Establishment of Ectomycorrhiza-inoculated *Pinus sylvestris* Seedlings on Coastal Dunes following a Forest Fire

AUDRIUS MENKIS[1*], VAIDOTAS LYGIS[2], DAIVA BUROKIENĖ[2] AND RIMVYDAS VASAITIS[1]

[1] *Department of Forest Mycology and Plant Pathology, Uppsala BioCenter, Swedish University of Agricultural Sciences, P.O. Box 7026, SE-750 07 Uppsala, Sweden*

[2] *Laboratory of Phytopathogenic Microorganisms, Institute of Botany at the Nature Research Centre, Žaliųjų Ežerų str. 49, LT-08406 Vilnius, Lithuania*

[*] *Corresponding author: Department of Forest Mycology and Plant Pathology, Uppsala BioCenter, Swedish University of Agricultural Sciences, P.O. Box 7026, SE-750 07 Uppsala, Sweden. E-mail: Audrius.Menkis@slu.se. Phone: +4618672729; Fax: +4618673599*

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Abstract

Replanting of coastal dunes following forest fires often result in low seedling survival, as the site is characterized by loose sand, low water and nutrient availability, lack of ectomycorrhizal (ECM) inoculum and increased susceptibility to insects and diseases. The aim of the present work was to check whether certain treatments of root systems have any impact on *Pinus sylvestris* seedling establishment as compared with standard planting procedures. Treatments included: i) inoculation with vegetative mycelium of *Cenococcum geophilum* (CGV); ii) inoculation with vegetative mycelium of *Suillus luteus* (SLV); moreover, in each of those two treatments damp sphagnum peat / sand mixture was added to roots upon planting; iii) inoculation with basidiospores of *S. luteus* applied during preceding growing season in the nursery (SLB), and iv) standard planting, no treatment (control). A two hectare experimental plantation of *P. sylvestris* was established in the spring of 2007 on coastal dunes of the Curonian Spit in western Lithuania on a site of approx.100 year-old *Pinus mugo* plantation that one year previously was devastated by a forest fire and clear cut. The three treatments and control, comprising 2,500 seedlings each, were planted in rows as 16 replicates (5000 seedlings /ha). Results showed that after the first growing season seedling survival was poor in all treatments (5.1 – 30.0%), but seedlings in CGV, SLV and SLB showed respectively 591%, 533% and 196% better survival than no-treatment controls. Thus, inoculations with vegetative mycelia and addition of peat/sand mixture were by far the best, although the most labour-consuming. After two growing seasons, the proportion of ECM-colonised roots in treatments was 27.3 – 39.8%, while 22.5% in the controls, and observed ECM types resembled those from forest nurseries. In conclusion, investigated treatments had a considerable positive impact on seedling establishment under high-risk site conditions.

Key words: ectomycorrhiza, reforestation, forest fire, Scots pine, seedling pathogens, fungal communities

Introduction

Coastal dunes of the Curonian Spit in the Baltic Sea represent probably one of the most extreme environments for tree establishment and growth in Lithuania (Olšauskas 2009). Historically, afforestation of the dunes was carried out in order to prevent drift of the sand and to protect dunes from rapid erosion (Strakauskaitė 2004). This was achieved by planting seedlings of a non-native dwarf mountain pine (*Pinus mugo*), which is a tree species known to be well adapted to harsh environmental conditions (Oyen 1999). As currently those plantations are overstocked, very prone to fire and of low recreational and aesthetical value (mainly due to the numerous root disease centres), it is generally considered that *P. mugo* should be gradually replaced by native Scots pine (*Pinus sylvestris*).

Forest fires are common in the area and often of high intensity. Such fires cause a major disturbance in the ecosystem, resulting in loss of forest stand, ground vegetation, and complete or partial burnout of litter and organic soil layer (Lygis et al. 2010 and references therein). Moreover, the fires lead to a drastic reduction of ectomycorrhizal (ECM) fungi in the soil (Dahlberg 2002). Simultaneously, they favour certain fire-adapted plant pathogens, as e.g. post-fire ascomycete *Rhizina undulata*, a cause of root-rot in conifers (Lygis et al. 2005 and references therein).

Reforestation of post-fire sites at the Baltic Sea coast is of key importance to sustain coastal dunes and prevent their erosion, but the efforts often result in low survival rates or even complete loss of outplanted seedlings already during the first vegetation season (V. Kolokšanskis, Curonian Spit National Park, personal communication). A complex of factors, e.g. low water and nutrient availability, reduced ECM inoculum, and damage caused by pathogens and insects are thought to be the most probable causes for low rates of seedling establishment. In particular, the ECM fungi are known to support plants with mineral nutrients and water, and protect against unfavourable abiotic and biotic stress factors, especially on low fertility sites and under harsh environmental conditions (Smith and Read 1997). To overcome deficiency of ECM fungi and to improve establishment and survival of tree seedlings in the field, artificial ECM inoculation of seedling roots was suggested (Trappe 1977, Dunabeitia et al. 2004, Menkis et al. 2007). Soil amendment with organic matter was also shown to have a positive effect on tree growth on poor fertility habitats (Rincon et al. 2006). Consequently, one could expect that combination of both methods and use of selected ECM fungal species adapted to the environmental conditions of the planting sites could further benefit vigour of tree seedlings (Menkis et al. 2007). The aim of the present work was to compare the outcome of standard planting procedures used in the Curonian Spit National Park, with three pre-planting treatments of seedling root systems aimed to promote the establishment and survival of *P. sylvestris* seedlings on sandy dunes under post-fire conditions.

Materials and methods

Study site and experimental design

The study was conducted on a sandy coastal dune of Curonian Spit peninsula located at the east coast of the Baltic Sea, western Lithuania (N 55°39', E 21°07'). The site comprised a shallow slope facing the sea, about 900 m from the coastline. There, in May 2006 a severe forest fire devastated approx. 250 hectares of approx. 100 year-old plantation of *P. mugo*, which subsequently has been clear-cut. In April 2007, a two hectare experimental plantation of *P. sylvestris* was established using two-year-old bare-root cultivated *P. sylvestris* seedlings produced in a local forest nursery. The three treatments and control, comprising 2500 seedlings each, were planted in rows in 16 replicates (5,000 seedlings/ha). Treatments included: i) inoculation with vegetative mycelium of *Cenococcum geophilum* (CGV); ii) inoculation with vegetative mycelium of *Suillus luteus* (SLV); moreover, in each of those two treatments damp sphagnum peat/sand mixture (ratio 3:1) was added to roots upon planting; iii) inoculation with basidiospores of *S. luteus* applied during preceding growing season in the nursery (SLB), and iv) standard planting, no treatment (control). In case of CGV and SLV treatments, the previous study has shown that addition of both ECM fungus and damp sphagnum peat/sand mixture to the root systems has significantly better effect on seedling establishment and growth than addition of damp sphagnum peat/sand mixture alone (Menkis et al. 2007), and therefore the later treatment was not included in the present study. In this study, no soil preparation was carried out prior to planting. This is a normal practice in dune reforestation on Curonian Spit, aimed to prevent soil erosion. Quality and mycorrhizal status of the seedlings was assessed during previous study (Menkis and Vasaitis 2011).

Meteorological conditions of study area

The climate in the area is transitional between maritime and continental, characterised by frequent and intense change of weather conditions, mild winters and relatively warm summers. Mean annual air temperature is 7°C, the absolute minimum -26°C and maximum 31°C. Maximum soil temperatures recorded at the closest meteorological station (10 km away) during the period 2006-2008, reached 56°C at the soil surface and 40°C at the depth of 5 cm (Klaipėda Coastal Meteorological Station, personal communication). On our study site respective surface temperatures were likely to be higher, as it was post-fire site covered by dark ash, which furthermore increases heat absorption. Average annual rainfall in the area is 660 mm, main precipitation occurring during October – February. Snow cover normally reaches 15-20 cm. Average wind velocity is 5.5 m/s, but 30-40 days per year it can reach up to 15 m/s. Such wind imposes intense movement of sand, especially on sea-exposed slopes, as that of the present study.

Soil characteristics

In order to determine soil characteristics, 20-cm-deep soil cores were taken at ten random locations more or less evenly distributed within the experimental plantation, pooled together and analyzed as a bulk sample. The analyses were carried out at the Institute of Botany, Vilnius, Lithuania, using standard procedures (Mineev et al. 1989). The main soil properties were as follows: pH_{KCl} – 4.59; humus content (calculated by oxidation method using a solution of $K_2Cr_2O_7$ and HCl) – 0.99%; $N_{mineralizable}$ – 0.0%; P_2O_5 – 146.8 mg/kg and K_2O – 14.6 mg/kg. All this (e.g., complete absence of nitrogen) indicates very low soil fertility.

Inoculum preparation, inoculation and planting

ECM inoculation of seedling roots was carried out employing two principally different methods. First, using vegetative mycelium of *C. geophilum* and *S. luteus*, as in CGV (i) and SLV (ii) treatments, respectively. Second, using basidiospore suspension of *S. luteus*, as in SLB (iii) treatment. *C. geophilum* and *S. luteus* were chosen for the experiment because both fungi occur naturally and establish ECMs with *P. sylvestris*, both in forest nurseries with seedlings and in coastal forests with old trees, and each of them has a distinguishable morphotype (Menkis et al. 2005, Menkis and Vasaitis 2011). For example, *S. luteus* was reported to form ECM with *P. sylvestris* seedlings in forest nurseries and in young forest plantations (Menkis et al. 2005, Menkis et al. 2007), and therefore could be expected to establish and increase overall mycorrhization in root systems of the inoculated plants. *C. geophilum* is known to be one of most drought tolerant ECM species (Melax and Reid 1973, Pigott 1982, Trappe 1977).

For the inoculations with vegetative mycelium (CGV and SLV treatments), isolates of *C. geophilum* (isolate UP162) and *S. luteus* (isolate UP72) were obtained from the culture collection of the Department of Forest Mycology & Pathology, Swedish University of Agricultural Sciences, Uppsala. The inoculum was prepared using a paper-sandwich technique described by Menkis et al. (2007). Briefly, round sheets of sterile filter paper (13 cm in diameter) were soaked in liquid Melin-Norkrans medium (MMN) (Marx, 1969) and pre-inoculated with a pure culture (a single genotype) of each fungus. Those were incubated for 30-37 days at room temperature in the dark. One day prior to the planting, pre-inoculated filter papers were soaked in sterile water to remove remaining nutrients, and on the day of planting they were transported to the planting site. In the field, roots of each seedling were shortened to 10-15 cm length and wrapped into one pre-inoculated filter paper. Then, a layer of damp sphagnum peat /sand mixture (ratio 3:1) was distributed round filter paper, which was then wrapped into a double layer of a paper towel. A total of 2500 seedlings were treated in this way with *C. geophilum*, and another 2500 with *S. luteus*, and each seedling was planted immediately after the inoculation.

For inoculations with basidiospores of *S. luteus* (SLB treatment), sporocarps of the fungus have been collected in the local stands of *P. sylvestris* in July 2006 and basidiospores were sampled from the spore prints. The spores have been washed-off by sterile water; their concentration was determined using haemocytometer and spore suspension was then prepared adjusting the concentration to ca. 10^5- 10^6 spores/ml. Inoculation with the prepared *S. luteus* spore suspension was carried out by watering roots of one-year-old bare-root seedlings of *P. sylvestris* cultivated in the forest nursery. This was done twice during the year 2006: at the end of July and in the middle of August. In May 2007, a total of 2500 SLB seedlings were excavated from the nursery and transported to the site for outplanting simultaneously with CGV and SLV treatments.

Furthermore, 2500 standard two-year-old bare-root cultivated seedlings of *P. sylvestris* were planted on the site. The seedlings originated from the same forest nursery. No specific treatment of those was accomplished, and planting procedures fully resembled routine afforestation practices normally conducted on coastal dunes in the Curonian Spit.

Assessment of seedling survival and fungal communities

Seedling survival in each treatment was assessed after the first growing season (year 2007) by counting all living / dead plants. In order to assess possible fungal involvement in seedling dieback, simultaneously 200 root systems of dead seedlings were randomly and proportionally sampled from different treatments and the control. Fungal isolations from those were done as described by Menkis et al. (2006). Briefly, roots have been washed in tap water and from each root system one ca. 1-cm-long segment was cut from lateral roots at the zone of advanced decay, the surface sterilized for 1 min in 33% hydrogen peroxide, rinsed three times in sterile water, placed on MMN agar medium in 9 cm Petri dishes (single segment per dish) and incubated at room temperature (21°C) in the dark. Isolated cultures were examined under a light microscope equipped with a long working-distance objective of 25×magnification, and grouped into mycelial morphotypes.

Establishment and persistence of the inoculated ECM fungi and the composition of fungal communities in roots were studied after the second growing season (year 2008). From each of three treatments and control twenty healthy-looking seedlings were randomly collected, their root systems were excised from stems, individually packed into plastic bags, transported to the laboratory and kept at 4ºC, and then investigated using methods described by Menkis et al. (2005). In brief, each root system has been washed with a tap water and twenty single root tips were randomly collected from its different parts using forceps. Sampled root tips were assessed for ECM colonisation using dissection microscope. In the presence of ECM colonisation, they were morphotyped using macro- and microscopic features.

Identification of fungi

For species identification, an internal transcribed spacer of the fungal ribosomal DNA (ITS rDNA) was sequenced using primers ITS1F and ITS4 (White et al. 1990). This was done for representatives of those sets:

i) mycelial morphotypes isolated from the decayed roots, and ii) ECM roots. Extraction of DNA from fungal mycelia and directly from the roots, and ITS rDNA amplification by PCR followed established methods described by Rosling et al. (2003). Sequencing was performed by Macrogen Inc., Seoul, Korea, utilising ABI 3730 XL automated sequencers (Applied Biosystems, Foster City, CA, USA). Raw sequence data were analysed using SeqMan version 8.1.3 software from DNASTAR package (DNASTAR, Madison, WI) and BioEdit version 7.0.5.2 (Hall 1999). Databases at GenBank (Altschul et al. 1997) and UNITE (Koljalg et al. 2005) were used to determine the identity of the sequences.

Statistical analyses

Comparison of seedling survival and ECM colonisation in different treatments was done using chi-square (χ^2) tests (Mead and Curnow 1983). As each of the datasets was subjected to three comparisons, confidence limits for *p*-values of the χ^2 tests were reduced three times, as required by the Bonferroni correction (Sokal and Rohlf 1995). ECM community structure and possible treatment effects were analysed with incorporated environmental variables (plant survival and mycorrhization) using Canonical Correspondence Analysis (CCA) in CANOCO 4.5 (ter Braak and Smilauer 1998). Comparison of ECM community structures in different treatments was assessed by calculating qualitative (S_S) Sorensen similarity indices (Magurran 1988).

Results

Seedling survival

After the first growing season in the plantation, seedling survival was poor in all treatments (5.1 - 30.0%). However, seedlings in CGV, SLV and SLB treatments showed significantly better survival (30.0%, 27.1% and 10.0%, respectively) than seedlings in the control (5.1%) (χ^2 test, p<0.0001). Furthermore, seedlings inoculated with vegetative mycelia (CGV and SLV) showed significantly better survival than those inoculated with basidiospores (SLB; χ^2 test, p<0.0001). Among inoculations using vegetative mycelium, survival of seedlings in CGV was slightly higher than that in SLV, but the difference was not statistically significant (χ^2 test, p=0.06).

ECM colonisation and community structure

After the second growing season, the proportion of roots colonised by ECM fungi was rather low in all treatments: 39.8% in CGV, 28.8% in SLV, 27.3% in SLB, but it was the lowest in untreated control seedlings, comprising 22.5%. Chi-square test revealed that mycorrhization of CGV-treated seedlings was significantly higher than in SLV, SLB or control (p<0.004), while neither of those three did differ significantly from each other (p>0.12). Out of the total of 80 root systems investigated, ECM has not been detected in only two, both originating from mycorrhiza-untreated controls,

Table 1. Frequency of occurrence of fungi (shown as percent of colonised plants / roots) in mycorrhizal root tips of *Pinus sylvestris* seedlings in relation to pre-planting treatment

Fungal species	GenBank accession no.	Treatments [a]				All
		CGV	SLV	SLB	Control	
		(20/400) [b]	(20/400)	(20/400)	(20/400)	(80/1600)
Ascomycetes						
Cadophora finlandica	HQ406816	-/-	-/-	10.0/4.6	10.0/6.7	3.8/2.3
Cenococcum geophilum	HQ406817	95.0/55.3	70.0/42.6	70.0/40.4	60.0/34.4	66.2/44.8
Phialophora sp. aurim712	DQ069046	10.0/2.5	10.0/5.2	40.0/18.3	15.0/7.8	8.8/7.8
Tuber sp. NS77	DQ068998	5.0/3.8	-/-	-/-	-/-	1.2/1.3
Wilcoxina mikolae	HQ406818	15.0/8.2	-/-	-/-	-/-	3.8/2.7
Wilcoxina sp. aurim735	DQ069051	25.0/6.3	15.0/6.9	10.0/3.7	-/-	6.2/4.7
All Ascomycetes		100.0/76.1	85.0/54.8	85.0/66.9	70.0/48.9	90.0/63.6
Basidiomycetes						
Hebeloma cavipes	HQ406819	10.0/2.5	15.0/4.3	-/-	15.0/5.6	6.2/3.0
Suillus granulatus	HQ406820	5.0/0.6	-/-	-/-	10.0/4.4	1.2/1.1
Suillus luteus	HQ406821	15.0/5.7	45.0/14.8	25.0/7.3	15.0/4.4	18.8/8.0
Thelephora terrestris	HQ406822	50.0/12.6	40.0/26.2	40.0/17.4	75.0/36.7	41.3/21.6
Tomentella ellisii	HQ406823	5.0/2.5	-/-	25.0/8.3	-/-	5.0/2.7
All Basidiomycetes		60.0/23.9	80.0/45.2	75.0/33.1	75.0/51.1	72.5/36.4

[a] Treatments: CGV – inoculated with vegetative mycelium of *C. geophilum* and supplemented with damp sphagnum peat /sand mixture; SLV – inoculated with vegetative mycelium of *S. luteus* and supplemented with damp sphagnum peat /sand mixture; SLB – inoculated with basidiospores of *S. luteus* applied during preceding growing season in the forest nursery; control – no treatment, standard planting.
[b] Numbers of examined root systems / root tips.

comprising there 10% among a total of 20 plants investigated. In the rest among all investigated plants (78) at least one ECM root tip was detected in analysed sample of each root system.

A total of 11 taxa comprised an overall ECM community, which in CGV, SLV and SLB was dominated by ascomycetes, while in the controls by basidiomycetes (Table 1). The reason for this in CGV, SLV and SLB treatments was the predominant root colonisation by *C. geophilum*, while in controls by *Thelephora terrestris*. The number of ECM taxa detected in each treatment was similar (χ^2 test, p>0.16): ten were detected in CGV, six in SLV, seven in SLB and seven in controls (Table 1). The CCA analysis showed 23.0% variation in ECM community structure that can be attributed to CGV, SLV, SLB treatments and the control (Figure 1). Ordination showed that all treatments and control were in rather close proximity on axis 1, indicating similar ECM community structures. The latter was also indicated by estimates of qualitative Sorensen similarity indices (S_S) which were high and ranged 0.71–0.77 (Figure 1). In the ordination, vector arrows pointed into virtually similar direction showing strong association between seedling survival and mycorrhization. As a result, both seedling survival and mycorrhization were favoured in CGV and SLV (inoculations using vegetative mycelia), but not in SLB (inoculation with basidiospores) and the controls.

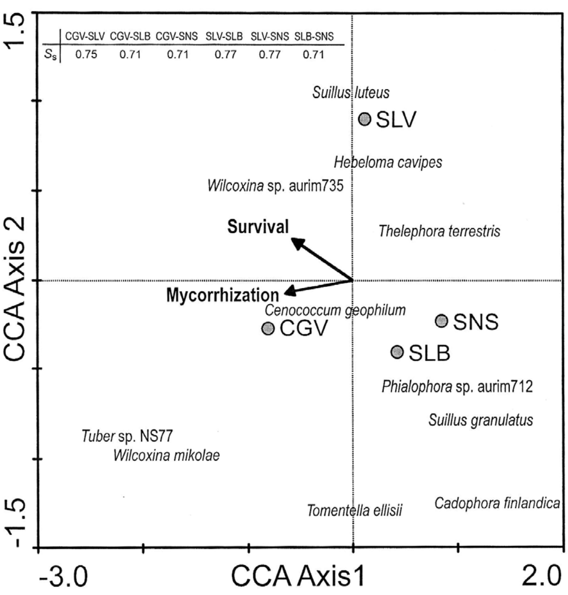

Figure 1. Ordination diagram based on Canonical Correspondence Analysis of ectomycorrhizal communities in roots of *P. sylvestris* seedlings inoculated with vegetative mycelia of *Cenococcum geophilum* (CGV) and *Suillus luteus* (SLV), basidiospores of *S. luteus* (SLB), and untreated control (SNS). The diagram shows part of the variation within the communities (23.0%) that can be attributed to the treatments along CCA axes 1 and 2. Arrows indicate the relative importance of environmental variables (plant survival and mycorrhization). Grey circles represent treatments. Taxonomic names correspond to a position in the ordination. Community structure is compared by calculating qualitative (S_S) Sorensen similarity indices (shown in the upper left corner of the diagram)

In the present study, only one of the inoculation methods / ECM species used had a profound effect on the establishment of ECM fungus in the root systems of inoculated plants. Consequently, the inoculation of seedling roots with vegetative mycelium of *C. geophilum* significantly increased subsequent mycorrhization as compared to any of other treatments (χ^2 test, p<0.0006) (Table 1). The inoculation with vegetative mycelium of *S. luteus* had less pronounced effect on mycorrhization, as the frequency of occurrence of this fungus in SLV was significantly higher only when compared to the controls (χ^2 test, p<0.02), but not when compared to CGV or SLB treatments (χ^2 test, p>0.2). Inoculation of seedling roots with basidiospores of *S. luteus* in the nursery was even less efficient, since the proportion of roots colonised by *S. luteus* in SLB did not differ significantly from any other treatment and control (χ^2 test, p>0.7) (Table 1).

Fungi in roots of dead seedlings

Fungal isolations from 200 segments of decayed roots yielded 261 pure cultures (1.3 cultures per root segment on average) representing nine distinct taxa. The most commonly isolated fungi were ascomycetes *Fimetariella rabenhorstii* (50.0%), *Phialocephala fortinii* (29.0%) and *Neonectria radicicola* (17.5%) (Table 2).

Table 2. Fungi isolated from decayed roots of *Pinus sylvestris* seedlings (n = 200)

Fungal species	GenBank accession no.	Proportion of colonised root systems, %
Ascomycetes		
Fimetariella rabenhorstii	HQ406808	50.0
Gibberella avenacea	HQ406809	0.5
Leptosphaeria sp. aurim1184	DQ0933683	0.5
Neonectria radicicola	HQ406810	17.5
Penicillium spinulosum	HQ406811	10.0
Phialocephala fortinii	HQ406812	29.0
Unidentified sp. 1276	HQ406813	12.0
Unidentified sp. 1277	HQ406814	10.5
All Ascomycetes		99.5
Zygomycetes		
Ambomucor seriatoinflatus	HQ406815	0.5

Discussion and conclusions

The survival of a planted seedling is largely determined during the first growing season. In case of success, the majority of survivors are likely to be established and usually persist at the site during the subsequent years (Menkis et al. 2007). In the present study, although the planting site was characterised by extremely unfavourable growth conditions, after the first growing season survival rate was worse than it could be expected. Factors contributing to this could be specific conditions on a site that occurred during that particular year. Following outplanting, the seedlings experienced exceptionally dry and warm period that lasted for nearly two months when their shoots were extensively grazed by *Strophosomus capitatus* (*Curculionidae*) beetles. Moreover, sporocarps of root pathogens *Rhizina undulata* and *Armillaria* spp. were commonly observed in the area, indicating a high activity of those fungi in the soil. Consequently, despite the low overall survival rates, the results of our study demonstrate that applied methods of artificial inoculation of roots with ECM, and in particular when this is combined with addition of damp peat-sand mixture, can significantly enhance seedling establishment even under extremely harsh environmental conditions and pressure of pests.

In related studies, although significantly positive effects of ECM inoculations on tree survival were reported, those were not always as strongly pronounced, and in particular when seedlings were planted on relatively nutrient-rich sites like former agricultural land (Menkis et al. 2007, Parlade et al. 2004). Furthermore, inoculation with ECM fungi did not improve seedling growth on forest clear-cuts characterised by relatively high availability of natural ECM inoculum (Loopstra 1988). Those results, together with the results of the present study, suggest that artificial ECM inoculation of seedling roots is most effective on dry and nutrient-deficient sites with low ECM availability.

ECM community detected in the current work (Table 1) to a large extent resembled ECMs from the nurseries, including the same nursery where from seedlings of this study had originated (Menkis et al. 2005, Menkis and Vasaitis 2011). This indicates that only limited changes did occur in communities of root fungi after seedling transfer and establishment on new sites such as coastal sandy dunes. Moreover, in a meanwhile natural colonization by both *C. geophilum* and *S. luteus* was typically observed on seedlings to which neither of the fungus has been inoculated (Table 1). The majority of fungi isolated from decayed roots of dead seedlings roots (Table 2) were also reported from both healthy-looking and diseased roots of conifer seedlings sampled in forest nurseries, clear-cuts and abandoned farmland (Menkis et al. 2005, Menkis et al. 2006, Menkis and Vasaitis 2011). The most common isolates in this study, *F. rabenhorstii* and *P. fortinii*, are characterised as root endophytes (Menkis et al. 2004, Sánchez Márquez et al. 2008) suggesting that behaviour of those fungi in roots might change depending on environmental conditions and/or plant health status (Menkis et al. 2004).

Furthermore, the inoculation of seedling roots with vegetative mycelium of *S. luteus* (SLV treatment) was far more effective when compared with the inoculation of the fungus using basidiospores in the nursery (SLB), which might be due to the fact that the method creates more suitable conditions for establishment of both ECM fungus and the host. Although in this study the most successful inoculations were while using paper towel pre-colonized with vegetative mycelia, this method would hardly receive large-scale application in practical forestry, mainly because it is highly labour-consuming. However, the method could be considered when aiming to rapid restoration of forest cover on high risk and fragile sites, as post-fire coastal ecosystems of high environmental, landscape, socio-economic and recreational value.

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СОЗДАНИЕ ПЛАНТАЦИЙ САЖЕНЦАМИ *PINUS SYLVESTRIS*, ИНОКУЛИРОВАННЫМИ ЭКТОМИКОРИЗОЙ НА ГАРЯХ В ПРИБРЕЖНЫХ ДЮНАХ

А. Менкис, В. Лигис, Д. Бурокене и Р. Васайтис

Резюме

Лесовосстановление в прибрежных дюнах после воздействия лесных пожаров часто характеризуется неудовлетворительной приживаемостью саженцев из-за влияния движущихся песков (засекание, засыпание саженцев), постоянным недостатком воды и питательных веществ, отсутствием эктомикоризы (ECM) в почве и, как следствие, повышенным риском повреждений вредителями и болезнями. Главная цель исследований было изучить влияние искусственной микоризации корневых систем саженцев *Pinus sylvestris* на их приживаемость по сравнению со стандартной процедурой посадки. Были подобраны следующие методы искусственной микоризации: i) инокуляция вегетативным мицелием гриба *Cenococcum geophilum* (CGV); ii) инокуляция вегетативным мицелием гриба *Suillus luteus* (SLV); кроме того, в каждом из этих вариантов непосредственно перед посадкой к корням саженцев была добавлена смесь сфагнового торфа и песка; iii) инокуляция почвы питомника базидиоспорами гриба *S. luteus* в предыдущем вегетационном сезоне (SLB), и iv) стандартная посадка (контроль). Экспериментальные культуры *P. sylvestris* площадью 2 га были высажены весной 2007-ого года в прибрежных дюнах Куршской косы на западе Литвы. Прежде на этом участке произрастали насаждения столетней *Pinus mugo*, которые пострадали от сильного пожара 2005 года и были вырублены в сплошную. Три варианта и контроль (каждый состоящий из 2500 саженцев) были посажены рядами по 16 повторностей (5000 саженцев /га). Результаты эксперимента показали, что после первого вегетационного сезона приживаемость саженцев была слабая во всех вариантах (5.1 – 30.0%), однако, по сравнению с контролем, саженцы CGV, SLV и SLB показали более высокую приживаемость на 591%, 533% и 196% соответственно. Таким образом, инокуляция вегетативным мицелием и добавка смеси торфа с песком дали лучшие показатели приживаемости саженцев, хотя этот метод оказался самым трудоемким. После второго вегетационного сезона, доля микоризных корней во всех вариантах составляла 27.3 – 39.8% (в контроле – 22.5%), и найденные типы ECM были сходны с типами, обнаруживаемыми в лесных питомниках. В заключении, искусственная микоризация имела значительный положительный эффект на приживаемость сосновых саженцев в сложных местных условиях.

Ключевые слова: эктомикориза, лесовосстановление, лесные пожары, патогены саженцев, сосна обыкновенная, грибные сообщества