

Heart Rot of Spruce and Alder in Forests of Latvia

Impact and Possibilities for Silvicultural Control

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Abstract

Heart rot causes great decrease in timber quality throughout the world. In the conifer forests of Northern Hemisphere the most significant losses are caused by fungi from the *Heterobasidion annosum* species complex. The spread of the disease is favored by forest management, as the fungus can use freshly cut stumps as an infection court, and so extend through root contacts to infect neighboring trees.

The country-scale inventory revealed that about 22% of Latvian spruce trees contained heart rot, which extended on average 6.6 m within the tree stem; the most commonly associated fungus being *H. parviporum*. These facts highlight the need to take actions to reduce the level of inoculum in already infested stands. Stump removal and the planting of resistant tree species are two options for that.

Long-term trials carried out in Scandinavia showed that stump removal could significantly decrease the proportion of infected trees in regenerated stands, although the effectiveness of this procedure may eventually decrease with time. In addition, this method is quite drastic and the adverse influence of continuous stump removal on stand biodiversity and productivity should be taken into consideration. The trial conducted in Finland showed that stump removal had a positive impact on seedlings growth, but influenced negatively mycorrhization and species richness.

The other management method which could be employed is the planting of resistant tree species, especially broadleaved trees. In the work undertaken for this thesis heart rot incidence, associated fungi and decay caused yield losses were studied in Latvian grey (*Alnus incana*) and black alder (*A. glutinosa*) stands. Despite the high decay incidence and the number of associated fungal species, no single *H. annosum* s.l. was found. Also, our data showed that the proportion and size of the heart rot column increased with tree age. These data suggest that a short rotation of fast growing broadleaved trees (not only *Alnus*, but also species of *Populus*, *Betula*, and *Salix*) could be used for clearing up the *Heterobasidion* inoculum.

Keywords: Decay incidence, *Heterobasidion*, *Picea abies*, *Alnus incana*, *Alnus glutinosa*, decay causing fungi, stump removal, mycorrhiza.

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Dedication

This work is dedicated to my mother, Maria, for all the love and support what you have given to me.

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List of Publications

This thesis is based on the work contained in the following papers, referred to by Roman numerals in the text:

- I Arhipova, N., Gaitnieks, T., Donis, J., Stenlid, J. & Vasaitis, R. (2011). Butt rot incidence, causal fungi, and related yield loss in *Picea abies* stands of Latvia. *Canadian Journal of Forest Research* 41: 2337-2345.
- II Cleary, M. R., Arhipova, N., Morrison, D. J., Thomsen, I. M., Sturrock, R. N., Vasaitis, R., Gaitnieks, T. & Stenlid, J. (2012) Stump removal to control root disease in Canada and Scandinavia: A synthesis of results from long-term trials. *Forest Ecology and Management*, in press.
- III Menkis, A., Uotila, A., Arhipova, N. & Vasaitis, R. (2010). Effect of stump and slash removal on growth and mycorrhization of *Picea abies* seedlings outplanted on a forest clear-cut. *Mycorrhiza* 20(7): 505-509.
- IV Arhipova, N., Gaitnieks, T., Donis, J., Stenlid, J. & Vasaitis, R. (2011). Decay, yield loss and associated fungi in stands of grey alder (*Alnus incana*) in Latvia. *Forestry* 84(4): 337-348.
- V Arhipova, N., Gaitnieks, T., Donis, J., Stenlid, J. & Vasaitis, R. (2012) Heart-rot and associated fungi in *Alnus glutinosa* (L.) Gaertn. stands in Latvia. *Scandinavian Journal of Forest Research*, 27(4): 327-336.

Papers I-V are reproduced with the permission of the publishers.

The contribution of Natalija Arhipova to the papers included in this thesis was as follows:

- I Part of the field work and most of the laboratory work. Analysis of data and writing up the paper in collaboration with the co-authors.
- II Field work done together with supervisors. All laboratory work, analysis of data and writing up the Brynne and Ramsåsa part of manuscript.
- III Conducted part of field work.
- IV Field work done together with supervisors. All laboratory work. Analysis of data and writing up the paper in collaboration with the co-authors.
- V All laboratory work. Analysis of data and writing up the paper in collaboration with the co-authors.

Abbreviations

BLAST	Basic Local Alignment Search Tool
CIAP	Calf Intestinal Alkaline Phosphatase
CTAB	Cetyl trimethyl ammonium bromide
DBH	Diameter at breast height
DNA	Deoxyribonucleic acid
ECM	Ectomycorrhizae
Exo I	Escherichia coli exonuclease I
ITS	Internal transcribed spacer
PCR	Polymerase chain reaction

1 Introduction

1.1 Heart rot

Heart rot in living trees causes serious economical losses due to mortality, growth reduction and decrease of timber quality throughout the world. The term “heart rot” is used to describe decay caused by fungi in the central core of the tree (Berry, 1973). Only in temperate regions of the Northern Hemisphere heart rots caused by different decay fungi reduce timber quality by several million cubic meters annually (Hinds, 1977; Woodward *et al.*, 1998). For example, inventories of spruce (*Picea abies* (L.) Karst.) stands in Finland demonstrated that direct losses in sawn timber volume due to stem rot varied on average from 8.5% to 30% (Kallio, 1972; Kallio & Tamminen, 1974; Tamminen, 1985). In broadleaved stands losses caused by heart rot can be even higher. For instance, inventories conducted in *Populus tremuloides* Michx. stands in different regions of the United States showed that on average 42-80% of aspen trees contained stem decay (Davidson *et al.*, 1959; Jones & Ostry, 1998; Warall & Fairweather, 2009).

Many fungal species, mainly basidiomycetes, are capable of causing considerable amount of stem decay in forest trees (Figure 1). Entry occurs through different infection courts, such as fire scars, dead branches, roots, parent stumps, or mechanical injuries (Berry, 1973). Heart rot in stems and roots could be considered as economically the most important cause of decrease of timber quality in managed forests, where cut stumps and wounds on living stems are abundant. For example, the highly destructive fungal pathogens from the genus *Heterobasidion*, which cause considerable heart rot in *P. abies*, are promoted by the forest management (Woodward *et al.*, 1998, Berglund *et al.*, 2012). As the disease is very common, in some studies heart rot of *P. abies* was associated with *Heterobasidion* based only on visual stump assessment (Enerstvedt & Venn, 1979; Nilsen, 1983; Vasiliauskas, 1989; Huse *et al.*,

1994; Vasiliauskas *et al.*, 2002). However, many other decay fungi can cause heart rot in living *P. abies* trees (Pechmann & Aufsess, 1971; Pechmann *et al.*, 1973; Kallio & Tamminen, 1974; Norokorpi, 1979; Hallaksela, 1984; Stenlid & Wästerlund, 1986; Piri *et al.*, 1990; Vasiliauskas *et al.*, 1996; Vasiliauskas & Stenlid, 1998; Vasiliauskas, 1998, 1999; Mäkinen *et al.*, 2007). Furthermore, communities of heart rot pathogens of *P. abies* may vary considerably depending on geographical region, stand age, site characteristics and management history, as reported from Germany (Pechmann *et al.*, 1973) and Finland (Kallio & Tamminen, 1974; Norokorpi, 1979; Hallaksela, 1984).



Figure 1. Heart rot in spruce tree. (Photo: T. Gaitnieks).

Forests cover about 50 % of the total land area of Latvia, and are the most important renewable natural resource in the country. Wood is one of the most essential forest products and makes up a significant component of all Latvian exports (Latvian Forest Industry Federation, 2008). Because of their significance Latvia's forests are intensively managed and, furthermore, the proportion of forest lands is likely to increase in the future due to a country scale policy of afforestation of abandoned farmlands. Despite this, little data are available about the distribution and importance of heart rot in spruce *P. abies* and alders, *Alnus incana* (L.) Moench. and *A. glutinosa* (L.) Gaertn. in Latvian forests. Early studies have shown that the proportion of decayed stumps in spruce clear-cuts varies from 18-68%, and in some stands – even reaches 87% (Eglītis, 1938; Jurēvičs, 1939; Lange, 1960; Mangalis, 1975; Gaitnieks *et al.*, 2004). However, fungi were not isolated in the cited studies, thus the causal agents of the disease remain unidentified. Studies on heart rot incidence in alders were never been previously performed in Latvia. The aim of the current work (Papers I-V) was to investigate frequency of infection by

stem decay fungi, the extent of decay they cause, the associated yield losses and management possibilities for reducing decay incidence in Latvian forests. The identification of stem rot causing fungi (Papers I, IV and V) also assist future silvicultural management decision-making and in helping in planning the most suitable stand regeneration strategies.

1.2 Heterobasidion root rot

Heterobasidion (or annosum) root rot is considered to be one of the most destructive conifer diseases in the north temperate regions of the world (Woodward *et al.*, 1998). In addition to root rot, *H. annosum* s.l. is able to cause extensive heart rot in a number of tree species, including *P. abies*. The disease usually occurs in managed conifer stands where the pathogen may cause direct tree mortality, growth reduction and root and stem decay (Risbeth, 1951c; Hodges, 1969; Bradford *et al.*, 1978; Bendz-Hellgren & Stenlid, 1997; Gibbs *et al.*, 2002). The *H. annosum* s.l. species complex consists of five species that have a wide geographical distribution, particularly, in many parts of Europe, Asia and North America (Dalman *et al.*, 2010). These fungi were once considered to belong to a single species under the name *Fomes annosus* (Fr.) Karst, or more recently, *Heterobasidion annosum* (Fr.) Bref., until mating experiments conducted by Korhonen (1978) revealed there were a number of host specialized intersterility groups. These intersterility groups were initially classified as the P, S (Korhonen, 1978), and F-type (Capretti *et al.*, 1990) based on their host preferences, namely pine, spruce and fir. However, as a result of subsequent phylogenetic studies, polymorphism in the DNA regions proved that three European (P, S and F) and two North American (P and S) intersterility groups belong to different clades, and that the *H. annosum* complex is in fact composed of five species (Johannesson & Stenlid, 1998; Otrrosina & Garbelotto, 2010; Dalman *et al.*, 2010). As a result, new scientific names were proposed: *H. annosum* (Fr.) Bref. s.s. (former European P type), *H. parviporum* Niemelä & Korhonen (former European S type) (Figure 2), *H. abietinum* Niemelä & Korhonen (former European F type), *H. irregulare* Otrrosina & Garbelotto (former American P type) and *H. occidentale* Otrrosina & Garbelotto (former American S type).



Figure 2. Fruitbodies of *Heterobasidion parviporum*.

These *Heterobasidion* species have two ways of dispersing: one - by airborne basidiospores, which invade through the surface of freshly cut stumps or fresh wounds, especially where these are close to the tree base and on roots; and another - by mycelia, which infect roots of neighboring trees via root contacts (Risbeth, 1951a, 1951b, 1951c; Meredith, 1959; Hodges, 1969; Roll-Hansen & Roll-Hansen, 1980; Vollbrecht & Stenlid, 1999). The disease symptoms in living trees, e.g. resin exudation, crown deterioration, butt swelling are not very characteristic and cannot be distinguished from symptoms, caused by other root pathogens (Woodward *et al.*, 1998). *H. annosum* species grow necrotrophically within the sapwood of the living trees, but with the exception of pine infections, extends to the heartwood, causing intensive heart rot, usually without any external symptoms (Gibbs, 1968; Woodward *et al.*, 1998). The decay caused by *H. annosum* is described as a white pocket rot. The color of the stain depends on host species. The incipient decay in most trees is pale-yellow or straw colored, usually surrounded by a darker stained zone, eventually becoming soft, fibrous and light-brown in the advanced stage, sometimes leading to hollowing in the center of stem (Woodward *et al.*, 1998). The spread rate within the individual stem depends on the tree vitality and wood moisture content and can reach rates of 2 m per year in roots and 1 m per year in stems (Hodges, 1969). The spread rate of disease foci through the stand depends on stand type, management history, tree species composition, soil properties and other factors (Risbeth, 1951b; Greig, 1962; Pratt & Greig, 1988; Lindén & Vollbrecht, 2002; Puddu *et al.*, 2003). In the northern temperate forests spread rate may reach on average 20-50 cm per year and the largest detected area occupied by a single genet was about 50 m at diameter (Stenlid, 1985; Piri *et al.*, 1990; Piri, 1996; Garbelotto *et al.*, 1997). The fungus can

survive in the wood residues (for example, stumps) for up to 60 years and still retain the ability to infect the next forest generation (Morrison & Johnson, 1978).

1.3 Disease management

About 46% of Latvian forests are composed of conifer stands, mainly of Scots pine (*Pinus sylvestris* (L.)) and Norway spruce (*P. abies*) (Central Statistical Bureau of Latvia, 2011). Both of these tree species are highly susceptible to infection by *H. annosum* s.l. and disease management is of high importance. As effective control of *H. annosum* s.l. is difficult, all practical methods that help to reduce the infection frequency would be of great value. There are three generally accepted ways to minimize damage in the future: stump removal, freshly-cut stump treatment with chemical or biological preparations and planting of resistant tree species in the succeeding stand generation (Hodges, 1969; Hallaksela & Nevalainen, 1981, Greig, 1984, Woodward *et al.*, 1998, Thor & Stenlid, 2005, Oliva *et al.*, 2008, Vasaitis *et al.*, 2008). Other possible control methods which have been suggested by different authors to reduce economic losses created by root rot are: planting mixed stands with a high proportion of deciduous trees, prescribed burning, seedling planting with wider spacing, reduced or delayed thinning in first rotation stands, site fertilization, and reducing the rotation time in diseased stands (Greig, 1984; Woodward *et al.*, 1998; Piri, 2000; Piri, 2003, Lygis *et al.*, 2004b, 2010).

1.3.1 Biological and chemical stump treatment as a control method

Treatment of freshly cut stumps with biological and chemical preparations is widely used to prevent infection by airborne basidiospores. Today, the most widespread method of *Heterobasidion* control is stump treatment with a *Phlebiopsis gigantea* (Fr.) Jülich spore suspension. The treatment was first suggested by Risbeth (1950, 1951b, 1952, 1961, 1963) and was found to be an effective way of preventing spore infection of freshly cut stumps, especially on different pine species (Greig, 1976; Annesi *et al.*, 2005; Nicolotti & Gonthier, 2005). Nowadays, a number of preparations based on *P. gigantea* have been produced in Europe: “PG Suspension” (UK), “PgIBL” (Poland), “Rotex” (Germany) and “Rotstop” (Finland). Stump treatment with *P. gigantea* preparations is widely used in many European countries (Pratt *et al.*, 2000) for reducing new infections by air-borne basidiospores, but has no effect, if disease is already present in stumps (Pettersson *et al.*, 2003).

Amongst chemical agents used for stump treatment, urea is the most commonly employed. The method works by changing the pH of the stump surface thus making it unsuitable for colonization of *Heterobasidion* airborne basidiospores (Johansson *et al.*, 2002). This method does have a negative impact on the stump inhabiting basidiomycete community (Vasiliauskas *et al.*, 2004) and also on plant species population within the stand (Westlund & Nohrstedt, 2000). However, it has been shown to be quite effective for controlling *Heterobasidion*, especially at urea concentrations of 20-30% (Brantberg *et al.*, 1996; Nicolotti & Gonthier, 2005; Lehtijärvi *et al.*, 2011; Wang *et al.*, 2012) and the toxicity of urea is low. As *P. gigantea* preparations are not as effective in spruce as in pine wood (Berglund & Rönnerberg, 2004), stump treatment with urea could be an alternative method for this species (Oliva *et al.*, 2008).

1.3.2 Stump removal as a control method

Stump removal (Figure 3) to reduce the probability of root disease transmission from stumps of the previous generation to newly planted trees was first suggested for controlling *H. annosum* by Hartig in 1878. Numerous experiments have shown that stump removal is an effective method for controlling *H. annosum* in heavily infested forest areas (Greig, 1980, 1984, Stenlid, 1987, Greig *et al.*, 2001, Vasaitis *et al.*, 2008).



Figure 3. Stump removal (Photo: T. Gaitnieks).

Stump removal has also been recommended for reducing the inoculum potential of other root pathogens, e.g. *Armillaria solidipes* Peck. and *Phellinus sulphurascens* Pilát. in British Columbia (B.C.), Canada (Cleary *et al.*, 2008, 2011; Strurrock *et al.*, 2006). Although this operation is expensive, there has been renewed interest in this procedure during the last few years because of the possibility of using stumps as an energy source (Björheden, 2006, Eriksson & Gustavsson, 2008; Laitila *et al.*, 2008; Walmsley & Godbold, 2010). Moreover, Saarinen (2006) showed that slash and stump removal will increase the work productivity and quality of mechanized forest regeneration. However, little is known about the long term impact of stump removal on root rot incidence. In paper II results from long-term experiments carried out in Sweden, Denmark and Canada for the control of the different root rot diseases: heterobasidion, armillaria and laminated root rot were compared. Data about the possible ecological impact of stump removal are scarce, especially from Europe (Walmsley & Godbold, 2010.). In paper III influence of stump and slash removal on the growth and mycorrhization of *P. abies* seedlings was studied.

1.3.3 Planting of resistant tree species as a control method

The relative resistance of tree species to pathogens is of special interest for reducing losses created by forest diseases. Planting of resistant species on the heavily infested sites has been suggested by number of authors (Stambaugh, 1989; Woodward *et al.*, 1998; Greig *et al.*, 2001). Broadleaved trees are considered to be more resistant to infection than conifers, as *H. annosum* s.l. does not occur or is very rare in pure deciduous tree stands (Domański, 1983; Lygis *et al.*, 2004a). Susceptibility to *Heterobasidion* infection varies greatly not only between different tree species, but also between individuals within one species (Swedjemark & Stenlid, 1995, 1997). Little information about planting alders on diseased sites is yet available, although several reports have been published dealing with the occurrence of *H. annosum* s.l. on *A. incana* (Rishbeth, 1957; Domański & Kowalski, 1987; Kraj & Kowalski, 2010). *Alnus glutinosa* is listed among the host species for *H. annosum* s.l (Woodward *et al.*, 1998). However, an inoculation experiment conducted in Sweden showed that both *H. annosum* s.s. and *H. parviporum* developed poorly in the wood of *A. glutinosa* (Swedjemark & Stenlid, 1995). In papers IV and V, the disease situation in natural *A. incana* and *A. glutinosa* stands was studied. Data on decay incidence and causal agents are very scarce and improved information would help in the planning of the best-practice management strategies in the future. Although broadleaved species are not generally as valuable

economically as conifers, in recent years interest about the short-term energy plantations has increased considerably, which gives a new aspect to this question. The work published in papers IV and V included the results of studies on the frequency of infection by stem decay fungi, extent of decay, and the associated yield losses in Latvian alder stands.

2 Objectives of the thesis

The overall aim was to study the incidence of heart-rot in *P. abies* and *Alnus* spp. stands, the associated fungi, their impact on forest yield and possible silvicultural measures to control *Heterobasidion* spp. with a special emphasis on stump removal.

More specifically the objectives were to evaluate:

- butt rot frequency in Latvian *P. abies* stands, associated fungi and the economical impact on a country scale
- the long-term effect of stump removal on root rot incidence
- the influence of stump and slash removal on the growth and mycorrhization of *P. abies* seedlings
- stem decay frequency in Latvian *A. incana* stands, associated fungi and related yield losses
- heart-rot in Latvian *A. glutinosa* stands, associated fungi and possible yield losses

3 Materials and methods

3.1 Study sites and field work

3.1.1 Butt rot incidence, causal fungi and related losses in *P. abies* stands of Latvia (Paper I)

The study was conducted during years 2005-2006 in 322 thinned and clear-cut *P. abies*-dominated stands distributed throughout Latvia. The sites represented *Oxalidososa* (102 sites), *Hylocomiosa* (86 sites), *Myrtillosa* (67 sites), *Mercurialiosa* (37 sites) and *Carico-phragmitosa*, *Oxalidososa* turf. mel. and *Myrtillosa* turf. mel. (together, 27 sites) forest site types according to the Latvian forest classification system (Bušs, 1997). The age of surveyed stands ranged from 27 to 164 years and their area from 0.2 to 8.7 ha. In total, 24 745 spruce stumps were examined using randomly oriented transects (Bloomberg *et al.*, 1980). Depending on the stand area, 15 to 240 stumps per site were measured and the presence or absence of decay in each was recorded. If present, the diameter of the decay zone was measured and the stage of decay recorded. In addition, cores were collected aseptically using an increment borer from a random selection of 1-10 decayed stumps at each site for subsequent isolation of decay-causing fungi (Section 3.2).



Figure 4. Collecting of wood sample using Presler increment borer.

To estimate the extent of heart rot in living stems 114 decayed mature *P. abies* trees were selected in four stands. All trees were cut, and height, diameter at stump level and DBH of trees, as well as diameter at stump level and total length of decay were measured. Discs were cut from these trees at the stem base (0.2 – 0.3 m above ground level), at breast height (1.3 m), at a height of 2.3 m from the stump, and then continuing at 3 m intervals until the final length of decay column was determined. Discs from the tree base, the middle and top of the decay column were brought to the laboratory and used for isolation of decay causing fungi (Section 3.2).

3.1.2 Stump removal to control *Heterobasidion* root rot (Paper II)

The study was conducted in 2008 in a stump removal trial established earlier at two localities: one at Brynge in northern Sweden and the other at Ramsåsa in southern Sweden. The trial was previously described by Hyppel (1978) and Stenlid (1987). Briefly, in two clear-cut spruce stands, heavily infected by *H. annosum* s.l., six sample plots were established: three at Brynge and three at Ramsåsa. In Brynge in one sample plot stumps were removed using a caterpillar tractor (B1), in other stumps were left intact (B2) and in the remaining plot stumps were treated with 15 g of sodium fluoride (NaF) through a bole hole in the middle of each stump (B3). All plots were planted with 2-year-old *P. abies* seedlings. In Ramsåsa, in the first sample plot stumps were removed using caterpillar tractor, soil was plowed and sieved, leaving no roots larger than 5 mm and two-year-old *P. abies* seedlings were planted at 1.5 m spacing (R1); at the second sample plot stumps left intact and *P. abies* seedlings planted at 1.2 m spacing (R2); and at the third stumps were also not removed and *A. incana* were planted (R3) (Hyppel, 1978). Tree condition and

the frequency of *Heterobasidion* infection were assessed twice before the present study: during 1975/78 by Hyppel (1978) and during 1984/85 by Stenlid (1987). In addition, all trees and stumps from the previous spruce generation and from the thinning (carried out in 1977 at Ramsåsa and in 1978 at Brynge) were mapped.

In 2008, both sites were reassessed. Due to wind storm, plots R2 and R3 at Ramsåsa were destroyed, but plot R1 though severely damaged was still functional. At this and all the Brynge plots (R1, B1, B2 and B3) tree DBH was measured and wood samples were taken using an increment borer. All trees were sampled twice at the stem base, but if the one of the wood samples indicated soft rot, a third sample was taken at breast height. Dead trees were also recorded and wood samples taken only from trees dead 1-3 years previously. All wood cores were used for subsequent fungal isolation.

3.1.3 Effect of stump and slash removal on growth and mycorrhization of *P. abies* seedlings outplanted on a forest clear-cut (Paper III)

This study was conducted in Hyytiälä Forestry Field Station of the University of Helsinki in Central Finland. The experiment was established in May 2005. On a clear-felled area four plots were established using different treatments: 1 – mounding (M); 2 – stump removal (K), 3 – mounding and slash removal (MH); 4 – stump and slash removal (HK). All plots were planted with 1-year-old containerized *P. abies* seedlings. In each treatment, the growth of all *P. abies* seedlings was measured twice: in September 2007 and in May 2009. In May 2009 also root systems from 12 randomly selected plants from each treatment were collected.

3.1.4 Decay, yield loss and associated fungi in stands of *A. incana* in Latvia (Paper IV)

This study was conducted in 46 recently cut clear-felled *A. incana* stands in different regions of Latvia. *Alnus incana* was dominant in all surveyed stands. The ages of the felled stands ranged from 25 to 65 years and represented mostly the *Oxalis* forest site type (Bušs, 1997). In total, 4 434 *A. incana* stumps were examined for the presence of decay (52 -100 stumps per site, depending on site size), using randomly oriented transects (Bloomberg *et al.*, 1980). For all stumps diameter of the cut surface, and diameter of the decay zone, if present, were measured using a ruler. The presence of spongy rot (decomposed wood squeezable with fingers) and hollowed stumps were also recorded. On each site, from 1 to 12 decayed stumps increment cores were collected for

subsequent isolation of decay causing fungi. In total, 227 stumps were sampled.

In addition, 431 living *A. incana* trees were sampled using an increment borer close to the stem base for evaluation of the presence or absence of heart rot in seven between 25- and 65-year-old stands (40-100 trees per stand). All collected wood cores were used for subsequent fungal isolation. In order to estimate the correlations between stem and decay parameters, 175 decayed stems from among those that were sampled, were felled, dissected and the following parameters recorded: age of tree, stump diameter, tree DBH, tree height, decay diameter at stump level and length of decay column. The presence of spongy heart rot in each stem was also recorded and its diameter at stump level and length measured.

3.1.5 Heart-rot and associated fungi in *A. glutinosa* stands in Latvia (Paper V)

This study was conducted in four between 51- and 84- year-old *A. glutinosa* stands located in the Kalsnava forest district in central-eastern Latvia. A total of 450 trees (100 – 150 trees per site) were sampled using an increment borer for evaluation of the presence or absence of heart rot. Each tree was sampled once close to the stem base and wood cores were collected for subsequent fungal isolation. In addition, 34 stems which showed the presence of heart rot were felled, dissected and the followed parameters recorded: tree age, stem diameter at stump level, tree DBH, tree height, decay and spongy rot diameter at stump level and length of the decay and spongy rot column.

3.2 Identification of fungi

All collected samples were transported to the laboratory and held in a refrigerator at +4 °C. Isolations from wood samples (Papers I, III, IV, V) were performed 1-2 days after field sampling. Each wood sample was flame-sterilized, placed on malt or Hagem agar media and incubated in the dark at 19 – 20 °C. To obtain pure cultures, the emergent mycelia were subcultured on Hagem agar media in separate Petri dishes. After 1-3 weeks of incubation, all pure cultures were examined under the light microscope (Leica DM400B) and grouped into cultural morphotypes according to macro- and microscopic morphological features of the fungal mycelia (Figure 5). For identification of fungal species and genera, morphological and molecular techniques were used. For identification of *Heterobasidion* species, intersterility tests were performed (Korhonen, 1978) using homokaryotic test cultures 96119 and 98036 (*H.*

parviporum); 03013 and 03015 (*H. annosum* s.s.). Clones of *Heterobasidion* were identified using somatic incompatibility pairing test (Stenlid, 1987).



Figure 5. a. Culture of *H. parviporum*. b. Conidiophores of *H. parviporum*

Spruce roots (Paper III) were washed in tap water, and 100 single root tips from each plant were randomly selected from different parts of root systems using forceps. ECM roots were identified using dissection microscope and morphotyped according to their morphological features.

One to three representatives from mycelial morphotypes (with some exceptions) and one to three randomly selected representatives of each distinct ECM morphotype from each root system were subjected to molecular identification following modified procedures from previous studies (Vasiliauskas *et al.* 2004, 2005). For DNA extraction, fungal mycelia were collected from Petri dishes using a flame-sterilized scalpel and placed into impact-resistant tubes with 800 μ l 2% CTAB solution and 3 glass beads. After that, mycelia were ground using FastPrep (MP Biomedicals) at speed 6000 and tubes placed for 1 hour in a heater (DB2P, Techne Dri-block®, Labasco) at 65 °C. After centrifugation (Biofuge Pico, Heraeus) 750 μ l of supernatant were placed into a new Eppendorf tube and under a fume hood 750 μ l of chloroform were added. After centrifugation 450 μ l of supernatant were very carefully removed from the upper part of the solution and placed in a new Eppendorf tube. The DNA was precipitated using 2-propanol in the proportion 1:2. After centrifugation 2-propanol was removed, and the DNA pellet was centrifugated with 200 ml of 70% ethanol. Then ethanol was carefully removed and DNA dissolved in 50 μ l of purified water (Milli-Q). The DNA concentration was determined using spectrophotometer (ND-1000, NanoDrop®).

For PCR amplification, primers ITS1F (CTTG GTCATTTAGAGGAAGTAA) and ITS4 (TCCTCCGCTTATTGATATGC) with an annealing temperature 55

°C were used (Kåren *et al.*, 1997). The thermal cycling was applied in a thermal cycler (Thermocycler GeneAmp®, PCR System 2700, Applied Biosystems). The ready PCR products were purified using CIAP and Exo I. After purification, PCR products were Sanger sequenced (Sanger *et al.*, 1977) by Macrogen (Seoul, Republic of Korea) using the primer ITS4 for every specimen. Sequencing was performed in one direction. All sequences were manually edited using the Lasergene software package SeqMen (version 5.07, DNASTAR, Madison, Wisconsin and BioEdit version 7.0.5.2. (Hall, 1999). BLAST searches were performed using three reference sequence databases: one at GenBank (<http://www.ncbi.nlm.nih.gov/blast>), one at the Department of Forest Mycology and Pathology, Swedish University of Agricultural Sciences and at UNITE (Kõljalg *et al.*, 2005). The ITS sequence homology was set at 98%-100% for delimiting fungal taxon (presumed species) and 94%-97% for delimiting at the genus level. ITS sequences of each sequenced morphotype (from pure fungal cultures and ECM) were deposited in GenBank.

3.3 Volume calculations and statistical analysis

Stand characteristics (species composition, age and site type) of Latvian trial plots were obtained from the stand inventory database in the Forest State Register (Mr. Indulis Brauners, Latvian State Forest Inc., personal communications). Calculations of decayed log volume were based on actual lengths of the columns of decay (Papers I, IV and V) and spongy rot (Paper IV and V) in the felled trees, according to Ozoliņš (2002). Calculations of decay volume in the stand were based on the volume of decayed log and the number of stems containing butt rot. Data were applied in the context of regional forest yield models and tables (Kuliešis, 1993). The calculations were accomplished following the procedure described in details in Papers I and IV. The correlation analyses were done using Microsoft Excel and significance levels were evaluated according to Liepa (1974). The analyses of proportions (chi-squared tests) and t-tests were conducted using Minitab® 15 software and their significance evaluated according to Fowler *et al.* (2001). In cases when the same dataset was subjected to repeated comparisons, confidence limits were reduced as required by the Bonferroni correction (Sokal & Rohlf, 1995). Analysis of similarity between fungal communities (Papers I, III, IV and V) was performed by calculation of qualitative Sorensen similarity indices (Magurran, 1988).

4 Results and discussion

4.1 Heart rot: incidence, causal fungi and related losses

4.1.1 Heart rot of spruce (Paper I)

Heart rot was detected in about one fifth (22%) of Latvian *P. abies* trees, a value which is compatible with the 10-30% rot frequency detected during previous studies done in Nordic and Baltic countries (Enerstvedt & Venn, 1979; Norokorpi, 1979; Nilsen, 1983; Vasiliauskas, 1989; Huse *et al.*, 1994; Vasiliauskas *et al.*, 2002). Significant relationships were found between both stand age and forest site type and rot incidence and intensity (the possibility of finding heart rot was higher in larger stumps), which generally agree with reports by other authors (Enerstvedt & Venn, 1979; Norokorpi, 1979; Nilsen, 1983; Mangalis, 1975). However, despite the reports of several authors that butt rot frequency in *P. abies* stands tends to decrease with an increasing proportion of deciduous and Scots pine trees (Enerstvedt & Venn, 1979; Huse *et al.*, 1994; Linden & Vollbrecht, 2002, Vasiliauskas *et al.*, 2002; Gaitnieks *et al.*, 2008), a positive correlation between an admixture of other tree species and rot frequency was not found in present study. This, as well as the generally high variation of rot incidence (from 0 to 83% in individual stands), may be due the different history of the surveyed stands: the previous tree rotation, the level of stem damages by big game or during forest operations, forest management operations during growing season and in previous generations, and others factors, all of which can have a significant influence on rot frequency (Isomäki & Kallio, 1974; Piri *et al.*, 1990; Swedjemark & Stenlid, 1993; Rönnerberg, 2000; Piri & Korhonen, 2001; Vasiliauskas, 2001; Piri, 2003; Mäkinen *et al.*, 2007; Piri & Korhonen, 2008).

The length of the heart rot columns in the felled trees was 6.6 m on average, which means that approximately 30% of tree stem of harvested trees, including the valuable butt log component, was spoiled by decay. In Paper I the relationship between the average volume of decayed logs and various stem parameters were presented in order to calculate actual timber losses due to heart rot in Latvian *P. abies* stands. The results indicated that 6-16% of the total standing volume in Latvian spruce stands could be damaged by decay, which generally agrees with the 8-30% of yield loss reported in Finnish studies (Kallio, 1972; Kallio & Tamminen, 1974; Tamminen, 1985). Yield losses increased with time. They were quite low in the relatively young spruce stands (40-50 years), two times higher in mature spruce stands (60-100) and three times higher in over-mature spruce stands (120-150 years).

Heterobasidion parviporum, following by *Stereum sanguinolentum* (Alb. & Schwein.) Fr. were identified as the main causes of heart rot in Latvian *P. abies* stands. Both of these fungi are known as common butt and stem rot agents in living *P. abies* trees (Pechmann & Aufsess, 1971; Pechmann *et al.*, 1973; Isomäki & Kallio, 1974; Hallaksela, 1984; Woodward *et al.*, 1998; Vasiliauskas, 1999, 2001, and references therein). In agreement with earlier reports, other decay-causing agents in living *P. abies*, namely *Amylostereum areolatum* (Chaillet ex Fr.) Boidin, *A. chailletii* (Pers.) Boidin, *Armillaria cepistipes* Velen., *Climacocystis borealis* (Fr.) Kotl. & Pouzar and *Porodaedalea chrysoloma* (Fr.) Fiasson & Niemelä were found only incidentally (Pechmann & Aufsess, 1971; Pechmann *et al.*, 1973; Norokorpi, 1979; Hallaksela, 1984). An unusual observation in the present study (Paper I) was the occurrence of some fungi, known to behave as dead wood decomposers or saprophytes, in the active decay columns of study trees. However, some of these species (namely *Bjerkandera adusta* (Willd.) Karst., *Gloeophyllum sepiarium* (Wulfen) Karst. and *Polyporus brumalis* (Pers.) Fr.) have been reported as occasional butt rot agents in southern Germany and northern Finland (Pechmann & Aufsess, 1971; Pechman *et al.*, 1973; Norokorpi, 1979).

The incidence of *Heterobasidion* - infected trees in Latvian spruce forests confirms that active management measures should be taken in order to reduce the extent of the yield losses caused by this disease. In 2007 the commercial Rotstop preparation was registered in Latvia and stump treatment now is a common procedure in thinned stands (Kenigšvalde *et al.*, 2011). However, as this stump treatment is only able to prevent the formation of new disease centers, other methods which could reduce the inoculum in already infected

stands should be taken into consideration. Two options that should be considered in a programme to reduce disease levels are the removal of decayed stumps and the planting of resistant tree species on heavily infected areas.

4.2 Heart rot of grey and black alders

The heart rot incidence found in this study in *A. incana* and *A. glutinosa* stands was generally high (1-54% in *A. incana* stands and 53-98% in *A. glutinosa* stands). The larger proportion of decay in *A. glutinosa* stands could be related to the older age of the surveyed stands, as well as to the fact, that a much smaller number of *A. glutinosa* stands (4) was investigated in comparison with the larger number of *A. incana* stands (46). The mean length of the heart rot column was generally similar for both species: 7.1 m for *A. incana* and 7.7 m for *A. glutinosa*. However, the mean length of spongy rot within the heart rot column was larger in *A. glutinosa* stems (4.2 m) than in *A. incana* stems (2.8 m). The spongy rot incidence was also higher in the *A. glutinosa* stands. The length of the heart rot zone correlated with tree age and tree size, indicating that the extent of rot tended to increase with time. However, in both cases the variation within the data was high, a result also found by other authors (Kärki, 1999; Kärki *et al.*, 2000).

Despite the fact that *A. incana* and *A. glutinosa* belong to the same genus, the most significant heart rot causing fungi were different in each species. In *A. incana* the most commonly isolated fungus was *Chondrostereum purpureum* (Pers.) Pouzar, whereas in *A. glutinosa* it was *Inonotus radiatus* (Sowerby) Karst. This may explain the lower incidence of spongy rot in the *A. incana* stands, compared with those of *A. glutinosa*, as well as the smaller extent of spongy rot within the decay column in *A. incana*, since *C. purpureum* is a less powerful decay agent. The biology of these two fungi is quite different; *I. radiatus* being a true heart rot fungus, while *C. purpureum* is a typical wound pathogen.

Yield losses caused by decay in alders tend to increase with time and may reach 20% of the standing volume by age 60-65 years for *A. incana* and over 50% of the standing volume at the age 80 years for *A. glutinosa*. The mean volume of wood degraded by the particularly serious spongy heart rot, which significantly reduces wood quality, was determined as 32.7 m³ha⁻¹ in fully stocked 60-65 years old *A. incana* stands and 94.4 m³ha⁻¹ in fully stocked 80 years old *A. glutinosa* stands. Since the harvesting age for *A. glutinosa* stands is prescribed at 71 years under Latvian law (corresponding clause nr. 9, Forest

Law of Latvia), high yield losses can be anticipated for diseased stands of this age. However, most of *A. glutinosa*-dominated stands currently have protected status in Latvia as key biotopes. For *A. incana*, the situation is better, as the harvesting age is not determined by law, and stands may be cut at any time. This should enable forest managers to avoid significant yield losses if stands are felled before heart rot increases to unacceptable levels. Also, a lower decay incidence was demonstrated in mixed *A. glutinosa* stands, and this question is worth further investigation.

4.3 Heart rot: possibilities for silvicultural control

4.3.1 Stump removal as method to control root rot (Papers II and III)

Stump removal cannot completely eradicate *H. annosum* s.l. from the stand, but it may help to reduce the inoculum potential in the next rotation stand and so reduce future timber losses. Numerous studies done in North America and Europe show that in most cases stump removal reduces root rot incidence in the next tree generation and increases stand productivity (Vasaitis *et al.*, 2008). Results of the long-term trials described in Paper II also show that stump removal significantly reduced damage caused by root rot fungi in the regenerated stands. Although the frequency of heterobasidion root rot increased with stand age, it was nevertheless 20-72% lower than in the control areas. Recent interest in the harvesting of tree stumps for fuel may give additional benefits and partially cover stump removal costs (Bjorheden, 2006; Laitila *et al.*, 2008). Historically, although stumps have previously been used as a fuel source, a subsequent decline in the demand for fuel wood had reduced its use for this purpose by the first part of 20th century (Walmsley & Godbold, 2010). Although interest in stump harvesting for energy production increased during the 1970s in Scandinavia, this source was eventually found to be too expensive (Hakkila, 2004). However, since the demands for forest fuels continue to grow, a re-evaluation of the possibility of stump harvesting had recommenced (Erikson & Gustavsson, 2008; Lazdiņš *et al.*, 2009; EUSTAFOR, 2010).

Data on the environmental impact of stump removal are scarce and more research is needed under European conditions. Some American trials showed a decrease in the amounts of C and N present in the soil after stump removal, especially on poor sites (Zabowski *et al.*, 2008; Butnor *et al.*, 2012). However, a comparative analysis done by Vasaitis *et al.* (2008) showed mostly a positive influence of stump removal on seedling establishment, tree growth and stand

productivity. Our results (Paper III) also revealed that stump removal improved seedling growth, which mainly agrees with reports by other authors (Vasaitis *et al.*, 2008; and references therein), but had a negative impact on seedling mycorrhization and diversity of mycorrhizal fungi. To date, a number of authors have recorded possible negative effects of stump removal on stand biodiversity, as many species of wood decay fungi, invertebrates, lichens and bryophytes use the stumps as their habitat (Jonsell, 2007; Rudolphi, 2007; Caruso, 2008; Hjältén *et al.*, 2010). However, cut stumps do not occur in natural forests and are attributed solely to the human activity in the recent hundreds years. Large quantities of stumps created each year in forest stands provide a favourable substrate for many pests and pathogens, such as the large pine weevil, *Hylobius abietis* L., and the root rot fungi belonging to *H. annosum* species complex. However, no evidence so far exists that any of the rare or protected species is restricted solely to a stump habitat. Moreover, not all stands are suitable for stump removal. Stump removal should not be attempted, for instance, on sites of slopes greater than 20° or with sandy or peat soils (the peat depth exceeding 45 cm) or on riparian buffer zones (EUSTAFOR, 2010). Large numbers of stumps are also created during stand thinning. The inventory conducted in Latvia (Paper I) showed that rather similar communities of wood inhabiting fungi were detected in *P. abies* stumps created during clear felling and thinning operations (Sorensen similarity index 0.68). However more species were found in stumps on the clear cut sites, a question which needs further investigation. In fact, *H. parviporum* was the most abundant fungus in both clear-felled and thinned stumps. The ability of this fungus to infect healthy trees through root contacts as well as survive in cut stumps for decades means that inoculum is able to accumulate in forest stand. Moreover the fruitbodies of *H. annosum* s.l. can be quite abundant on colonised stumps (Vasiliauskas *et al.*, 2002; Müller *et al.*, 2007) as well as on cull pieces left in the forest (Schütt *et al.*, 1979; Müller *et al.*, 2007; Stivriņa *et al.*, 2010), providing an additional risk of infection not only to neighboring trees, but also to adjacent stands.

Results from trials reported in Paper II indicate that stump removal has a long-term impact and significantly decreases *Heterobasidion* infection 50 years after planting. Furthermore, although the disease incidence increased with time, it still remained on an acceptable level. Combining stump removal with other root rot control methods could achieve even better results. Treatment of the thinned stumps of the subsequent generation with *P. gigantea* spore suspensions, thinning during the cold season, when the probability of spore infection is low (Yde-Andersen, 1962) and avoiding damage to living trees

from heavy machinery (Vasiliauskas, 2001) will prevent the development of new infection centers and help to reduce losses. Modern methods of stumping have improved the quality of stump lifting by extracting the whole root system with minimal soil damage. However, more research is needed for the evaluation of the possible ecological effects of removing such a great amount of dead wood within stands, especially if stump removal will become a standard forest management procedure. The long-term consequences of stump removal on stand productivity, soil quality, biodiversity and tree growth in different geographical regions of Europe also needs further investigation.

4.3.2 Planting resistant tree species

As stump removal should be avoided on some sites because of the soil type, slope or other factors, the planting of *Heterobasidion*-resistant trees, especially broadleaved species, could be considered as one of the disease management options (Woodward *et al.*, 1998; Piri, 2003; Pavlov *et al.*, 2008). For example, Silver birch *Betula pendula* Roth was found to be resistant to *H. parviporum* infection (Piri, 1996), although *H. annosum* s.s. can quite heavily infect this tree species (Lygis *et al.*, 2004a). Concerning the two alder species, *A. incana* and *A. glutinosa*, little is known about the susceptibility of pure stands of these hosts to infection by *H. annosum* s.l. Although the decay incidence in the evaluated alder stands was high (Papers IV and V) and large number of different fungal species was detected, no species from the *H. annosum* species complex was found among them. Frequency of decay in these studies was mostly attributed to the stand age and timber losses caused by heart rot significantly increased in stands older than 50 years. This finding indicates that the 10- to 20-year-rotation age proposed for energy plantations in Scandinavia and Baltic countries (Rytter, 1995, 1996; Lohmus *et al.*, 1996; Daugavietis *et al.*, 2009) may be suitable not only for biomass production, but also to improve forest health. The additional beneficial effect of planting *Alnus* spp. on heavily infested sites could result from the antagonistic effect of alder rhizosphere microflora to *H. annosum* s.l. (Johansson & Marklund, 1980; Arhipova *et al.*, 2008), as well as the ability of alders to fix atmospheric nitrogen, which could improve soil productivity (Huss-Danell & Lundmark, 1988; Uri *et al.*, 2002; 2003; Kuznetsova *et al.*, 2010). Even if the tested species is not fully resistant to *H. annosum* s.l. infection, a short rotation time is likely to reduce the build-up of new inoculum. Stands of coppice origin, especially on sites with a high decay incidence, are better avoided, as the newly emerging sprouts could be infected by decay fungi from the parent stump. This could reduce sprouting incidence, decrease growth and survival, and could lead to stem decay at a very

young age (Roth & Sleeth, 1936; Berry & Lombard, 1978; Kärki *et al.*, 2000; Wolken *et al.*, 2009; Bakys *et al.*, 2011). For example, the most common heart rot causal agent in Latvian *A. incana* stands, *C. purpureum*, is known as a sprout killer and this species were proposed by some authors for forest vegetation management (Harper *et al.*, 1999; Pitt *et al.*, 1999). Also, the ability of *Heterobasidion* sp. to infect stumps of short rotation tree species is unknown. Therefore, it would be useful to establish trials in short term rotation stands of fast-growing native *Alnus*, *Populus*, *Betula*, *Salix* and other broadleaved species on sites, heavily infected by *H. annosum* s.l., in order to study their resistance potential in more detail under field conditions. Such studies might give promising results, especially taking into account the recommended short rotation age (10 – 20 years), so that the planted species might escape serious damage by *Heterobasidion*, even if the host trees are not completely resistant.

4.4 Conclusions

Studies conducted in this thesis show that *H. parviporum* is the main cause of heart rot in *P. abies* stands of Latvia and that the associated yield losses are significant. The ability of this pathogen to spread within stands through root contacts and to survive in plant residues for decades indicates that such forest sites are permanently infested, and that the disease will persist over future forest generations. Stump treatment could prevent, or at least, reduce infections of healthy stands from airborne basidiospores. However, as the disease is already present in a great number of Latvian *P. abies* stands, other methods for reducing the future yield losses should be attempted, especially on heavily infected sites. The most promising methods, both from a forest health and an economic point of view, are likely to be stump removal and the planting of fast-growing broadleaved tree species, such as native species of birch, aspen and alder, on heavily infected sites. However, as the long term impact of this type of forest management is still unknown, more research is needed concerning its potential ecological impact. Stump and slash removal, as well as the introduction of short rotation energy plantation could lead to a dramatic decrease in nutrient concentrations, at least in some types of forest soils with the removal of large amounts of biomass for energy production. The long-term consequences of this procedure to future forests productivity are still unclear. However, taking ecological and silvicultural consideration into account, optimally healthy and productive forest stands could be achieved.

5 Future prospects

New stump removal trials should be set up in different geographical regions of Europe to evaluate the long term effects of this treatment on site productivity, biodiversity and other related factors. A number of associated questions also deserve to be answered. In particular, what is the size limit of the *Heterobasidion* colonised wood remnants left on the site after stump removal which will still make the infection of new tree generation possible? There is little interest in the use of small dimension stumps (less than 10 cm in diameter) as an energy resource due to their small size and yet stumps of these dimensions are not usually treated with *P. gigantea* preparations. The question therefore arises as to whether and to what extent they may serve as a substrate for colonisation by *H. annosum* s.l. basidiospores and act as a centre for the establishment of new disease foci in the next generation. Also, could fruitbodies produced on colonised stumps and other wood residues left in forest increase the possibility of infection of neighbouring healthy stands by *Heterobasidion* spores?

Experimental energy plantations of native fast growing broadleaved species should be planted on infected sites and their potential resistance to heterobasidion root rot evaluated under the field conditions. Also, the economic aspects should be taken into consideration when designing such trials.

References

- Annesi, T., Curcio, G., D'Amico, L., Motta, E. (2005). Biological control of *Heterobasidion annosum* on *Pinus pinea* by *Phlebiopsis gigantea*. *Forest Pathology*, 35(2): 125-134.
- Arhipova, N., Gaitnieks, T., Nikolaeva, V., Vulfa, L., Mihailova, A. (2008). Impact of grey alder on microflora of rhizosphere of Norway spruce and on its antagonism against *Heterobasidion annosum*. *Mežzinātne*, 17(50): 9-21 [In Latvian with English abstract].
- Bakys, R., Vasiliauskas, A., Ihrmark, K., Stenlid, J., Menkis, A., Vasaitis, R. (2011). Root rot, associated fungi and their impact on health conditions of declining *Fraxinus excelsior* stands in Lithuania. *Scandinavian Journal of Forest Research*, 26(2): 128-135.
- Bendz-Hellgren, M., Stenlid, J. (1997). Decreased volume growth of *Picea abies* in response to *Heterobasidion annosum* infection. *Canadian Journal of Forest Research*, 27(10): 1519-1524
- Berry, F.H. (1973) Heart rots of central region hardwoods. *USDA Forest Service Forest Pest Leaflet*, 140, 8 p.
- Berry, F.H., Lombard, F.F. (1978) Basidiomycetes associated with decay of living oak trees. *USDA Forest Service Research Paper NE-413*, 8 p.
- Berglund, M., Rönnberg, J. (2004). Effectiveness of treatment of Norway spruce stumps with *Phlebiopsis gigantea* at different rates of coverage for the control of *Heterobasidion*. *Forest Pathology*, 34(4): 233-243.
- Berglund, H., Hottola, J., Panttilä, R., Siitonen, J. (2011) Linking substrate and habitat requirements of wood-inhabiting fungi to their regional extinction vulnerability. *Ecography*, 34: 864-875.
- Björheden, R. (2006) Drivers behind the development of forest energy in Sweden. *Biomass & Bioenergy*, 30 (4): 289-295.
- Bloomberg, W.J., Cumberbirch, P.M., Wallis, G. W. (1980) A ground survey method for estimating losses caused by *Phellinus weirii* root rot. II. Survey procedures and data analysis. *Canadian Forest Service, Pacific Forest Research Centre, Victoria, B.C., Information Report, BC-R-4*.
- Bradford, B., Alexander, S. A., Skelly, J. M. (1978) Determination of growth loss of *Pinus taeda* L. caused by *Heterobasidion annosum*. (Fr.) Bref. *European Journal of Forest Pathology*, 8(3): 129-134.

- Brandtberg, P.-O., Johansson, M., Seeger, P. (1996) Effects of season and urea treatment on infection of stumps of *Picea abies* by *Heterobasidion annosum* in stands on former agricultural land. *Scandinavian Journal of Forest Research*, 11(1-4): 261-268.
- Bušs, K. (1997) Forest ecosystems classification in Latvia. In: *Proceedings of Latvian Academy of Sciences, Section B*, 51, 204-218.
- Butnor, J.R., Johnsen, K.H., Sanchez, F.G., Nelson, C.D. (2012) Impacts on pine species, stump removal, cultivation, and fertilisation on soil properties half a century after planting. *Canadian Journal of Forest Research*, 42(4): 675-685.
- Capretti, P., Korhonen, K., Mugnai, L., Romagnoli, C. (1990) An intersterility group of *Heterobasidion annosum* specialized on *Abies alba*. *European Journal of Forest research* 20(4): 231-240.
- Caruso, A., Rudolphi, J., Thor, G. (2008) Lichen species diversity and substrate amounts in young planted boreal forests: a comparison between slash and stumps of *Picea abies*. *Biological Conservation*, 141(1): 47-55.
- Central Statistical Bureau of Latvia (2012) Natural Resources. Tree species in Forests of Latvia. Available online at: <http://www.csb.gov.lv/en/statistikas-temas/natural-resources-key-indicators-30500.html> [Cited 11 June 2012].
- Cleary, M., van der Kamp, B., Morrison, D. (2008) British Columbia's southern interior forests: Armillaria root disease. *Stand Establishment Decision Aid*, BC JEM. 9, 60-65.
- Cleary, M., Sturrock, R., Hodge, J. (2011) Southern Interior Forest Region: Laminated root disease. *Stand Establishment Decision Aid*, BC JEM. 12, 17-20.
- Dalman, K., Olson, A., Stenlid, J. (2010) Evolutionary history of the conifer root rot fungus *Heterobasidion annosum sensu lato*. *Molecular Ecology*, 19(22): 4979 – 4993.
- Daugavietis, M., Daugaviete, M., Bisenieks, J. (2009) Management of grey alder (*Alnus incana* (L.) Moench.) stands in Latvia. In: Malinovska, L., Mezote, A., Kronbergs, E., Nulle, I. (eds.) *Proceedings of 8th International Scientific Conference Engineering for Rural Development*, Jelgava, May 28-29. Latvian University of Agriculture, Jelgava, Latvia, pp. 229 – 234.
- Davidson, R.W., Hinds, T.E., Hawksworth, F.G. (1959) Decay of Aspen in Colorado. *USDA Forest Service*, Station Paper No. 45. 14 p.
- Domański, S. (1983) Fungi that destroyed *Populus tremula* stand in Lagów Lubuski. *European Journal of Forest Pathology*, 13(3): 166-173.
- Domański, S., Kowalski, T. (1987) Fungi occurring on forests injured by air pollutants in the Upper Silesia and Cracow industrial regions. X. Mycoflora of dying young trees of *Alnus incana*. *European Journal of Forest Pathology*, 17(6): 337-348.
- Eglītis, M. (1938) *Augu slimības* [Plant diseases]. Rīga, p. 255. (In Latvian).
- Enerstvedt, L. I., Venn, K. (1979) Decay in mature *Picea abies* (L.) Karst. stands. A study on clear-cuttings in Øvre Eiker. *Meddr Norsk Institutt for Skogforskning*, 35: 237-264. [In Norwegian with English summary].
- Eriksson, L.N., Gustavsson, L. (2008) Biofuels from stumps and small roundwood – costs and CO₂ benefits. *Biomass and Bioenergy*, 32(10): 897-902.
- EUSTAFOR (2010) Biomass and Bioenergy Report. *WG Booklet*. Available at: http://www.eustafor.eu/failed/File/Publications/Biomass_Booklet.pdf.

- Fowler, J., Cohen, L., Jarvis, P. (2001) *Practical statistics for field biology*. 2nd ed. John Wiley and Sons, Chichester, UK.
- Gaitnieks, T., Arhipova, N., Donis, J., Stenlid, J., Vasaitis, R. (2008) Butt rot incidence and related losses in Latvian *Picea abies* (L.) Karst. stands. In: Garbelotto, M., Gontier, P. (eds.) *Proceedings of the 12th International Conference on Butt and Root Rots of Forest Trees*, Berkley, California – Medford, Oregon, 12-19 August, 2007. The University of California, Berkley, California. pp. 177-179.
- Gaitnieks, T., Šica, L., Indriksons, A., Grantiņa, L. (2004) Root-rot detection in Norway spruce stands using pulsed electronic tool. In : Mańka, M., Łakomy, P. (eds). *Proceedings of the 11th International Conference on Root and Butt Rots of Forest Trees*, Poznań and Białowieża, Poland, August 16-22, 2004. The August Cieszkowski Agricultural University, Poznań, Poland, pp. 379-385.
- Garbelotto, M., Slaughter, G., Popunuck, T., Cobb, F.W., Burns, T.D. (1997) Secondary spread of *Heterobasidion annosum* in white fir root-diseases centre. *Canadian Journal of Forest Research*, 27: 766-773.
- Gibbs, J.N. (1968) Resin and resistance of conifers to *Fomes annosus*. *Annals of Botany*, 32(3): 649-665.
- Greig, B.J.W., Low, J. D. (1975) An experiment to control *Fomes annosus* in second rotation pine crops. *Forestry*, 48 (2): 147-163.
- 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(2): 191-202.
- Greig, B.J.W. (1962) *Fomes annosus* (Fr.) Cke. and other root-rotting fungi in conifers on ex-hardwood sites. *Forestry*, 35(2): 164-182.
- Greig, B.J.W. (1976) Biological control of *Fomes annosus* by *Peniophora gigantea*. *European Journal of Forest Pathology*, 6(2): 65-71.
- Greig, B.J.W. (1980) Chemical, biological and silvicultural control of *Fomes annosus*. In: Dimitri, L. (ed.) *Proceedings of 5th IUFRO conference of problems of root and butt rot in conifers*, Germany, 1978. Hesswische Forstliche Versuchsanstalt, Hann. Munden, p. 75-84.
- Greig, B.J.W. (1984) Management of East England pine plantations affected by *Heterobasidion annosum* root rot. *European Journal of Forest Pathology*, 14(7), 392-397.
- Greig, B.J.W., Gibbs, J.N., Pratt, J.E. (2001) Experiments on susceptibility of conifers to *Heterobasidion annosum* in Great Britain. *Forest Pathology*, 31(4): 219-228.
- Hakkila, P. (2004) Developing technology for large-scale production of forest chips. In: *Wood Energy Technology Programme 1999-2003*, National Technology Agency, Helsinki, Finland. 98 p.
- Hall, T. A. (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series*, 41: 95-98.
- Hallaksela, A.-M. (1984) Causal agents of butt-rot in Norway spruce in Southern Finland. *Silva Fennica*, 18(3): 237-243.
- Hallaksela, A.M., Nevalainen, S. (1981) Control of root rot fungus (*Heterobasidion annosum*) by treating Norway spruce stumps with urea. *Folia Forestalia*, 470: 1-10. [In Finnish with English summary].

- Harper, G.J., Comeau, P.G., Hintz, W., Wall, R.E., Prasad, R., Becker, R.M. (1999) *Chondrostereum purpureum* as a biological control agent in forest vegetation management. II. Efficacy on Sitka alder and aspen in western Canada. *Canadian Journal of Forest Research*, 29(7): 852-858.
- Hartig, R. (1874) Important diseases of forest trees. Contributions to mycology and phytopathology for botanists and foresters. *Phytopathological classics*. No. 12. American Phytopathological Society. St. Paul, MN. 127 pp.
- Hartig, R. (1878) Die Zersetzungserscheinungen des Holzes der Nadelholzebaume und der Eiche in Forstlicher, Botanischer und Chemischer Richtung. Springer-Verlag, Berlin, 151 pp.
- Hinds, T.E. (1977) Heart rots of Engelmann spruce and subalpine fir in the Central Rocky Mountains. *USDA Forest Service Forest Insect & Disease Leaflet*, 150, 9 p.
- Hjältén, J., Stenbacka, F., Andersson, J. (2010) Saproxylic beetle assemblages on low stumps, high stumps and logs: Implications for environmental effects of stump harvesting. *Forest Ecology and Management*, 260(7): 1149-1155.
- Hodges, C.S. (1969) Modes of infection and spread of *Fomes annosus*. *Annual Review of Phytopathology*, 7: 247-266.
- Huse, B. J., Solheim, H., Venn, K. (1994) Råte i gran registrert på stubber etter hogst vinteren 1992 [Rot in spruce registered at stumps after harvesting at winter 1992]. *The Norwegian Forest and Landscape Institute, Rapport fra Skogforsk 23/94*: 26 p [In Norwegian with English summary].
- Huss-Danell, K., Lundmark, J.-E. (1988) Growth of nitrogen-fixing *Alnus incana* and *Lupinus* spp. for restoration of degenerated forest soil in northern Sweden. *Studia Forestalia Suecica*, 181: 1-20.
- Hyppe, A. (1978) Rotröta och stubbrytning. *Projekt heltrådsutnyttjande rapport nr. 60*. Swedish University of Agricultural Sciences. [In Swedish].
- Isomäki, A., Kallio, T. (1974) Consequences of injury caused by timber harvesting machines on the growth and decay of spruce. (*Picea abies*). *Acta Forestalia Fennica*, 136: 1-25.
- Johannesson, H., Stenlid, J. (2003) Molecular markers reveal genetic isolation and phylogeography of the S and F intersterility groups of the wood-decay fungus *Heterobasidion annosum*. *Molecular Phylogenetics and Evolution*, 29(1): 94-101.
- Johansson, M., Marklund, E. (1980) Antagonists of *Fomes annosus* in the rhizosphere of grey alder (*Alnus incana*) and Norway spruce (*Picea abies*) *European Journal of Forest Pathology*, 10(7): 385-395.
- Johansson, S.M., Pratt, J.E., Asiegbo, F.O. (2002) Treatment of Norway spruce and Scott pine stumps with urea against the root and butt fungus *Heterobasidion annosum* – possible modes of action. *Forest Ecology and Management*, 157(1-3): 87-100.
- Jones, A.C., Ostry, M. E. (1998) Estimating white trunk rot in aspen stands. *Northern Journal of Applied Forestry*, 15 (1): 33-36.
- Jonsell, M. (2007) Effects on biodiversity of forest fuel extraction, governing by processes working on a large scale. *Biomass and Bioenergy* 31(10), 726-732.
- Jūrēvičs, B. (1939) *Eglu sakņu un stubbru trupe*. [Spruce root and stem rot]. In: *Mežsaimniecības rakstu krājums*, 17. Rīga, p. 81-107. [In Latvian].

- Kallio, T. (1972) An example of the economic loss caused by decay in growing spruce timber in Southern Finland. *Silva Fennica*, 6: 116-124. [In Finnish with English summary].
- Kallio, T., Tamminen, P. (1974) Decay of spruce (*Picea abies* (L.) Karst.) in the Åland Islands. *Acta Forestalia Fennica*, 138: 1-42.
- Kärki, T. (1999) Prediction the value of grey alder (*Alnus incana*) logs based on external quality. *Silva Fennica*, 33(1): 13-23.
- Kärki, T., Maltamo, M., Eerikäinen, K. (2000) Diameter distribution, stem volume and stem quality models for grey alder (*Alnus incana*) in eastern Finland. *New Forests*, 20(1): 65-85.
- Kären, O., Högberg, N., Dahlberg, A., Jonsson, L., Nylund, J.E. 1997. Inter- and intraspecific variation in the ITS region of rDNA of ectomycorrhizal fungi in Fennoscandia as detected by endonuclease analysis. *New Phytologist*, 136(2): 313-325.
- Kenigsvalde, K., Donis, J., Korhonen, K., Gaitnieks, T. (2011) Biological control of Heterobasidion root rot of coniferous stumps by *Phlebiopsis gigantea* – literature review. *Mežzinatne*, 23(56):25-40. [In Latvian with English abstract].
- Korhonen, K. (1978) Intersterility groups of *Heterobasidion annosum*. *Communicationes Instituti Forestalis Fenniae*, 94(6), 1-25.
- Kraj, W., Kowalski, T. (2010) Identification of *Heterobasidion* spp. in Poland by RFLP analysis of laccase and manganese dependent peroxidase. *Dendrology*, 63: 11-19.
- Kõljalg, U., Larsson, K.-H., Abarenkov, K., Nilsson, N.H., Alexander, I. J., Eberhardt, U., Erland, S., Höiland, K., Kjoller, R., Larsson, E., Pennanen, T., Sen, R., Taylor, A.F.S., Tadersoo, L., Vrålstad, T., Ursing, B.M. (2005) UNITE: a database providing web-based methods for the molecular identification of ectomycorrhizal fungi. *New Phytologist*. 166(3): 1063-1068.
- Kuliesis, A. (1993) *Forest yield models and tables in Lithuania*. LMS, Kaunas, Lithuania, 383 pp.
- Kuznetsova, T., Lukjanova, A., Mandre, M., Lõhmus, K. (2010) Aboveground biomass and nutrient accumulation dynamics in young black alder, silver birch and Scots pine plantations on reclaimed oil shale mining areas in Estonia. *Forest Ecology and Management*, 262(2): 56-64.
- Laitila, J., Ranta, T., Asikainen, A. (2008) Productivity of stump harvesting for fuel. *International Journal of Forest Engineering*, 19(2): 37-47.
- Lange, V. (1960) [Silvicultural characteristics of Norway spruce with different branching]. In *Proceedings of the Latvia University of Agriculture*, 9, Rīga, p. 537-542. [In Latvian.]
- Latvian Forest Industry Federation. (2008) Forest sector in Latvia 2008. (Brochure) Publisher: LFIF, Rīga, p. 7. Available online at: http://www.zm.gov.lv/doc_upl/Meza_nozare_katalogs_EN.pdf [cited 13 Jun 2012].
- Lazdiņš, A., Von Hofsten, H., Lazdiņa, D., Lazdāns, V. (2009) Productivity and costs of stump harvesting for bioenergy production in Latvian conditions. In: Malinovska, L., Mezote, A., Kronbergs, E., Nulle, I. (eds.) *Proceedings of 8th International Scientific Conference Engineering for Rural Development*, Jelgava, May 28-29. Latvian University of Agriculture, Jelgava, Latvia, pp. 194-201.
- Lehtijärvi, A., Doğmuş- Lehtijärvi, T., Aday, A. G., Oskay, O. (2011) The efficacy of selected biological and chemical control agents against *Heterobasidion abietinum* on *Abies cilicica*. *Forest Pathology*, 41(6): 470-476.
- Liepa, I. (1974) *Biometrija*. [Biometrics]. Zvaigzne, Riga, Latvia, 332 pp. [In Latvian].

- Lindén, M., Vollbrecht, G. (2002) Sensitivity of *Picea abies* to butt rot in pure stands and in mixed stands with *Pinus sylvestris* in Southern Sweden. *Silva Fennica*, 36(4): 767-778.
- Lõhmus, K. Mander, U., Tullus, H., Keedus, K. (1996) Productivity, buffering capacity and resources of grey alder forests in Estonia. In: K. Perttu, A. Koppel (eds) *Short rotation willow coppice for renewable energy and improved environment*. Swedish University of Agricultural Sciences, Uppsala, Sweden. 95-105.
- Lygis, V., Vasiliauskas, R., Stenlid, J. (2004a) Planting *Betula pendula* on pine sites infested by *Heterobasidion annosum*: disease transfer, silvicultural evaluation, and community of wood-inhabiting fungi. *Canadian Journal of Forest Research*, 34(1): 120-130.
- Lygis, V., Vasiliauskas, R., Stenlid, J., Vasiliauskas, A. (2004b) Silvicultural and pathological evaluation of Scots pine afforestations mixed with deciduous trees to reduce the infections by *Heterobasidion annosum* s.s. *Forest Ecology and Management*, 201(2-3): 275-285.
- Lygis, V., Vasiliauskaite, I., Stenlid, J., Vasaitis, R. (2010) Impact of forest fire on occurrence of *Heterobasidion annosum* s.s. root rot and other wood-inhabiting fungi in roots of *Pinus mugo*. *Forestry*, 83(1): 83-92.
- Magurran, A. E. (1988) *Ecological diversity and its measurements*. Princeton University Press, Princeton, NJ, 179 pp.
- Mangalis, I. (1975) [*Heterobasidion annosum* in spruce plantations and spruce forests]. In: [Spruce and spruce forests in Latvia]. *Zinātne*, p. 35-42. (in Latvian).
- Mäkinen, H., Hallaksela, A.-M., Isomäki, A. (2007) Increment and decay in Norway spruce and Scots pine after artificial logging damage. *Canadian Journal of Forest Research*, 37(11): 2130 – 2141.
- Meredith, D.S. (1959) The infection of pine stumps by *Fomes annosus* and other fungi. *Annals of Botany*, 23(3): 455-476.
- Morrison, D.J., Johnson, A.L.S. (1978) Stump colonization and spread of *Fomes annosus* 5 years after thinning. *Canadian Journal of Forest Research*, 8(2): 177-180.
- Müller, M.M., Heinonen, J., Korhonen, K. (2007) Occurrence of *Heterobasidion* basidiocarps on cull pieces of Norway spruce left on cutting areas and in mature spruce stands. *Forest Pathology*, 37(6): 374-386.
- Nicolotti, G., Gonthier, P. (2005) Stump treatment against *Heterobasidion* and *Phlebiopsis gigantea* and some chemicals in *Picea abies* stands in the Western Alps. *Forest Pathology*, 35(5): 365-374.
- Nilsen, P. (1983) The occurrence of rot in old stands of Norway spruce (*Picea abies* (L.) Karst.), related to tree data and stand properties. *Rapport Norsk institut for skogforskning 2/83*: 1 – 34. [In Norwegian with English summary].
- Norokorpi, Y. (1979) Old Norway spruce stands, amount of decay and decay-causing microbes in Northern Finland. *Communicationes Instituti Forestalis Fenniae*, 97(6): 1-77.
- Oliva, J., Samils, N., Johansson, U., Bendz-Hellgren, M., Stenlid, J. (2008) Urea treatment reduced *Heterobasidion annosum* s.l. root rot in *Picea abies* after 15 years. *Forest Ecology and Management*, 255(7): 2876-2882.
- Otrosina, W.J., Garbelotto, M. (2010) *Heterobasidion occidentale* sp. nov. and *Heterobasidion irregulare* nom. nov. : A disposition of North American *Heterobasidion* biological species. *Fungal Biology*, 114(1): 16-25.

- Pavlov, I.N., Korhonen, K., Gubarev, P.V., Cherepnin, V.L., Barabanova, O.A., Mironov, A.G., Ageev, A.A. (2008) Закономерности образования очагов *Heterobasidion annosum* (Fr.) Bref. S. Str. в географических культурах сосны обыкновенной (Минусинская котловина). [Formation of *Heterobasidion annosum* (Fr.) Bref s.s. disease centers in the provenances of Scots pine (Minusinsk Basin).] *Хвойные бореальной зоны*, XXV, 1-2: 28-36.
- Pechmann, H. V., Aufsess, H. V. (1971) Untersuchungen über die Erreger von Stammfäulen in Fichtenbeständen. *Forstw. Cbl.*, 90: 259-284.
- Pechmann, H. V., Aufsess, H. V., Rehfuess, K. E. (1973) Ursachen und Ausmaß von Stammfäulen in Fichtenbeständen auf verschiedenen Standorten. *Forstw. Cbl.*, 92: 68-89.
- Pettersson, M., Rönnerberg, J., Vollbrecht, G., Gemmal, P. (2003) Effect of thinning and *Phlebiopsis gigantea* stump treatment on the growth of *Heterobasidion parviporum* inoculated in *Picea abies*. *Scandinavian Journal of Forest Research*, 18(4): 362-367.
- Piri, T. (1996) The spreading of the S type of *Heterobasidion annosum* from Norway spruce stumps to subsequent tree stands. *European Journal of Forest Pathology*, 26(4): 193-204.
- Piri, T. 2000. Response of compensatory-fertilized *Pinus sylvestris* to infection by *Heterobasidion annosum*. *Scandinavian Journal of Forest Research*, 15(2): 218-224.
- Piri, T. (2003) Silvicultural control of *Heterobasidion* root rot in Norway spruce forests in Southern Finland. Regeneration and vitality fertilization of infected stands. *Academic dissertation in Forest Pathology, Finnish Forest Research Institute*, Helsinki, Finland, 64 pp.
- Piri, T., Korhonen, K. (2001) Infection of advance regeneration of Norway spruce by *Heterobasidion parviporum*. *Canadian Journal of Forest Research*, 31(6): 937-942.
- Piri, T., Korhonen, K. (2008) The effect of winter thinning on the spread of *Heterobasidion parviporum* in Norway spruce stands. *Canadian Journal of Forest Research*, 38(10): 2589 – 2595.
- Piri, T., Korhonen, K., Sairainen, A. (1990) Occurrence of *Heterobasidion annosum* in pure and mixed stands in Southern Finland. *Scandinavian Journal of Forest Research*, 5(1-4): 113 – 125.
- Pitt, D.G., Dumas, M.T., Wall, R.E., Thompson, D.G., Lanteigne, L., Hintz, W. (1999) *Chondrostereum purpureum* as a biological control agent in forest vegetation management. I. Efficacy on speckled alder, red maple, and aspen in eastern Canada. *Canadian Journal of Forest Research*, 29(7): 841-851.
- Pratt, J.E., Greig, B.J.W. (1988) *Heterobasidion annosum*: development of butt rot following thinning in two young first rotation stands of Norway spruce. *Forestry*, 61(4): 339-347.
- Pratt, J.E., Niemi, M., Sierota, Z.H. (2000) Comparison of three products based on *Phlebiopsis gigantea* for the control of *Heterobasidion annosum* in Europe. *Biocontrol Science and Technology*, 10(4): 467-477.
- Puddu, A., Luisi, N., Capretti, P., Santini, A. (2003) Environmental factors related to damage by *Heterobasidion abietinum* in *Abies alba* forests in Southern Italy. *Forest Ecology and Management*, 180(1-3): 37-44.
- Risbeth, J. (1951a) Observations on the biology of *Fomes annosus*, with a particular reference to East Anglian pine plantations. II. Spore production, stump infection, and saprophytic activity in stumps. *Annals of Botany*, 15(1): 1-22.

- Risbeth, J. (1951b) Observations on the biology of *Fomes annosus*, with a particular reference to East Anglian pine plantations. III. Natural and experimental infection of pines, and some factors affecting severity of the disease. *Annals of Botany*, 15(1): 221-246.
- Risbeth, J. (1951c) Butt rot by *Fomes annosus* Fr. in East Anglian conifer plantations and its relation to tree killing. *Forestry*, 24(2): 114-120.
- Risbeth, J. (1952) Control of *Fomes annosus* Fr. *Forestry*, 25(1): 41-50.
- Risbeth, J. (1957) Some further observations on *Fomes annosus*. *Forestry*, 30: 69-89.
- Risbeth, J. (1961) Inoculation of pine stumps against infection by *Fomes annosus*. *Nature*, 191: 826-827.
- Risbeth, J. (1963) Stump protection against *Fomes annosus* III. Inoculation with *Peniophora gigantea*. *Annals of Applied Biology*, 52: 63-77.
- Roll-Hansen, F., Roll-Hansen, H. (1980) Microorganisms which invade *Picea abies* in seasonal stem wounds. 1. General aspects. *Hymenomyces*. *European Journal of Forest Pathology*, 10(7): 321-339.
- Roth, E., Sleeth, B. (1936) Decay in sprout oak stands. *USDA Bur. Plant Ind., Emergency Conservation Work*, 4 p.
- Rönnerberg, J. (2000) Logging operation damage to roots of clear felled *Picea abies* and subsequent spore infection by *Heterobasidion annosum*. *Silva Fennica*, 34(1): 29-36.
- Rudolphi, J. (2007) Bryophytes, lichens and dead wood in young managed boreal forests. Doctoral thesis, *Faculty of Natural Resources and Agricultural Sciences, Department of Ecology*, SLU, Uppsala, 32 p.
- Rytter, L. (1995) Effects of thinning on the obtainable biomass, stand density, and tree diameters on intensively grown grey alder plantations. *Forest Ecology and Management*, 73(1-3): 135-143.
- Rytter, L. (1996) The potential of grey alder plantation forestry. In: Perttu, K., Koppel, A. (eds) *Short rotation willow coppice for renewable energy and improved environment*. Swedish University of Agricultural Sciences, Uppsala, Sweden, pp. 89-94.
- Sanger, F., Nicklen, S., Coulson, A. R. (1977) DNA sequencing with chain-terminating inhibitors. *Proceedings of the National Academy of Sciences of the United States of America*, 74, 5463-5467.
- Schütt, P., Schuck, H.J., Lautenschlager, K., Prestle, W., Stimm, B. (1979) *Fomes annosus* sporocarps – their abundance on decayed logs left in the forest. *European Journal of Forest Pathology*, 9(1): 57-61.
- Sokal, R.R., Rohlf, F. J. (1995) *Biometry*. 3rd edition. WH Freeman, New York, NY, USA.
- Stambaugh, W. J. (1989) Annosus root rot disease in Europe and the Southwestern United States: occurrence, research, and historical perspective. *USDA Forest Service Gen. Tech. Report*, PSW – 116, 9 p.
- Stenlid, J. (1985) Population structure of *Heterobasidion annosum* as determined by somatic incompatibility, sexual incompatibility, and isoenzyme patterns. *Canadian Journal of Botany*, 63: 2268-2273.
- Stenlid, J. (1987) Controlling and predicting the spread of *Heterobasidion annosum* from infected stumps and trees of *Picea abies*. *Scandinavian Journal of Forest Research*, 2(1-4): 187-198.

- Stenlid, J., Wåsterlund, I. (1986) Estimating the frequency of stem rot in *Picea abies* using an increment borer. *Scandinavian Journal of Forest Research*, 1(1-4): 303-308.
- Stivriņa, B., Kenigšvalde, K., Korhonen, K., Gaitnieks, T. (2010) Importance of large dimension decaying logging residues of spruce in the spread of *Heterobasidion* root rot. *Mežzinātne*, 22(55): 88-102. [In Latvian with English summary].
- Strurrock, R., Zeglen, S., Turner, J. (2006) British Columbia's coastal forests: laminated root rot. *Forest health stand establishment decision aid*, BC JEM 7: 41-43.
- Swedjemark, G., Stenlid, J. (1993) Population dynamics of the root rot fungus *Heterobasidion annosum* following thinning of *Picea abies*. *Oikos*, 66: 247 – 254.
- Swedjemark, G., Stenlid, J. (1995) Susceptibility of conifers and broadleaf seedlings to Swedish S and P strains of *Heterobasidion annosum*. *Plant Pathology*, 44(1): 73-79.
- Swedjemark, G., Stenlid, J. (1997) Between-tree and between-isolate variation for growth of S-group *Heterobasidion annosum* in sapwood of *Picea abies* cuttings. *Canadian Journal of Forest Research*, 27(5): 711-715.
- Tamminen, P. (1985) Butt-rot in Norway spruce in Southern Finland. *Communications Instituti Forestalis Fenniae*, 127: 1-51.
- Thor, M., Stenlid, J. (2005) *Heterobasidion annosum* infection of *Picea abies* following manual and mechanized stump treatment. *Scandinavian Journal of Forest Research*, 20(2): 154-164.
- Uri, V., Löhmus, K., Tullus, H. (2003) Annual net nitrogen mineralization in a grey alder (*Alnus incana* (L.) Moench.) plantation on abandoned agricultural land. *Forest Ecology and Management*, 184(1-3): 167-176.
- Uri, V., Tullus, H., Löhmus, K. (2002) Biomass production and nutrient accumulation in short-rotation grey alder (*Alnus incana* (L.) Moench.) plantation on abandoned agricultural land. *Forest Ecology and Management*, 161(1-3): 169-179.
- Vasaitis, R., Stenlid, J., Thomsen, I.M., Barklund, P., Dahlberg, A. (2008) Stump removal to control root rot in forest stands. A literature study. *Silva Fennica*, 42(3): 457-483.
- Vasiliauskas, A. (1989) *Root fungus and the resistance of coniferous forests' ecosystems*. Mokslas publisher, Vilnius, 174 p. [In Russian with English summary].
- Vasiliauskas, R. (1998) Five basidiomycetes in living stems of *Picea abies*, associated with 7-25 year-old wounds. *Baltic Forestry*, 1: 29-35.
- Vasiliauskas, R. (1999) Spread of *Amylostereum areolatum* and *A. chailletii* decay in living stems of *Picea abies*. *Forestry*, 72(2): 95-102.
- Vasiliauskas, R. (2001) Damage to trees due to forestry operations and its pathological significance in temperate forests: a literature review. *Forestry*, 74(4): 319-336.
- Vasiliauskas, R. (1999) Spread of *Amylostereum areolatum* and *A. chailletii* decay in living stems of *Picea abies*. *Forestry*, 72(2): 95-102.
- Vasiliauskas, R., Juška, E., Vasiliauskas, A., Stenlid, J. (2002) Community of *Aphylophorales* and root rot in stumps of *Picea abies* on clear-felled forest sites in Lithuania. *Scandinavian Journal of Forest Research*, 17(5): 398-407.
- Vasiliauskas, R., Lygis, V., Thor, M., Stenlid, J. (2004) Impact of biological (Rotstop) and chemical (urea) treatments on fungal community structure in freshly cut *Picea abies* stumps. *Biological Control*, 31(3): 405-413.

- Vasiliauskas, R., Lygis, V., Larsson, K.-H., Stenlid, J. (2005) Airborne fungal colonization of coarse woody debris in North Temperate *Picea abies* forest: impact of season and local spatial scale. *Mycological Research*, 109(4): 487-496.
- Vasiliauskas, R., Stenlid, J., Johansson, M. (1996) Fungi in bark peelings wounds of *Picea abies* in Central Sweden. *European Journal of Forest Pathology*, 26(6): 285-296.
- Vasiliauskas, R., Stenlid, J. (1998) Fungi inhabiting stems of *Picea abies* in managed stand in Lithuania. *Forest Ecology and Management*, 109(1-3): 119-126.
- Vollbrecht, G., Stenlid, J. (1999) Transfer of the P-type of *Heterobasidion annosum* from old-growth stumps of *Picea abies* to *Picea abies* and *Larix x eurolepis*. *European Journal of Forest Pathology*, 29(2): 153-159.
- Walmsley, J.D., Godbold, D. L. (2010) Stump harvesting for bioenergy – a review of the environmental impacts. *Forestry*, 83(1): 17-38.
- Wang, L.Y., Pålsson, H., Rönnberg, J. (2012) The effect of *Phlebiopsis gigantea* and urea stump treatment against spore infection of *Heterobasidion* spp. on hybrid larch (*Larix x eurolepis*) in Southern Sweden. *Forest Pathology*, in press. DOI: 10.1111/j.1439-0329.2012.00777.
- Warrell, J.J., Fairweather, M.L. (2009) Decay and discoloration of aspen. *USDA Forest Service Forest Insect & Disease Leaflet*, 149, 7 p.
- Westlund, A., Nohrstedt, H.-Ö. (2000) Effects of stump-treatment substances for root rot control on ground vegetation and soil properties in a *Picea abies* forest in Sweden. *Scandinavian Journal of Forest Research*, 15(5): 550-560.
- Wolken, J.M., Lieffers, V.J., Landhäusser, S.M., Mulak, T. (2009) Spring frost and decay fungi are implicated in suppressing aspen re-growth following partial cleaning in juvenile stands. *Annals of Forest Science*, 66: 805.
- Woodward, S., Stenlid, J., Karjalainen, R., Hüttermann, A. (ed.) (1998) *Heterobasidion annosum: biology, ecology, impact and control*. CAB International, Wallingford, UK, 589 p.
- Yde-Andersen, A. (1962) Seasonal incidence of stump infection in Norway spruce by air-borne *Fomes annosus* spores. *Forest Science*, 8: 98-103.
- Zabowski, D., Chambreau, D., Rotramel, N., Thies, W.G. (2008) Long-term effects of stump removal to control root rot on forest soil bulk density, soil carbon and nitrogen content. *Forest ecology and Management*, 255(3-4): 720-727.

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