. Forest Pathol. 8, 135–145.
Burdekin, D.A., 1972. A study of losses in scots pine caused by *Fomes annosus*. Forestry 45, 189–196.
Cherubini, P., Fontana, G., Rigling, D., Dobbertin, M., Brang, P., Innes, J.L., 2002. Tree-life history prior to death: two fungal root pathogens affect tree-ring growth differently. J. Ecol. 90, 839–850.
Cruickshank, M.G., Morrison, D.J., Lalumière, A., 2011. Site, plot, and individual tree yield reduction of interior Douglas-fir associated with non-lethal infection by Armillaria root disease in southern British Columbia. Forest Ecol. Manage. 261, 297–307.
Froelich, R.C., Cowling, E.B., Collicott, L.V., Dell, T.R., 1977. Fomes annosus reduces height and diameter growth of planted slash pine. For. Sci. 23, 299–306.
Garbelotto, M., Slaughter, G., Popenuck, T., Cobb, F.W., Bruns, T.D., 1997. Secondary spread of *Heterobasidion annosum* in white fir root-disease centers. Can. J. Forest Res. 27, 766–773.
Gibbs, J.N., Greig, B.J.W., Pratt, J.E., 2002. Fomes root rot in Thetford Forest, East Anglia: past, present and future. Forestry 75, 191–202.
Greig, B.J.W., Pratt, J.E., 1976. Some observations on the longevity of Fomes annosus in conifer stumps. Eur. J. Forest Pathol. 6, 250–253.
Greig, B.J.W., 1998. Field recognition and diagnosis of *Heterobasidion annosum*. In: Woodward, S., Stenlid, J., Karjalainen, R., Hüttermann, A. (Eds.), *Heterobasidion annosum*: Biology, Ecology, Impact and Control. CAB International, Cambridge, UK, pp. 35–41.
Hallsby, G., 2013. Plantering av barrträd. In: Skogsskötselserien, p. 59.
Isomäki, A., Kallio, T., 1974. Consequences of injury caused by timber harvesting machines on the growth and decay of spruce (*Picea abies* (L.) Karst.). Acta Forestalia Fennica 136, 1–25.
Joseph, G., Kelsey, R.G., Thies, W.G., 1998. Hydraulic conductivity in roots of ponderosa pine infected with black-stain (*Leptographium wageneri*) or annosus (*Heterobasidion annosum*) root disease. Tree Physiol. 18, 333–339.
Kajimoto, T., Matsuura, Y., Sofronov, M.A., Volokitina, A.V., Mori, S., Osawa, A., Abaimov, A.P., 1999. Above- and belowground biomass and net primary productivity of a *Larix gmelinii* stand near Tura, central Siberia. Tree Physiol. 19, 815–822.
Korhonen, K., Lipponen, K., Bendz, M., Johansson, M., Ryen, I., Venn, K., Seiskari, P., Niemi, M., 1993. Control of *Heterobasidion annosum* by stump treatment with 'rotstop', a new commercial formulation of *Phlebiopsis gigantea*. In: Johansson, M., Stenlid, J. (Eds.), The Eighth International Conference on Root and Butt Rots. Info/Repro, Uppsala, Wik, Sweden and Haikko, Finland, pp. 675–683.
Korhonen, K., 1978. Intersterility groups of *Heterobasidion annosum*. Communicationes Instituti Forestalis Fenniae 94, 1–25.
Korhonen, K., 2001. Simulated stump treatment experiments for monitoring the efficacy of *Phlebiopsis gigantea* against *Heterobasidion*. In: Laflamme, G., Bérubé,

J.A., Bussières, G. (Eds.), 2012, Root and Butt Rots of Forest Trees: 10th International Conference on Root and Butt Rots. Québec City, Canada, pp. 206–210.

Kozlowski, T.T., Pallardy, S.G., 1997. Chapter 11. Absorption of water and ascent of sap. In: Physiology of Woody Plants. Academic Press Inc., California, USA, pp. 237–268.

Kurkela, T., 2002. Crown condition as an indicator of the incidence of root rot caused by *Heterobasidion annosum* in Scots pine stands. Silva. Fenn. 36, 451–457.

Laine, L., 1976. The occurrence of *Heterobasidion annosum* (Fr.) Bref. in woody plants in Finland. Metsantutkimuslaitoksen Julkaisuja 90, 1–53.

Morrison, D.J., Pellow, K.W., Norris, D.J., Nemec, A.F., 2000. Visible versus actual incidence of Armillaria root disease in juvenile coniferous stands in the southern interior of British Columbia. Can. J. Forest Res. 30, 405–414.

Morrison, D.J., Pellow, K.W., Nemec, A.F., Norris, D.J., Semenoff, P., 2001. Effects of selective cutting on the epidemiology of armillaria root disease in the southern interior of British Columbia. Can. J. Forest Res. 31, 59–70.

Oliva, J., Thor, M., Stenlid, J., 2010. Reaction zone and periodic increment decrease in *Picea abies* trees infected by *Heterobasidion annosum* s.l. Forest Ecol. Manage. 260, 692–698.

Oliva, J., Julio Camarero, J., Stenlid, J., 2012. Understanding the role of sapwood loss and reaction zone formation on radial growth of Norway spruce (Picea abies) trees decayed by *Heterobasidion* annosum s.l. Forest Ecol. Manage. 274, 201–209.

Pukkala, T., Möykkynen, T., Thor, M., Rönnberg, J., Stenlid, J., 2005. Modeling infection and spread of *Heterobasidion annosum* in even-aged Fennoscandian conifer stands. Can. J. Forest Res. 35, 74–84.

Pukkala, T., 1989. Predicting diameter growth in even-aged Scots pine stands with a spatial and non-spatial model. Silva. Fenn. 23, 101–116.

Redfern, D.B., Pratt, J.E., Hendry, S.J., Low, J.D., 2010. Development of a policy and strategy for controlling infection by *Heterobasidion annosum* in British forests: a review of supporting research. Forestry 83, 207–218.

Reich, P.B., Oleksyn, J., Modrzynski, J., Tjoelker, M.G., 1996. Evidence that longer needle retention of spruce and pine populations at high elevations and high latitudes is largely a phenotypic response. Tree Physiol. 16, 643–647.

Rishbeth, J., 1951a. Observation on the biology of *Fomes annosus*, with particular reference to East Anglian pine plantations. II Spore production, stump infection and saprophytic activity in stumps. Ann. Bot. 15, 1–21.

Rishbeth, J., 1951b. Observation on the biology of *Fomes annosus*, with particular reference to East Anglian pine plantations. III Natural and experimental infection of pines, and some factors affecting severity of the disease. Ann. Bot. 15, 221–247.

Rishbeth, J., 1963. Stump protection against *Fomes annosus*. III. inoculation with *Peniophora gigantea*. Ann. Appl. Biol. 52, 63–77.

Rönnberg, J., Petrylaitė, E., Nilsson, G., Pratt, J., 2006. Two studies to assess the risk to *Pinus sylvestris* from *Heterobasidion* spp. in southern Sweden. Scand. J. Forest Res. 21, 405–413.

Rönnberg, J., Berglund, M., Johansson, U., Cleary, M., 2013. Incidence of *Heterobasidion* spp. following different thinning regimes in Norway spruce in southern Sweden. Forest Ecol. Manage. 289, 409–415.

Skogsstyrelsen, 1984. Gallringsmallar – Södra Sverige. In: Skogsstyrelsen, Jönköping, Sweden, p. 35.

Skogsstyrelsen, 2012. Swedish statistical yearbook of forestry. In: Official Statistics of Sweden. Swedish Forest Agency, Jönnköping, Sweden, p. 380.

Stamp, N., 2003. Out of the quagmire of plant defense hypotheses. Q. Rev. Biol. 78, 23–55.

Stenlid, J., Redfern, D.B., 1998. Spread within the tree and stand. In: Woodward, S., Stenlid, J., Karjalainen, R., Hüttermann, A. (Eds.), *Heterobasidion annosum*: Biology, Ecology, Impact and Control. CAB International, Cambridge, UK, pp. 125–142.

Thor, M., 1996. Stump treatment against root and butt rot cause by root fomes (*Heterobasidion annosum*) – a study of the literature. In: Research Paper of Skogforsk, Oskarshamn.

Thor, M., 2001. Operational stump treatment against *Heterobasidion annosum* in European forestry – current situation. In: Laflamme, G., Bérubé, J.A., Bussières, G. (Eds.), Root and Butt Rots of Forest Trees: 10th International Conference on Root and Butt Rots. Québec City, Canada, pp. 170–175.

WinSTEM, 2004. Manual for WinSTEM 2004a,b. In: Regent Instruments Inc., Canada.

Yuan, Z.Y., Chen, H.Y.H., 2010. Fine root biomass, production, turnover rates, and nutrient contents in boreal forest ecosystems in relation to species, climate, fertility, and stand age: literature review and meta-analyses. Crit. Rev. Plant Sci. 29, 204–221.

Zhang, J., 2012. A study of root distribution and the effect of *Heterobasidion* spp. root infection on the growth of live Scots pines (*Pinus sylvestris*) in Southern Sweden. In: Southern Swedish Forest Research Center. Swedish University of Agricultural Sciences, Alnarp, Sweden.